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**From the Research Center Borstel - Leibniz Lung Center
Priority Research Area Infections
Department Molecular and Experimental Mycobacteriology
Of the University of Lübeck
Director: Prof. Dr. Ulrich Schaible**

***MYCOBACTERIUM TUBERCULOSIS* RESISTANCE EVOLUTION:
FROM UNDERSTANDING TO PREVENTION**

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Teresa Marlene Walz
from Offenburg, Germany

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First referee: Prof. Dr. Stefan Niemann

Second referee: Prof. Dr. med. Peter König

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„It takes all the running, to stay at the same place! “

Lewis Carroll

from Alice`s Adventures in Wonderland & Through the Looking-Glass

Abstract

With nearly half a million new multidrug resistant (MDR) tuberculosis (TB) cases estimated annually, TB is a major contributor to the current antimicrobial resistance crisis. MDR-TB is defined as an infection with a *Mycobacterium tuberculosis* complex (MTBC) strain resistant to the two most effective first-line drugs rifampicin (RIF) and isoniazid. To effectively address this public health crisis, it is essential to understand resistance mechanisms and evolution. This will better guide the development of new antibiotics and treatment strategies, and avoid rapid resistance development, as seen with bedaquiline (BDQ). This work explores resistance mechanisms to one key MDR-TB treatment drug, namely BDQ, and new evolutionary medicine informed treatment strategies based on negative hysteresis.

BDQ, one of the most important drugs for treating MDR-TB, is facing increasing resistance in clinical MTBC strains in several high incidence settings. Clinical MTBC strains acquire BDQ resistance through mutations in the *atpE* gene, which encodes the ATP synthase (primary target of BDQ), and in the *Rv0678* gene, which regulates the *mmpS5-mmpL5* efflux pump system. Most clinical resistance is associated with mutations in *Rv0678*, leading to increased expression of the efflux pump genes. In this work, RNA sequencing (RNAseq) analysis of BDQ-resistant mutants, harbouring different mutations in *Rv0678* (total loss-of-function mutations and reduced-function mutations), was conducted. The results revealed that efflux pump expression varied among clones, with baseline *mmpS5-mmpL5* efflux pump gene expression upregulated without BDQ exposure. Surprisingly, under BDQ challenge, the wild-type H37Rv strain upregulated the *mmpS5-mmpL5* efflux pump to similar levels as the *Rv0678* mutant clones, pointing to other mechanisms involved in resistance development. Analysis of the transcriptional response of *Rv0678* mutant clones and H37Rv at different BDQ exposure timepoints revealed a complex response indicating that resistance is mediated not by a single mechanism, but rather by an enhanced metabolic plasticity of the *Rv0678* mutants, which allows them to shift to a dormant-like state under stress while maintaining some cellular functions.

The second focus of this work was to explore evolution-informed treatment strategies aimed at enhancing drug efficacy and circumvent resistance evolution by using negative hysteresis as potential mechanism in sequential treatments to limit the evolutionary potential of MTBC strains. An *in vitro* model was designed based on short pre-exposure with one drug, followed by main-exposure with the second drug utilizing sub-lethal concentrations which potentially induces cellular hysteresis, a phenomenon where antibiotic effectiveness is enhanced based on the order of drug administration. Sequential exposure of MTBC reference strain H37Rv with ethambutol (EMB) followed by RIF led to a sequence-dependent reduction in bacterial load, attributed to the negative hysteresis effect, where

EMB pre-exposure enhanced RIF's efficacy. RNAseq was employed to investigate the underlying mechanisms, indicating that the cellular hysteresis effect, induced by the respective drug sequence, was likely driven by a cell surface stress response and metabolic remodelling by a *dank*-mediated stress response.

Taken together, the results obtained in this thesis contradicts common knowledge that BDQ resistance induced by *Rv0678* mutations is solely conferred by upregulation of the mmpS5-mmpL5 efflux pump system only, but rather points to the role of an enhanced metabolic plasticity in antibiotic responses as important survival strategy. It also shows, that comprehensive studies based on mutant generation combined with OMICS tools are needed to understand pathogen drug response, and escape mechanisms. Further, to the best of our knowledge, for the first time the potential of negative hysteresis induced by fast drug switching for enhancing drug efficacy even at sub-minimum inhibitory concentration (MIC) conditions was observed in *M. tuberculosis*. This has great potential for optimizing TB treatment concepts especially for drug resistant MTBC strains, where drug options are limited and cycling therapies with a lower number of antibiotics significantly enhance treatment possibilities. Accordingly, evolutionary informed treatment strategies should be urgently further exploited.

Zusammenfassung

Mit schätzungsweise fast einer halben Million neuer multiresistenter (*multidrug-resistant*, MDR) Tuberkulosefälle pro Jahr trägt die Tuberkulose (TB) erheblich zur aktuellen Antibiotikaresistenzkrise bei. MDR-TB ist definiert als eine Infektion mit einem *Mycobacterium tuberculosis* Komplex (MTBK)-Stamm, der gegen die beiden wirksamsten Erstlinienmedikamente, Rifampicin (RIF) und Isoniazid, resistent ist. Um der globalen Bedrohung durch Antibiotikaresistenzen wirksam begegnen zu können, ist es unerlässlich, sowohl die Entstehung als auch die zugrunde liegenden biologischen Mechanismen der Resistenzen zu verstehen. Dies kann dazu beitragen, die Entwicklung neuer Antibiotika und Behandlungsstrategien besser zu steuern und eine rasche Resistenzentwicklung, wie sie beispielsweise bei Bedaquilin (BDQ) beobachtet wird, zu verhindern.

In dieser Arbeit werden die Resistenzmechanismen gegen das wichtige MDR-TB-Behandlungsmedikament BDQ und neue, auf negativer Hysteresis basierende Behandlungsstrategien der Evolutionsmedizin untersucht. BDQ, eines der wichtigsten Medikamente zur Behandlung von MDR-TB, zeigt in mehreren Gebieten mit hoher TB-Inzidenz zunehmende Resistenz in klinischen MTBK-Stämmen. Diese Stämme erwerben BDQ-Resistenz durch Mutationen im *atpE*-Gen, das für die ATP-Synthase (das primäre Ziel von BDQ) kodiert, sowie im *Rv0678*-Gen, das das *mmpS5-mmpL5*-Exportpumpen System reguliert. Die meisten Resistenzen von klinischen MTBK-Stämmen basieren auf Mutationen in *Rv0678*, was zu einer erhöhten Expression von Exportpumpen Genen führt.

In dieser Studie wurde eine RNA-Sequenzierungsanalyse (RNAseq) von BDQ-resistenten Mutanten mit unterschiedlichen Mutationen in *Rv0678* durchgeführt, darunter solche mit vollständigem Funktionsverlust und Mutationen mit reduzierter Funktion. Die Ergebnisse zeigten, dass in der Tat die Exportpumpen Gene *mmpS5-mmpL5* ohne BDQ-Exposition in *Rv0678* Mutanten hochreguliert waren, allerdings variierte das Expressionsniveau zwischen den *Rv0678* Mutanten. Überraschenderweise wurde *mmpS5-mmpL5* beim Wildtyp-Stamm H37Rv unter BDQ-Behandlung auf ein vergleichbares Niveau hochreguliert wie bei den *Rv0678*-Mutanten, was auf die Existenz weiterer resistenzvermittelnder Mechanismen in den Mutanten hindeutet. Die Analyse der globalen Genexpression von *Rv0678*-Mutantenklonen und dem sensiblen Referenzstamm H37Rv zu verschiedenen BDQ-Behandlungszeitpunkten ergab eine komplexe Transkriptionsantwort, die darauf hindeutet, dass die Resistenz nicht auf einen einzigen Mechanismus zurückzuführen ist. Stattdessen scheint eine erhöhte metabolische Plastizität es den *Rv0678*-Mutanten zu ermöglichen, unter Stressbedingungen in einen „Dormancy-ähnlichen“ Zustand zu übergehen und dennoch einige zelluläre Funktionen aufrechtzuerhalten.

Der zweite Schwerpunkt dieser Studie bestand darin, evolutionsbasierte Behandlungsstrategien zu untersuchen, die darauf abzielen, die Wirksamkeit von Medikamenten zu erhöhen und die Entwicklung von Resistenzen zu verhindern. Hierzu soll negative Hysteresis als potenzieller Mechanismus in zyklischen Therapien genutzt werden, um das evolutionäre Potenzial von MTBK-Stämmen zu begrenzen. Um die Anwendung von negativer Hysteresis bei MTBK-Stämmen zu testen, wurde ein *in vitro*-Modell entwickelt, in dem die Bakterien zunächst kurz mit einem Medikament vor exponiert wurden, um dann nachfolgend die Hauptexposition mit einem zweiten Medikament durchzuführen. Beide Medikamente werden dabei in subletalen Konzentrationen eingesetzt. Hierdurch sollte die negative zelluläre Hysteresis induziert werden, ein Phänomen, bei dem die Wirksamkeit eines Antibiotikums in Abhängigkeit von der Reihenfolge der Medikamentenverabreichung erhöht wird.

In diesem Modell führte die sequentielle Behandlung des MTB-Referenzstamms H37Rv mit Ethambutol (EMB), gefolgt von RIF, zu einer sequenzabhängigen Reduzierung der Bakterienlast. Dieses Ergebnis wurde auf den Effekt der negativen zellulären Hysteresis zurückgeführt, bei dem die Vorbehandlung mit EMB die Wirksamkeit von RIF steigerte. Um die zugrunde liegenden Mechanismen zu untersuchen, wurde RNAseq genutzt, dies zeigte, dass der durch diese spezifische Medikamentenabfolge induzierte negative zelluläre Hysteresis-Effekt wahrscheinlich mit einer *dnaK*-assoziierten Stressantwort zusammenhängt, bei der die Zelloberfläche umstrukturiert wurde.

Insgesamt widersprechen die in dieser Arbeit erzielten Ergebnisse der gängigen Annahme, dass die durch *Rv0678*-Mutationen hervorgerufene BDQ-Resistenz ausschließlich durch eine Hochregulierung der *mmpS5*-*mmpL5* Exportpumpen vermittelt wird. Vielmehr weisen sie auf die entscheidende Rolle einer erhöhten metabolischen Plastizität bei der Antibiotikareaktion als wichtige Überlebensstrategie hin. Dies zeigt auch, dass umfassende Studien, die auf der Erzeugung von Mutanten in Kombination mit OMICS-Tools basieren, notwendig sind, um die Reaktion des Pathogens auf Medikamente und die Mechanismen der Resistenzentwicklung besser zu verstehen

Darüber hinaus wurde nach unserem Kenntnisstand zum ersten Mal das Potenzial der negativen zellulären Hysteresis aufgezeigt, welche durch einen raschen Wechsel der Wirkstoffe hervorgerufen wird, um die Wirksamkeit des Medikaments auch in Konzentrationen unterhalb der minimalen Hemmkonzentration (MHK) zu steigern. Dies birgt erhebliches Potenzial für die Optimierung von Tuberkulose-Behandlungskonzepten, insbesondere bei arzneimittelresistenten MTBK-Stämmen, bei denen die Behandlungsoptionen begrenzt sind. Zyklische Therapien mit einer reduzierten Anzahl von Antibiotika könnten die Behandlungsmöglichkeiten bei MDR-TB signifikant verbessern. Es ist daher dringend geboten, evolutionsbasierte Behandlungsstrategien weiterzuentwickeln und zu nutzen.

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1 Introduction

1.1 *Mycobacterium tuberculosis* complex

Mycobacterium tuberculosis complex bacteria (MTBC) are fascinating pathogens, being intertwined with humanity throughout history, leaving its mark on societies, art, and literature (1–4). MTBC strains includes a range of species, such as *Mycobacterium tuberculosis* (*M. tuberculosis*), *Mycobacterium africanum*, *Mycobacterium bovis* (*M. bovis*) and others, that can cause tuberculosis (TB) (5,6). TB also known as the white plague, predominantly infects the lungs but has the potential to disseminate throughout the body (7). Despite the passage of time and advances in medical science, TB remains one of the most fatal disease globally as stated by the World Health Organization (WHO) in 2024 (1,3,8,9). Bacteria of the MTBC have highly conserved genomes but have distinct host specifications (6,10).

TB, an airborne disease, presents a unique challenge for diagnosis and treatment (8). Initially, an estimated 5-10 % of infected individuals progress to active disease, the majority enters a latent phase, during which they are non-contagious (8). However, the bacterium can reactivate years after infection under conditions such as immunosuppression or malnutrition, leading to disease onset (11). Reactivation of latent TB cases is often indicated by unspecific symptoms such as cough, chest pain, weakness, and weight loss (11). The complex interplay between MTBC bacteria and our immune system remains a subject of ongoing investigation (12–14). Therefore, a deeper understanding of the multifaceted immune response to MTBC strains infections is ever evolving. The bacterium is known to attack the host's innate immune defences, evading elimination and invading host cells to survive (14). Inside the macrophage, it survives within the phagosome, preventing its maturation and so evading lysosomal destruction (15). Immune cells form granulomas around the bacteria containing the infection (16). This pathological hallmark of TB unfortunately, complicate treatment by providing a hide-out for the bacteria, resulting in poor antibiotic penetration and treatment escape (17).

However, despite being a curable and preventable disease, the global burden of TB remains around 10 million new cases every year, and a distressing 1.3 million fatalities (in 2022), intensified by SARS-CoV-2 pandemic (8). The WHO's ambitious campaign to end TB by 2030 has encountered obstacles with a complex interplay of socioeconomic, geopolitical, healthcare-related factors and the drug resistance crisis (8,18–20). The global TB report of 2023 states three big 2025 milestones. The most challenging target appears to be the general reduction of the incidence rate by 50 %, with only an 8.7 % reduction achieved between 2015 and 2022. However, progress towards the other two milestones is more encouraging. The goal of reducing TB-related deaths by 75 % has seen a 19 % reduction over the same period. The third milestone, which aims to reduce the cost burden for people with TB to zero by

2025, has already seen nearly 50 % of this target achieved by 2023 (8). Despite these advancements, it seems unlikely that the 2025 targets will be fully met within this year.

As highlighted by the 2025 milestones, a critical challenge in the fight against TB is among other factors like drug-resistant TB, is the persistent gap between rich and poor nations (8,19,21). In 2022, the WHO identified thirty countries with high TB incidence, accounting for nearly 90 % of global cases. Notably, eight of these countries contribute to two-thirds of the world's total TB cases. According to the 2023 Global TB Report, these countries are: India (27 %), Indonesia (10 %), China (7.1 %), the Philippines (7.0 %), Pakistan (5.7 %), Nigeria (4.5 %), Bangladesh (3.6 %) and the Democratic Republic of the Congo (3.0 %) (8). The strong correlation between low gross domestic product (GDP) and high TB burden emphasizes the urgent need for global action to address these inequalities (Figure 1.1) (8). Poverty, low GDP, and malnutrition among those infected significantly contribute to the worsening of TB disease outcomes (8). This association becomes apparent as nations with lower GDPs tend to have higher TB incidence rates per 100 000 population (Figure 1.1); and nations with a low percentage of undernourished population have low TB incidences (8).

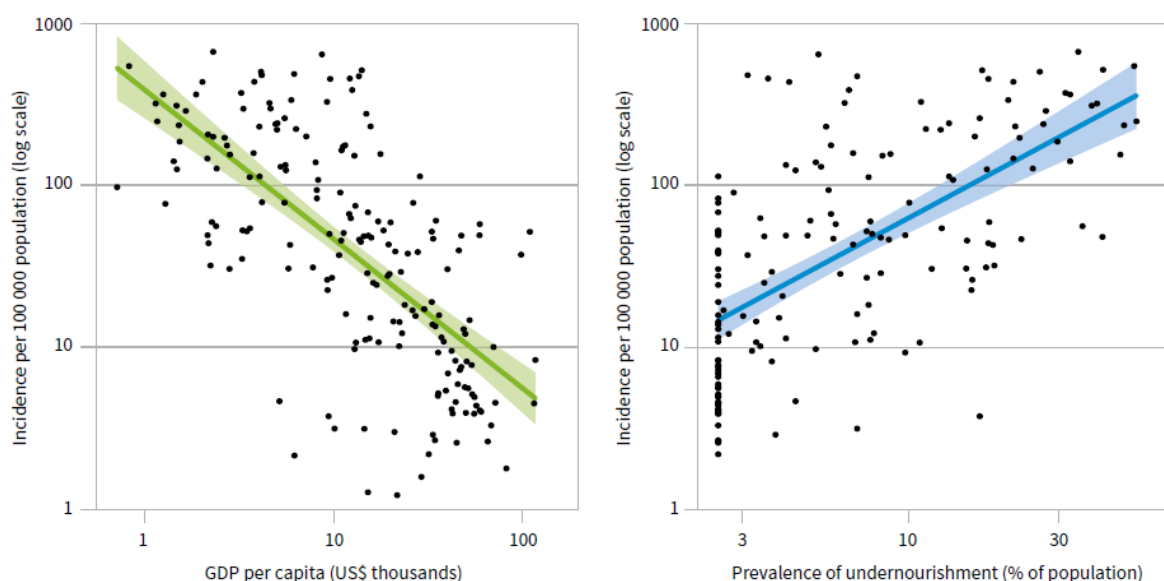


Figure 1.1: *M. tuberculosis* burden and fatalities in relation to gross domestic product and malnourishment. (Left) Correlation between the gross domestic product (GDP) per capita, and TB incidences per 100 000 population. (Right) Prevalence of undernourishment as percent of population. Countries and regions are represented as dots. Data and figure sourced from WHO's Global TB Report 2023. A study by Carter et al., further underscores the impact of socioeconomic factors, suggesting that eradicating poverty could reduce global TB incidence by about 30 %, while improved social protection coverage could lead to an almost 80 % reduction of TB incidences by 2023. Enhancing these factors could result in an annual incidence decrease of about 9 % (21). Additionally, the spread of multidrug-resistant (MDR) TB poses a significant challenge to global TB control efforts (22–24).

1.2 Drug resistant and multidrug resistant tuberculosis – an ongoing public health crisis

The rapid emergence of resistance to both novel and established antibiotics is a significant challenge for TB control globally (8). With an estimate of half a million rifampicin-resistant (RR) and MDR-TB cases annually, it is fuelling the ongoing global antimicrobial resistance crisis (8,22). MDR-TB is defined as resistance to the two essential first-line drugs rifampicin (RIF) and isoniazid (INH) (8,22). If patients with an MDR / RR-TB infection acquire resistance to any fluoroquinolone, they are classified as having pre-extensively drug resistant TB (pre-XDR-TB) (22). XDR-TB, is defined by additional resistance to at least one of the other Group A drugs bedaquiline (BDQ) and linezolid (LIN) (8). Approximately 3 % of new TB cases are multidrug-resistant (MDR-TB), with significant regional variability (8). In some areas, up to 40 % of newly detected cases are MDR-TB (8,24). Notably, the spread of drug-resistant *Mycobacterium tuberculosis* complex (MTBC) strains is primarily driven by transmission dynamics rather than de novo resistance mutations in patients (24,25). A study on MDR-TB transmission in Georgia found that the epidemic is primarily driven by MDR Central Asia L2 strains, which are particularly fit and transmissible due to both specific and compensatory mutations in RNA polymerase, rather than L4 strains (24).

Furthermore, the relatively low detection rate of drug-resistant TB leads to suboptimal treatment outcomes and significantly hinders the progress of initiatives such as the WHO's End-TB project (26,27). Moreover, countries in the Far and Middle East are facing alarming transmission rates, with over 25 % of new TB patients diagnosed with MDR-TB (Figure 1.2A) (8). In response to this crisis, the United Nations' high-level meeting on TB set a milestone to treat 1.5 million MDR / RR-TB patients between 2018 and 2022, but only 55 % of this target was met (Figure 1.2B) (8). A major obstacle is detection of MDR-TB cases, as it requires bacteriological confirmation and testing for drug resistance by molecular diagnostic tests, culture methods, and sequencing technologies (8,22). Furthermore, even if standard rapid molecular drug susceptibility tests or Xpert MTB / RIF assays are available, not all resistance conferring mutations are necessarily detected, further adding to a suboptimal treatment outcome (22,28).

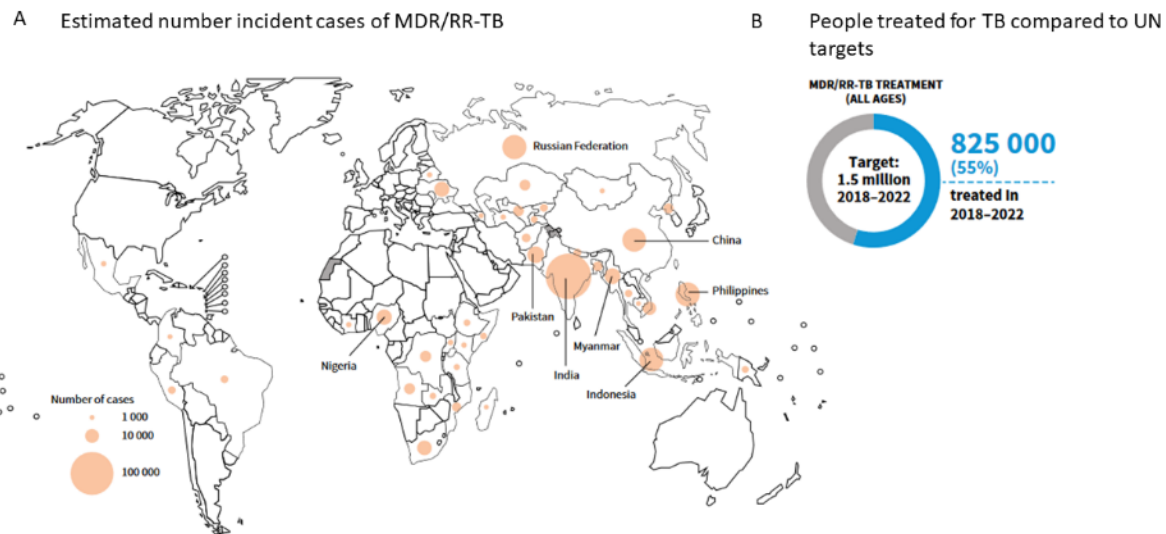


Figure 1.2: Global incident cases of multi-drug resistant and rifampicin resistant tuberculosis (MDR / RR-TB) vs. people treated against MDR / RR-TB. (A) According to the WHO Global TB Report the highest incidences (> 10 000) of MDR / RR-TB are in the Russian Federation, India, Pakistan, China, the Philippines, Myanmar, Indonesia and also parts of Sub-Saharan Africa like Nigeria. (B) Treatment of MDR / RR-TB cases reached 825 000 people between 2018 - 2022, which is only about 50 % of the treatment goal of WHO End-TB programme. Data and figure sourced from WHO's Global TB Report 2023.

1.3 Tuberculosis treatment regimens

The WHO advocates a 6-month regimen (2HRZE / 4HR) for infections caused by susceptible MTBC strains, with variations depending on the case. The standard regimen consists of a 2-month phase including INH, RIF, pyrazinamide (PZA), and ethambutol (EMB), followed by 4-months of INH and RIF (29,30). RIF is a cornerstone in this treatment, which inhibits the bacterial Deoxyribonucleic acid (DNA) polymerase and thereby prevents DNA transcription (31,32) (Figure 1.3). INH, a prodrug activated by the catalase-peroxidase *katG*, disrupts mycolic acid biosynthesis via radical and adduct generation (33,34). EMB primarily targets dividing cells by inhibiting the formation of arabinogalactan a specific cell wall component (35). PZA targets non-replicating cells, its active metabolite pyrazinoic acid disrupts membrane energetics and various cellular functions (36).

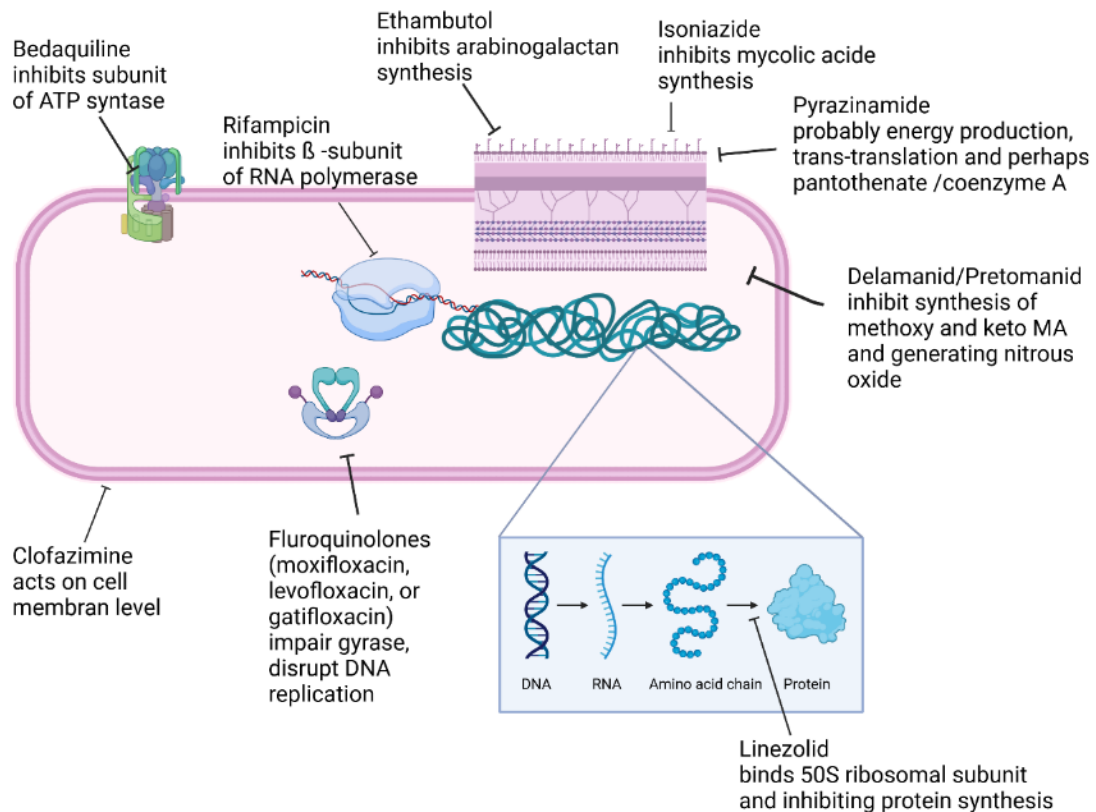


Figure 1.3: Mode of action antimycobacterial therapeutics. Illustration of MTBC bacterial cell, highlighting known targets for a range of anti-TB drugs. Figure modified from Darby et al.

However, the treatment regimens are hampered by high pill burdens, long treatment durations, and frequent onset of toxic side effects. These challenges often lead to low patient compliance, contributing to treatment failure, disease relapse and development of drug resistance (37). In response, the development of shorter-term regimens has been a major public health focus. A novel 4-month regimen has been recently introduced, replacing RIF and EMB with rifapentine and MFX (a fluoroquinolone (FQ)) (2HPMZ / 2HPM). The new regimen, which includes (INH, rifapentine, MFX, and PZA, has shown comparable efficacy to the standard 6-month treatment and has been recommended by WHO since December 2022 for patients, as well as for children and adolescent aged 3-month and older (29,37,38).

Research underscores the strong correlation between high TB burdens and low social protection, addressing these factors could reduce incidence rates by up to 80 % (21,39). To mitigate patient noncompliance, the WHO has implemented Direct Observed Treatment, Short-course (DOTS), which has significantly increased treatment success rate. However, challenges remain, particularly in regions with limited access to necessary infrastructure, as studies from Ethiopia have demonstrated (37,40–43). Effort to improve compliance resulted in the design of a fixed-dose combinations (FDC), which reduce the pill burden (44). Notably, the global success rate of FDC was around 86 % as reported in 2020 (44), although concerns about bioavailability of RIF in this formulation persists (45,46).

Despite these advancements, TB remains at the forefront of the global antimicrobial resistance crisis. Drug resistance has been swiftly emerging to every new antibiotic on the market and spreading (35). Treatment for MDR-TB has seen improvements, initially shortening the duration to 9-12-month, replacing injectable kanamycin or capreomycin with amikacin. Due to the permanent side effects caused by the toxicity of injectables, WHO recommended a 9-month all-oral regimen containing BDQ in 2019 (29,47). This complex regimen involves 6-month BDQ application combined with FQ (levofloxacin / MFX) ethionamide, EMB, INH, PZA and 4 month clofazimine (CLZ) (duration increases if sputum smear remains positive), then 5 months again FQ (levofloxacin / MFX), CLZ, EMB and PZA (47). However, the use of FQ and the variety of antibiotics makes this regimen unsuitable for patients with complicated resistance profiles or FQ resistance. Studies like the Nix-TB and TB-PRACTECAL trials have explored novel treatment regimens, leading to the development of BPaLM, which combines BDQ, pretomanid (PRE), LIN and MFX for 6-month (up to 9-month if smear positive after 6-month) (47). While BPaLM is a major improvement, progress in development of antibiotics remains slow (38). Longer treatment regimens, such as 9-month or 18-month courses, remain necessary for cases where shorter regimens are unsustainable due to drug intolerance, resistance, or other concerns (38).

In an effort to improve treatment approaches, host-directed therapeutics (HDTs) and precision medicine have emerged. HDTs aim to modulate the immune system in preventing non-productive inflammation and immune responses (43). Often utilizing repurposed compounds, HDTs represent a promising strategy for fast-tracking approval by regulatory authorities (43). However, the ultimate advancement in treatment strategies lies in precision medicine, where treatment strategies are tailored to both the individual patient and the infectious MTBC strains (48,49). This approach combines cutting-edge scientific discoveries and technological advances to foster personalized drug dosing and treatment durations. Technologies like next-generation sequencing (NGS) enable rapid assessment of drug susceptibility prediction within a week of diagnosis. Additionally, modulating the host immune system and considering host genetics could further personalize treatment (48). Despite the promise of these new technologies, their high costs limit widespread adoption. Therefore, ongoing development of standard treatment strategies remain crucial until precision medicine becomes accessible to all patients.

1.4 MTBC bacteria resistance mechanism

Resistance mechanisms arise when MTBC bacteria undergo genetic mutations that enable it to survive despite the presence of antibiotics (50). As drug-resistant strains spread, they complicate treatment regimens and undermine control efforts (37,49,50). The transmission of these drug-resistant strains further exacerbates the problem, making it more difficult to contain and eradicate TB on a global scale (51).

To effectively fight the tuberculosis crisis, resistance mechanisms need to be fully understood (52). Generally, drug resistance can be classified as either intrinsic or acquired (35). Intrinsic resistance refers to innate characteristics (e.g., physiological) of the bacteria, where no additional mutations are needed to render the antimicrobials ineffective. In MTBC strains, intrinsic resistance can involve genes of virulence and growth, such as cell wall-associated genes or lipids which foster drug resistance (53,54). The waxy cell wall, a major physiological factor, makes most drugs unable to penetrate (35). The complex structure consists of, in brief, peptidoglycans attached to high branched arabinogalactan layer and long chain mycolic acids, finished by acyl lipids, surface proteins and lipoarabinomannans (LAMs) spanning the whole structure (55). Drugs have been designed to specifically target this structure, for example EMB, which targets arabinosyltransferase (53). Furthermore, antibiotic degrading enzymes such as Beta(β)-lactamases induce resistance to β -lactam antibiotics (e.g. meropenem), as they hydrolyse a specific bond in the β -lactam ring structure (56). Acquired resistances evolve in the pathogen as mutations in target genes i.e., the direct drug target or mycobacterial enzymes which activate prodrugs. These can result in either high-level or low-level resistance (30). Some species are able to acquire resistances by sharing plasmids through horizontal gene transfer (HGT). This is not observed in MTBC strains, which gain drug resistance solely through chromosomal mutations (57). High-level resistance often results from point mutations in the direct target or mycobacterial enzymes. A selection of mechanisms associated with acquired resistance is depicted in Table 1.1.

Table 1.1: Mechanisms associated with acquired resistance against rifampicin, ethambutol and bedaquiline. Adapted from Dookie et al. 2018 (49), sources referenced in the text.

Drug	Gene	Resistance mechanism
Rifampicin	Mutation in <i>rpoB</i> , encoding RNA polymerase. Mutation in Rifampicin Resistance Determining Region (RRDR)	Reduced binding affinity of RIF on target, bactericidal, inhibits RNA synthesis.
Ethambutol	Mutation in <i>embB</i> , inhibit arabinogalactan biosynthesis	<i>embCAB</i> operon encodes arabinosyl transferase enzyme
Bedaquiline	-Mutations in the <i>atpE</i> gene, encoding the beta subunit of ATP synthase. -Mutations in <i>Rv0678 (mmpR)</i> coding transcription regulator of efflux pump system	-Inhibits the adenosine triphosphate (ATP) synthase, essential for energy production. High-level resistance -Off-target mutations confer low level resistances as supposed to enhance <i>mmpS5–mmpL5</i> efflux pump system (30,58–61).

Active export of antibiotics

Efflux pumps are membrane-spanning exporter and / or importer proteins that play a crucial role in the active transport of antibiotics and other substrates. These pumps vary in their energy sources, with some being driven by the proton motive force, while others rely on ATP. In MTBC strains, a significant transporter family involved in this process is the Mycobacterial membrane protein Large (MmpL), which belong to the Resistance-Nodulation-Cell Division (RND) transporter subclass known for MDR pumps (62). A well-known example within this family is the MmpS5-MmpL5 efflux pump system, which is most notably responsible for exporting BDQ, but also CLZ and others. The mycobacterial membrane protein Small (MmpS) is located in close proximity to the *mmpL* genes and are thought to act as membrane fusion proteins, locating the complex in the membrane. However, the precise mechanism is insufficiently understood (62).

In the context of low-level BDQ resistance, mutations frequently occur in the gene *Rv0678*, which encodes a transcriptional regulator. This regulator is believed to inhibit the transcription of the efflux pump genes *mmpS5*-*mmpL5* (*Rv0676c*, *Rv0677c*) located downstream of its promoter by binding to DNA (Figure 1.4) (63,64). Mutations in *Rv0678* can impair this regulatory function, leading to increased transcription of the efflux pump (61,65). These mutations are dispersed throughout the *Rv0678* gene, contributing to the development of resistance by promoting the overexpression of the efflux pump.

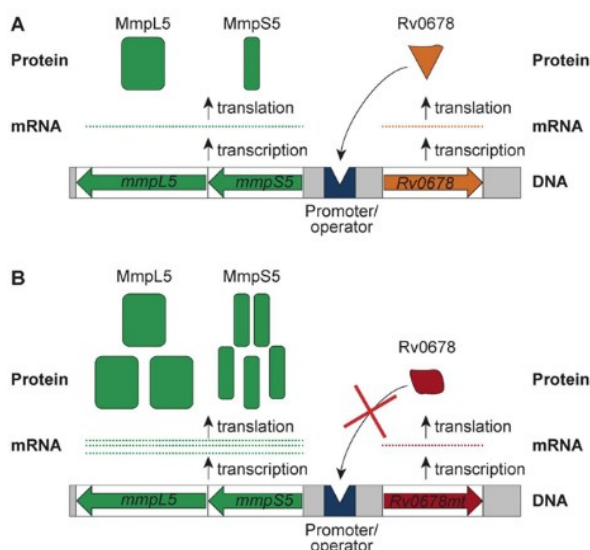


Figure 1.4: Low-level bedaquiline resistance mechanism in clones with *Rv0678* mutations. (A) In wild-type strains, the *Rv0678* transcriptional regulator controls the expression of the *mmpS5*-*mmpL5* efflux pump system. It is thought that the *Rv0678* protein binds to the intergenic promoter region, thereby inhibiting the downstream transcription of the *mmpS5* and *mmpL5* genes. (B) Clones with *Rv0678* mutations, it is supposed that specific mutation types can impair the function of the *Rv0678* regulator. This protein malfunction leads to insufficient binding to the promoter region, resulting in less inhibition of transcription of the *mmpS5* and *mmpL5* efflux pump genes. Consequently, this causes an increased expression of the efflux pump. Figure sourced from Andries et al. 2014.

Metabolic plasticity and decreased antibiotic efficacy

The adaptability of MTBC strains is a key factor for their success, enabling them to sense and respond to various environmental changes with remarkable versatility. MTBC strains are capable of growth both intracellularly and extracellularly, with their metabolic plasticity allowing them to switch between different metabolic pathways depending on the availability of nutrient sources (66). They primarily utilize three major pathways: tricarboxylic acid (TCA) cycle, pentose phosphate pathway (PPP) and glycolytic (Embden–Meyerhof–Parnas (EMP)) pathway, which are augmented by enzymes linking glycolysis and the TCA cycle (67). Additionally, the methyl citrate cycle and methylmalonyl pathway are employed to metabolize propionyl-CoA from sterols, uneven branched chain fatty acids, and amino acids (68). Propionyl-CoA is crucial for virulence and cell wall stability, as it serves as a precursor for the synthesis of phthiocerol dimycocerosate (PDIM), polyacylated trehalose and sulfolipids (important lipids of the cell wall) (69,70). Nevertheless, its accumulation can also be toxic for the bacteria (71).

It has been reported that under environmental stress like nutrient starvation, hypoxia, or antibiotic treatment, subpopulations of MTBC strains reduce their metabolic activity towards a non-replicating state, which increases their tolerance to antibiotics (72). For example, studies on the relationship between fatty acid metabolism and drug resistance in knockout strains have shown that depletion of *prpD*, a key entry gene of the methylcitrate cycle, results in decreased tolerance compared to the ancestral strain (72).

This metabolic plasticity is crucial for maintaining a balance of essential cofactors (66). While an accumulation of these cofactors can become toxic, they are also vital in adequate amounts for key cellular processes (67,73,74). For example, iron plays a key role in various biological functions. In response to iron depletion, MTBC strains can upregulate iron importers like *irtA-B*. Conversely, when iron levels are excessive, they increase the expression of iron storage proteins, such as *bfrB*, to prevent toxicity (73). Such environmental changes are primarily sensed by two-component systems (TCSs) (75). In these systems, a sensor kinase detects environmental fluctuations and activates a response regulator, which in turn induces specific pathways. MTBC strains possess 11 TCSs, including MprRAB, PhoPR, PrrAB and DosR / DosS which are known to induce virulence (76,77). Of these, DosS / DosR play a central role in the regulation of dormancy by reducing crucial metabolism in response to unfavourable conditions like hypoxia (77–81).

Dormancy and drug resistance

Dormancy in MTBC bacilli increases drug tolerance and complicates treatment (82). The reduced metabolic activity of dormant bacilli not only makes them harder to detect and eradicate by the immune

system but also reduces the efficacy of antibiotics(83). TB infections arise from a heterogeneous population of MTBC strains including both actively replicating and dormant bacilli (83). Bacterial drug resistance is typically identified by an increase in the minimum inhibitory concentration (MIC) of a drug (84). The MIC refers to the *in vitro* measurement of a bacterial strain's susceptibility or resistance to specific antibiotics (85). This differs from drug tolerance, which is measured by an increased duration required to kill 99 % of the bacterial population (MDK₉₉), and persistence, which refers to a subpopulation of bacteria that survives prolonged antibiotic exposure without growing (30,84,86).

1.5 Evolutionary medicine

Given the immense ability of MTBC strains to adapt to various treatments, there is an urgent need for innovative approaches to avoid resistance development or render treatment regimens more effective, especially in light of the growing public health crisis posed by MDR- and XDR-TB (87). Historically, every time a new anti-TB drug was introduced, resistance to it emerged shortly thereafter (88). As explored in the previous section, various types of resistance mechanisms (via chromosomal mutations) lead to increased MIC or render the pathogen no longer treatable (49). Regimens to further decelerate the pace of resistance evolution during the standard treatment regimens, alternative strategies need to be investigated. These insights can further be harnessed to tailor treatment regimens for individual patients (precision medicine) (87).

The field of evolutionary medicine, also known as Darwinian medicine, is an interdisciplinary concept that applies principles of evolutionary biology to understand health and disease (89). This approach provides different scopes and out of the box observations to investigate how evolutionary processes such as natural selection, genetic drift, and adaption have shaped our physiological response to various diseases (90). Central to evolutionary medicine is the concept that many health issues arise from mismatches between our evolved biology and the modern environment (89,90). Furthermore, evolutionary medicine investigates the role of genetic variation and trade-offs in disease susceptibility (91). Were some genetic traits give advantages in diseases outcome they may be disadvantageous in a more general sense, for example like the sickle cell anaemia protects against malaria but can cause vessel occlusion (91).

Understanding the evolutionary basis of disease also involves examining how pathogens evolve and respond to treatment. Applying principles of evolutionary medicine can advance TB therapy and might restrict or slow down resistance evolution, thus be a potent option, especially for treatment and prevention of MDR-TB cases (48,87,89). In applying evolutionary medicine to TB research, the most interesting topics include: antibiotic synergy and antagonism, cellular hysteresis, collateral sensitivity, and cross-resistance (87).

1.5.1 Antibiotic synergy und antagonism

Determining the interaction type between drugs and chemicals has become increasingly important in the biomedical field (92–94). As treatment regimens grow more complex and incorporate various compounds, it is crucial to understand how these agents interact. Antibiotic synergy is observed when two or more agents have a stronger killing or suppressive effect than when applied individually (95). In theory, this effect would allow lower dosing of each agent with the same therapeutic effect, thereby reducing potential side effects (95).

Antagonism, on the other hand, occurs when the combined effect between two or more reagents is lower than both individual effects (96). This can result in a reduced therapeutic effect, and the drug concentration or combination needs to be adapted for treatment success (96). An example for this would be a shared receptor, where both drugs compete for the receptor site. In contrast to synergy or antagonism, additive effect occur when two or more compounds act independently, and the overall effect is just the sum of their individual effects (96). The Loewe Additivity and the Bliss Independence models can be used for detection and quantification of synergism and antagonism (97–99). The Loewe Additivity assumes that the agents have similar mechanisms of action and assesses whether their combined effect deviates from the expected additive effect (97). The Bliss Independence Model supposes that the agents act independently of each other and evaluates whether their combined effect differs from the expected outcome based on their individual actions (96,97). A study on measuring drug interactions by diagonal measurement of *n*-way drug interactions using the Bliss multiplicative model in MTBC strains, reported that, most synergy was measured in combination of two antibiotics rather than combination of three or four antibiotics (100).

In summary, synergism and antagonism are important concepts that describe how agents interact to produce combined effects that are either greater or less than the sum of their individual effects (97). However, Larkins-Ford and colleagues reported that some combinations, despite showing strong antagonism *in vitro*, can still be highly effective *in vivo*, as demonstrated by the BPAL regimen (101). This highlights that the treatment success may not hinge on a single factor but rather on a combination of factors such as dosing and the microenvironment (102). Notably, dosing was identified as being more critical to treatment success than synergy (101). Such investigations are crucial for optimizing therapeutic strategies to enhance patient outcomes.

1.5.2 Cellular Hysteresis

Cellular hysteresis refers to an immediate physiological effect due to the interaction between two antibiotics (103). This long-term effect has the potential to influence treatment efficacy positively or negatively, and in some cases, can circumvent evolution of resistance (103). This has been demonstrated *in vitro* for pathogens such as *Pseudomonas aeruginosa* and *Escherichia coli* (104,105).

The sequential administration of one antibiotic followed by another, rather than concurrently, has been shown to result in a treatment enhancing effect known as negative cellular hysteresis (103,104,106). This approach leverages the immediate physiological changes induced by the first antibiotic, which can enhance the efficacy of the second antibiotic (103). This effect is thought to be non-genetic, as the switch between antibiotics occurs too rapid for genetic alterations to take place, usually below the generation time of the bacteria (103,106). This phenomenon can be drug sequence and combination dependent, as in one direction the treatment can result in negative cellular hysteresis, subduing the pathogen, and when the drug order is switched it could have no effect, or a protecting effect for the pathogen, i.e., positive cellular hysteresis (107).

Rapid drug switching is believed to have the potential to circumvent or delay the evolution of resistance, making it a promising strategy for managing drug resistance (103,106). However, as far as our knowledge goes, it has not been investigated in MTBC strains. Understanding and applying this concept in the context of TB treatment could provide new avenues for combating this challenging disease (108).

In summary, cellular hysteresis represents an immediate, non-genetic response to antibiotic treatment that can significantly impact the effectiveness of sequential antibiotic therapies (103,107,108). By exploiting this phenomenon, it may be possible to enhance treatment outcomes and slow down or circumvent development of drug resistance in various bacterial pathogens (103,106,108).

1.5.3 Collateral Sensitivity

Collateral sensitivity is a phenomenon in which, in contrast to cellular hysteresis or synergy, the evolution of resistance to one drug increases the susceptibility to another drug (109–111). The mechanisms underlying this effect are complex and depend on the drugs and organisms involved (110). Two mechanisms resulting in collateral sensitivity include: metabolic trade-offs, where resistance to one drug induces a metabolic burden on the organism, making it more susceptible to another drug that targets another metabolic pathway; and specific genetic mutations, which confer resistance to one drug can increase on the other hand sensitivity to another drug (108,109).

In MTBC, drug-resistant strains have been found to display increased susceptibility to β -lactam antibiotics (112). Research revealed that treatment with standard TB drugs such as EMB, ethionamide,

levofloxacin, aminoglycosides, streptomycin, INH, PZA, and RIF induces genes like *blal* (*Rv1846c*) that suppress β -lactamase activity (113). This phenomenon, offers potential for combating drug resistance (113). However, more research is needed to fully understand and harness this mechanism for therapeutic purposes.

1.5.4 Cross resistance

Cross-resistance arises when a mechanism that a pathogen uses to resist one drug also provides protection against another drug (64). This can occur through various mechanisms, including genetic mutations, efflux pumps, and changes in cellular targets (114). For instance, efflux pumps, which are protein complexes that actively pump a wide range of drugs out of the cell, reducing their intracellular concentrations and effectiveness (62,115–117). Overexpression of efflux pumps, such as *mmpS5-mmpL5* can confer resistance to multiple antibiotics, most famously BDQ and CLZ (61,117–120). Similarly, mutations in the *inhA* gene, the target of INH and ethionamide, demonstrate cross-resistance mechanisms that were confirmed 30 years after their initial hypothesis (121). Advances in molecular technology now allow for better detection of resistance and cross-resistance, a critical factor in designing effective treatment regimens (28).

1.6 Next generation sequencing history and importance in MTBC strains control

Next-generation sequencing (NGS) has revolutionized the field of genomics, providing unprecedented speed and accuracy in sequencing DNA and RNA (122,123). These techniques are crucial for understanding evolution and response mechanisms of MTBC strains to environmental changes such as antibiotic treatments (124). This knowledge on resistance mechanisms can be harnessed to improve resistance detection and design tailored treatment regimens (precision medicine) (87,124,125).

The advent of next-generation sequencing technology dates back to the late 20th century with the introduction of Sanger sequencing, developed by Frederick Sanger in 1977 (126,127). This chain-termination method, determined the nucleotide sequence of the DNA by incorporating chain-terminating dideoxynucleosides during DNA replication (126). Herby, DNA fragments with varying length are generated and the size separation reveals the original DNA strand (127). This method remained the gold standard for sequencing for decades due to its precision in reading nucleotide sequences (123,126). In 1986, Applied Biosystems automated this technique, which increased the speed and efficiency (123,128). The initiation of The Human Genome Project, an international research initiative, aimed to map the whole human genome utilizing Sanger sequencing (129). This project paved the way for higher throughput and lower cost sequencing technologies i.e., NGS (130). In 1995, the first whole-genome

sequencing of an organism (*Haemophilus influenzae*), demonstrated NGSs feasibility for pathogen genome analysis (131,132). In 1998, Cole and colleagues sequenced the complete genome of *M. tuberculosis* H37Rv (133). In 2005, the first commercial NGS platform, 454 Pyrosequencing, was introduced on the market by Roche (134,135). This technology combined pyrosequencing with a stationary DNA template immobilized on nano beads, enabling massive parallel sequencing compared to Sanger methods (135). Only a year later Illumina launched the Genome Analyzer, revolutionizing the market with a new technique, sequencing by synthesis (SBS), increasing throughput and accuracy (122). In 2010, Life Technologies introduced the Ion Torrent Sequencing, the first semiconductor-based sequencer (136). From 2010 onwards the development and improvement of NGS-technologies remained steady (132).

Whole genome sequencing (WGS) of MTBC strains has become a crucial component of TB diagnosis, treatment, and surveillance efforts (137,138). Recent studies on MTBC strains, including one analysing over 10 000 isolates, have demonstrated that genotypic predictions of drug susceptibility based on WGS are highly consistent with phenotypic susceptibility results for first-line TB drugs (139). This accord emphasizes the potential of WGS as a reliable method for determining drug resistance, which can significantly improve TB management (137–140). The advantages of WGS over traditional phenotypic tests include shorter processing times, enhanced reliability, and potentially lower costs, underscoring its growing importance in TB control (137,139,141). Despite these advantages, there are currently multiple workflows (e.g., MTBseq, UVPReSeqTB) for WGS analysis, each employing different methodologies (138,142,143). The lack of standardized approaches in variant calling and validation poses a challenge to ensure that all detected genetic variants are accurate and relevant (138,143). Therefore, establishing common validation criteria for WGS pipelines would be a critical next step, which should be prioritized by regulatory bodies to ensure the accuracy and consistency of WGS data (138). Once such regulation criteria are established WGS-based DST has the potential to become the new standard (138). Furthermore, WGS offers a comprehensive view of genetic variation across the entire genome, surpassing current WHO-recommended genotypic tests, which focus primarily on specific hotspot regions and a limited set of antibiotics (137,140,143). WGS can identify a wider range of resistance-conferring mutations, providing a more complete picture of drug susceptibility (138,143). However, for WGS to be widely adopted in clinical settings, its usage and interpretation must also be simplified and standardised (140). Healthcare professionals need user-friendly tools and clear guidelines for interpreting WGS data to ensure its effective integration into routine TB diagnosis and treatment (137,138,140). Developing such frameworks will be essential for increasing the potential of WGS in combating TB on a global scale (138).

Illumina NGS

Illumina NGS technology enables high-throughput sequencing based on SBS. This method involves fragmenting the DNA into small pieces called reads, adapters are then attached to the ends, which are then immobilized on a solid surface like a glass slide called a flow cell (144). Each fragment is amplified to form clusters of identical DNA fragments, which significantly increases the fluorescence-based signal during sequencing (Figure 1.5). The sequence of the reads is determined when fluorescently labelled nucleotides are incorporated; these are added one at a time to the increasing DNA strand by DNA polymerase (145). Fluorescence is emitted after each incorporation, allowing for identification of each base upon its incorporation, and captured with a high definition (HD)-camera (145). The sequence of the DNA is determined based on the order of this fluorescent signals across millions of spots on the flow cell, thereby allowing for massive parallel sequencing of millions of fragments (reads) (73).

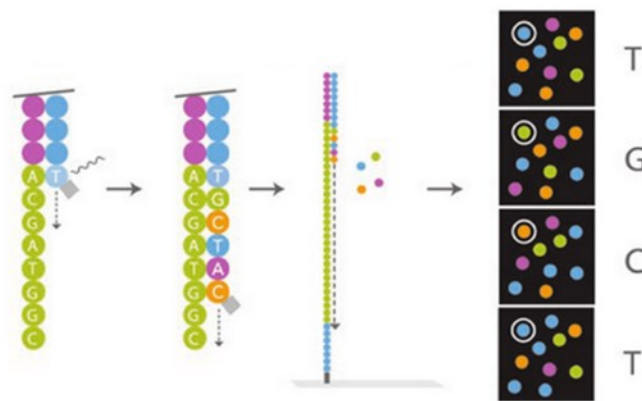


Figure 1.5: Sequencing step of Illumina® technology overview. After the cluster generation step (not shown), the sequencing step follows by extension of the first base, reading and deblocking. This extension is repeated to extend the strand and base calls are detected via the HD-camera. From Mathieu Lemaire researchgate.net and images courtesy of Illumina, Inc., Dan Diego, CA, USA.

Study objectives

To enhance treatment strategies and discover approaches to circumvent or slow down the evolution of resistance, it is essential to gain a comprehensive understanding of the mechanisms that lead to drug resistance. Ultimately, understand resistance and exploring alternative treatment strategies is crucial to address the ongoing evolutionary arms race between humanity and pathogens. The specific objectives of this study are:

Better understand bedaquiline resistance mechanisms:

- Assess the transcriptome of drug-resistant MTBC strains with and without exposure to their target drugs.
- Identify key genetic and regulatory changes associated with resistance.
- Understand the molecular basis of drug resistance by comparing the transcriptome between resistant and non-resistant strains.

Explore alternative treatment strategies:

- Investigate the potential of sequential antibiotic treatments to induce negative cellular hysteresis in MTBC strains.
- Exploit the immediate physiological changes induced by the first antibiotic to enhance the efficacy of the second antibiotic.
- Understand the transcriptomic changes associated with sequential treatment to identify the molecular mechanisms underlying this effect.

2 Materials and Methods

2.1 Materials

2.1.1 Consumables

Product	Order number	Company
6-well plate, flat bottom, sterile	CLS3516	Corning Costar
96-well plate, flat bottom, sterile	83.3924	Sarstedt
Adhesive film for microplates Breath®easy	Z380059	Sigma-Aldrich
AMPure® PB Beads	100-265-900	PacBio
BD MGIT™ Tubes	245113	Becton Dickinson
Filter Tips 10 µL	70.1114.210	Sarstedt
Filter Tips 100 µL	70.760.212	Sarstedt
Filter Tips 200 µL	70.760.211	Sarstedt
Filtertips 1000 µl	70.762.211	Sarstedt
Sterican® 0,4 x 25 mm (27G)	9180117	Braun
Cuvettes 10x4x45 mm	67.742	Sarstedt
Cuvette single-use lids	XK25.1	Roth
Inoculation loops (sterile)	612-2498	VWR
Latex gloves (Medium & Small)	MAG 08029	Dahlhausen
Löwenstein-Jensen with glycerol	TV5114C	Artelt-Enclit
Lysing Matrix B 2 mL	116911100	MPBiomedicals
Microscope slides	03-0060	R. Langenbrinck
Nitrile gloves (Small)	MAG 08047	VWR
Nonstick, RNase-free Tubes, 1.5 mL	AM12450	Thermo Fisher
Parafilm®	P7793-1EA	Pechiney
Petri dishes 92 x 16 mm	82.1472	Sarstedt
Petri dishes, square, PS	688161	Greiner
RNeasy Protect Bacteria Mini Kit	74524	Qiagen
RNase free 1,5 mL reaction tubes; low binding	710176	Biozym
RNase free tubes (1,5 and 2 ml)	72.705.200	Sarstedt
Roller bottle (490 cm ²)	430195	Corning
Screw Cap Tube 1.5 mL with lid	72.692	Sarstedt
Seal gas permeable tissue culture 96-well-plates, sterile	045290 Lot	Porvair Sciences
Square media bottle, sterile, PETG, 30 mL	2019-0030	Nalgene™
Stripette® serological pipette 10 mL	4488	Corning Inc.
Stripette® serological pipette 25 mL	4489	Corning Inc.

Product	Order number	Company
Stripette® serological pipette 5 mL	4487	Corning Inc.
Syringe 1 mL	303172	Becton Dickinson
Syringe 20 mL	352054	Becton Dickinson Bioscience
Syringe filtration unit Filtropur 0.2 µm	83.1826.001	Sarstedt
Millex®-LG hydrophilic Fluoropore™ (PTFE) Filter unit 0.2 µm, sterile, PTFE / LCR	SLLG013SL	Merck Millipore Ltd
Filtropur S 0.2 µm sterile, non-pyrogenic	83.1826.001	Sarstedt
Tube 1.5 mL safety cap	S 72690001	Sarstedt
Tube 15 mL, polypropylene screw caps	62.554.502	Sarstedt
Tube 50 mL, polypropylene screw caps	62.547.254	Sarstedt
Tubes 0.5 ml (Qubit)	732-0675	Axygen

2.1.2 Reagents and Buffers

Product	Order number	Company
2-Propanol (Isopropanol)	AE73.1	Roth
BBL™ Brain Heart Infusion Broth, Modified	299070	Becton Dickinson
Buffer AE (5 nM 10 mM Tris / HCl, pH 8.5)	740917.1	Macherey -Nagel
Blood agar TSA, 5 % Sheep blood	254087	Becton Dickinson
CTAB (N-Cetyl-N,N,N- trimethylammonium-bromide)	1023421000	Merck
DEPC water	T143.3	Roth
Distilled H ₂ O (DNase / RNase free; sterile; not DEPC treated)	10977-035	Gibco (life technologies)
Difco™ Middlebrook 7H10 Agar 500 g	262710	Becton Dickinson
Difco™ Middlebrook 7H9 Broth 500 g	1173715	Becton Dickinson
DNase Max™	15200-50	Qiagen
DNA / RNA shield	R1100-250	Zymo
Dimethyl Sulfoxide, sterile filtered (DMSO)	A7248,0010	PanReac AppliChem
Elution Buffer (for PacBio sample prep)	101-633-500	Pac-Bio
Elution Buffer AE (5 mM Tris / HCL, pH 8.5)	12716563	Fisher Scientific
Ethanol 100 %	108543	Merck
Glycerol (Glycerin) anhydrous A2926,1000	0P011742	PanReac AppliChem
HCl 0.1 mol / L	2315957	Roth
HCl-ethanol 3 % for histology	6477.3	Roth
Isoamyl alcohol	T870.1	Roth

Product	Order number	Company
LB-Broth 1 kg	L3022	Sigma-Aldrich
Methylene blue solution	AE64.3	Roth
Multiply- μ Stripe 0.2 ml chain	72.985.002	Sarstedt
NaCl	9265.2	Roth
NaOH	20765.03	Serva
OADC Middlebrook Enrichment 500 mL	212351	Becton Dickinson
Oelic acide	A4420	Applichem
Proteinase K	19133	Qiagen
Pursept® A Xpress	230131	Schülke & Mayr
PBS, sterile w / o Ca und Mg	PO436500	Pan Biotech
Resazurin sodium salt	R7017-1G	Sigma-Aldrich
SDS	2326.2	Roth
RNase-ExitusPlus™	A7153,0500	Applichem
RNase-Free DNase Set	79254	Qiagen
Sodium Chloride	131659.1211	PanReac AppliChem
TRIzol®	19698001	Ambion by life technologies
Tris-Hydrochlorid	9090.2	Roth
Tween® 80	P4780	Sigma-Aldrich
Taq DNA Polymerase with Standard Taq Buffer	M0273S	New England Biolabs
Water, sterile	0082479E	Braun

2.1.3 Antibiotics

Drug	Form	Reconsti- tuted in	Order number	Company
Bedaquiline	powder	DMSO	43211	AstaTech
Rifampicin CRS	Powder	DMSO	000j8u	European Pharmacopeia Reference Standart
Ethambutol Hy- drochloride CRS	powder	DMSO	007DEf	European Pharmacopeia Reference Standart

2.1.4 Kits

Product	Order number	Company
Qubit dsDNA HS assay kit (500)	Q32854	Life Technologies
RNAese® Mini Kit (250)	74106	Qiagen
RNA clean & concentrator-5™ (DNase Included) -5 (50 rxns)	R1013	Zymo Research
Mo Bio DNase Max Kit	15200-50	Qiagen

Product	Order number	Company
HS RNA Kit (15NT)	DNF-472-0500	Agilent
HS NGS Fragment Kit (1-6000bp)	DNF-474-1000	Agilent
NGS Fragment Kit (1-6000bp) (SS NGS Fragment Kit)	DNF_473-1000	Agilent
Byam2015:		
Illumina Tagment DNA Enzyme and Buffer Large Kit	20034198	Illumina
IDT Illumina DNA / RNA UDI A Tag 96 Idx 96 Spl	20027213	Illumina
IDT Illumina DNA / RNA UDI B Tag 96 Idx 96 Spl	20027214	Illumina
IDT- Illumina DNA / RNA UDI C Tag 96 Idx 96 Spl	20042666	Illumina
IDT- Illumina DNA / RNA UDI D Tag 96 Idx 96 Spl	20042667	Illumina
Celero EZ:	0568-24	Tecan
Celero EZ DNA-Seq Core Module	30188860	Tecan
10 nt UDI-A 96-Plex Adaptor Plate	30219997	Tecan
10 nt UDI-B 96-Plex Adaptor Plate	30219998	Tecan
10 nt UDI-C 96-Plex Adaptor Plate	30219999	Tecan
10 nt UDI-D 96-Plex Adaptor Plate	30220000	Tecan
NuQuant NGS Library Quantification Module	30188703	Tecan
Zymo-Seq RiboFree Total RNA Library Kit genutzt erst die Version 1.3.0	R3003	Zymo Research
Zymo-Seq RiboFree Total RNA Library Kit genutzt erst die Version 2.0.0	R3003	Zymo Research

2.1.5 Self-made media

Middlebrook 7H9 medium 1 L	
4.7 g	7H9 Middlebrook
900 mL	Dissolve in dH ₂ O
900 mL	Dissolve in dH ₂ O
+ 2 mL	Glycerol (final conc. 0.2 %)
+2.5 mL	20 %Tween80 (final conc. 0.05 %)
	Sterile filter or autoclave: dissolve amount of 7H9, add Glycerol, 20 min / 121° C)
	add amount of Tween after autoclaving
	Store at 4° C for ~6 month
+ 100 mL	OADC Middlebrook enrichment

Middlebrook 7H9 for Transcriptomics	
4.7 g	7H9 Middlebrook
900 mL	Dissolve in dH ₂ O
+ 5 mL	Glycerol (final conc. 0.5 %)
	Sterile filter or Autoclave: Dissolve amount of 7H9, add Glycerol, 20 min / 121° C)
	Add Tyloxapol after autoclaving

	Store at 4° C for ~6 month
0.25 mL	Tyloxapol 20 %
+ 100 mL	OADC Middlebrook (final conc. 10 %)

Middlebrook 7H10 plates 1 L	
Middlebrook 7H10 Agar last 3 month / 1 L = 40 Plates a 25 mL	
19 g	7H10 Middlebrook
900 mL	dH ₂ O (1 L glass bottle) let Stirrer in bottle!
+ 5 mL	Glycerol
	Autoclave 20 min / 121° C Cool down in water bath (45-50° C) or on magnet stirrer
+100 mL	Bovine sera (inactivated)
	(+antibiotics) (+10 mL 1 % Cycloheximide final conc. 0.01 %, → optional only when fungi contamination risk) 25 mL per plate, work sterile (safety cabinet, use disposable pipette)

0.05 % Tween80 + aqua for dilution rows 2L	
5 mL	20 % Tween80
2000 mL	Aqua (sterile) Sterile filtration in safety bench

OADC self-made	
8.5 g	NaCl
20 g	D-Glucose (sin aqua), dilute in oleic acid
50 g	Albumin Fr. 5 pH 7.0, dilute in oleic acid
0.03 g	Catalase
	Oleic acid

2.1.6 Laboratory equipment

Product	Company
Analytical Balance EMB 2000-2	KERN
Autoclave	Systec
Synergy 2 plate reader	BioTek (Agilent)
Centrifuge 5430	Eppendorf
Centrifuge 5430	Eppendorf
Multifuge	Eppendorf
Fragment Analyzer™	Advanced Analytica
Heating Block	Biozym Scientific GmbH
Hera Safe – Safety bench	Thermo, Germany
Incubator	Memmert GmbH + Co KG, Schwäbisch Hall, Modell: 100-800

Product	Company
Incubator	Firma: Heraeus Instruments, Hanau, Typ B20
Incubator	Thermo Scientific / Thermo Electron LED GmbH, Langenselbold, Typ: Heratherm IMH400-S
Multi-channel pipettes (100µl, 300µl)	Eppendorf
NextSeq2	Illumina
NextSeq1000	Illumina
Refrigerator	Privileg
Refrigerator	Liebherr Profiline
Pipet boy	Integra Biosciences
Pipettes (10µl, 100µl, 200µl, 1000µl)	Eppendorf
Freezer -80° C	SANYO Electric Co., Ltd; Ultra-low temperature freezer, Typ MDF-U73V
Qubit 3.0	Life technologies
NanoDrop™	Thermo Fischer Scientific
FastPrep24	MP Biomedicals™
Spectrophotometer DS-11 FX	DeNovix
Thermocycler	Analytik Jena
Thermocycler C1000 Touch	Biorad
Vortex Genie2	Scientific Industries
Vortex Genius 3	IKA Vortex
Water bath	Köttermann

2.1.7 Software

Software	Developer	Free Software Source
Gen5™	BioTek	www.agilent.com/en/product/cell-analysis/cell-imaging-microscopy/cell-imaging-microscopy-software/biotek-gen5-software-for-imaging-microscopy-1623226
Microsoft® Office	Microsoft Corporation	www.microsoft.com
RStudio version 23.09.1+494	© The R Foundation	https://cran.r-project.org/
R version 4.3.2	© The R Foundation	https://svn.r-project.org/R-dev-web/trunk/index.html
DESeq2 package version 1.42.0	Love MI, Huber W, Anders S (2014)	https://bioconductor.org/packages/release/bioc/html/DESeq2.html
Venny 2.1	Juan Carlos Oliveros, BioinfoGP	https://bioifogp.cnb.csic.es/tools/venny/
DAVID	Dr. Weizhong Chang, Mr. Brad T. Sherman	https://david.ncifcrf.gov

2.2 Methods

2.2.1 Basic laboratory procedures

2.2.1.1 Colony forming unit plating and counting

Quantifying a viable population involves plating and counting colony-forming units (CFU) on a solid medium. To begin, a serial dilution of the bacteria from 10^{-1} to 10^{-4} , with a dilution factor of 1:10, was performed in a medium comprising sterile water and 0.05 % Tween80, a detergent that prevents mycobacteria from forming aggregates.

First, 225 μL of the water and 0.05 % Tween80 was dispensed into each well of a 96-well plate. Then, 25 μL of the bacterial culture was added into the initial well. The culture was thoroughly mixed by pipetting approximately five times, with a multi-channel pipette, the pipette tips discarded after each mixing. Subsequently, 25 μL from the initial well was transferred to the next, repeating this process for each subsequent dilution. All dilutions were performed in technical triplicates, with the final 25 μL discarded after the last dilution step. Next, 100 μL of each dilution was spread onto half a petri dish containing 7H10 Middlebrook medium supplemented with 0.5 % glycerol and either 10 % inactivated bovine serum or 10 % OADC medium (OADC in the case of antibiotic usage). Alternatively, spot plating was done by dropping 10 μL of each dilution on the agar plates. The plates were then sealed with Parafilm®, bagged in plastic, and incubated upside down at 37° C. CFU enumeration was performed after 14, 21, and 28 days of incubation, with CFU counts determined using Formula 1:

(1)

$$n \text{ colonies} * \text{dilution factor} * 10 = \frac{\text{CFU}}{\text{mL}}$$

2.2.1.2 Determination of colony forming units on antibiotic selection plates

To quantify mutants within a population over time, CFU counts were conducted on antibiotic selection plates. The serial dilution and enumeration procedure closely followed the method described for normal CFU plating (Chapter 2.2.1.1). Instead of bovine serum, 10 % OADC was used to prevent interactions between the serum and the antibiotics. The antibiotic of interest was added at the final concentration specified in Table 2.1 to a portion of the medium, and thoroughly mixed using a magnetic stirring rod. Plates were examined after 14, 21, and 28 days. The mutant-to-CFU ratio was calculated according to Formula 2.

(2)

$$\frac{\text{Mutant CFU/mL}}{\text{Total CFU/mL}} * 10^6 = \text{Mutant per million CFU}$$

Table 2.1: Antibiotic concentrations used in this thesis. The MIC was determined by resazurin emission micro-titer assay (REMA) and concentrations were validated according to WHO (141).

Drug	MIC (REMA assay) [µg / mL]	MIC (literature) [µg / mL]	Mutant selection con- centration [µg / mL]
Ethambutol	2	1 - 5	5
Rifampicin	0.008	0.06 - 0.25 / 0.5	0.25
Bedaquiline	0.12	≤ 0.008 - 0.25 with a mode at 0.06	1.2

2.2.1.3 Optical density measurement

In cuvettes

To estimate the growth of the bacterial culture, turbidity was assessed by measuring the optical density at a wavelength of 600 nm (OD₆₀₀) using a spectrophotometer. Calibration was performed by filling a cuvette (1 cm diameter), with 1 mL of the respective culture media. Then, 1 mL of the culture of interest was introduced (in a new cuvette) into the spectrophotometer for measurement. The method relies on detecting the fraction of light scattered through the medium. The qualitative maximum detection limit is approximately an OD of 1.0. To measure highly turbid samples, 500 µL of the experimental medium was mixed with 500 µL of the sample and then measured. The OD was calculated when diluted by multiplying the obtained value by a factor of 2. Optical density measurement serves as an indicator of culture growth, in real time, and helps to determine the growth phase. However, it should be complemented with CFU plating.

2.2.1.4 Contamination test

2.2.1.4.1 Blood agar sterility check

Blood agar, enriched with 5 % sheep blood, was utilized in this study to observe the growth of fastidious microorganisms as a contamination control. For every culture of *M. tuberculosis*, blood agar was used as a sterility indicator. Approximately 25-50 µL of culture were inoculated onto blood agar plates and then incubated at 37° C for one week. For contamination investigations, blood agar plates were examined in front of a light source. Any growth observed on these plates within the first day or up to 7 days after inoculation suggested potential contamination, necessitating further testing.

2.2.1.4.2 Brain heart infusion medium sterility check

To assess the presence of both fastidious and non-fastidious microorganisms, comprising aerobic and anaerobic bacteria, Brain Heart Infusion (BHI) medium was employed. This medium served as a sterility control for other bacteria or fungi. Initially, 2 x 10 mL of BHI were aliquoted into separate 15 mL screw cap tubes. Then, 50 - 100 µL of the bacterial culture of interest was transferred into one tube, and both tubes were incubated at 37° C for two weeks. Growth was monitored on days 7 and 14 of incubation. The presence of turbidity in the sample medium, in comparison to the control, indicated potential contamination of the culture.

2.2.1.4.3 Ziehl - Neelsen-Staining

The Ziehl - Neelsen staining method was employed to detect gram positive bacteria and distinguish acid-fast bacteria like *M. tuberculosis*. After staining, mycobacteria appeared as pink stained rods, while other structures such as cocci or non-acid-fast bacilli were coloured blue and identified as contamination. This technique served as an additional sterility control in select experiments. First, 10 µL of bacterial culture was spread onto a microscopic glass slide using a sterile loop and allowed to dry at room temperature. Subsequently, the slide underwent two rounds of passing through a Bunsen burner flame (5 flame passes), with a cooling step in between. The slides were then transferred from the BSL3 facility to a chemical hood in a BSL2 laboratory.

Next, the slides were completely covered with carbolic fuchsin solution and gently heated using a Bunsen burner until steam appeared, careful not to boil the solution. After cooling, two additional rounds of gentle heating until steaming were performed. Following a 10-min cooling period, the carbolic fuchsin solution was rinsed off with distilled water. Decolorization was done by dipping the slide in 3 % HCl-ethanol solution, followed by washing with distilled water. To counterstain the decolorized structures, the slide was immersed in a diluted methylene blue solution (1:10 diluted in water) for 1 min and washed with distilled water. Finally, the air-dried slides were examined under oil immersion microscopy.

2.2.2 Phenotypic assays

2.2.2.1 Resazurin Emission Microtiter Assay (REMA)

A swift and cost-effective method for phenotypic DST and determining MIC is the REMA. In contrast to MIC determination using solid media, the REMA assay provides results within 7 days rather than 14 – 21 days. The assay relies on measuring resazurin reduction, indicated by a colour change from blue to pink, with pink coloration indicating cell respiration, thus living bacteria.

First, a bacterial culture was started from frozen stock, and grown at 37° C until reaching an OD₆₀₀ of 0.3-0.4, in either a 30 mL or 60 mL square bottle (Nalgene®), depending on the amount of antibiotics being tested. The day before the experiment, blood agar was inoculated with the culture to confirm sterility as described Chapter 2.2.1.4.1. The culture was then diluted until reached an OD of 0.3 if needed, the culture with OD of 0.3 was diluted 1:20. To prevent evaporation, sterile water was filled in the outer rows of the 96-well plates. Then, 100 µL of 7H9 Middlebrook medium supplemented with 0.2 % glycerol, 0.05 % Tween80, and 10 % OADC was distributed into all wells, with three wells for medium-only controls. The first column was filled with 200 µL of antibiotic culture. Then, 100 µL from the first row was serially transferred to subsequent wells using a multichannel pipette, ensuring thorough mixing and changing tips between transfers, the last 100 µL were discarded. A minimum of three columns with only medium and culture served as antibiotic-free growth controls.

Within the BSL3 facility, 100 µL of the 1:20 diluted bacterial culture was added to each well. No bacterial culture was added to the blank wells. The plates were then sealed with breathable, sterile well plate seals and covered with aluminium foil for light protection, followed by incubation at 37° C for 6 days. On day six, 30 µL of resazurin solution (0.01 % w / v) was added to all wells. The resazurin solution was prepared by reconstituting 1 mg of powder in 10 mL sterile water (Braun), followed by sterile filtration using a Millipore® filter. After an additional 24-h incubation at 37° C, fluorescence and absorbance were measured at 544 / 590 nm and 570 nm, respectively. Plates were read using the Synergy 2 plate reader and the Gen5 software. Visual colour change was also documented by photography (Figure 2.1). The combination of visual colour change with fluorescence and absorbance measurements allowed for determination of the final MIC.

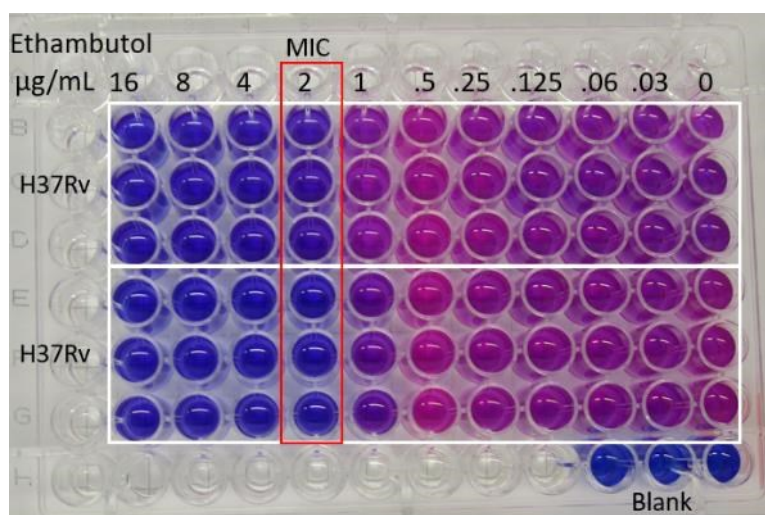


Figure 2.1: Resazurin Emission Microtiter Assay (REMA). Example of the MIC test for *M. tuberculosis* H37Rv with EMB in triplicates. The antibiotic stock was 1:2 serial diluted starting from 16 µg / mL to 0.03 µg / mL. The last column has 0 µg / mL and is the positive growth control. Row H, column 10-12 depict the negative control also use as blank (medium only). The blank is used to subtract the background of the medium from the turbidity of the culture. The red square depicts the MIC, which is the lowest concentration inhibiting the growth. In combination with fluorescence / absorbance measurement in the Synergy2 plate reader, the MIC was determined. Positive growth = pink, inhibited growth = blue. N = 1, technical triplicates.

2.2.2.2 Checkerboard assay

In combination therapy, drug interactions can exhibit positive, negative, or neutral effects, known as synergism, antagonism, or additivity, respectively. To confirm the nature of these interactions, a checkerboard assay was employed. In this thesis the assay was used to determine the interaction between ethambutol and rifampicin. Interaction types were predicted based on a colour change from blue (indicating no growth) to pink (indicating growth) due to resazurin reduction, as observed in the REMA assay (Chapter 2.2.2.1). In addition to assessing the interaction, the MIC of each drug was determined, along with the Fractional Inhibitory Concentration Index (FICI) using the following formula 3:

(3)

$$FICI = FICA + FICB$$

$$FICA = \frac{(MIC \text{ of } A \text{ in presence of } B)}{(MIC \text{ of } A \text{ alone})}$$

$$FICB = \frac{(MIC \text{ of } B \text{ in presence of } A)}{(MIC \text{ of } B \text{ alone})}$$

Interpretation of FICI result was as follows: $FICI \leq 0.5$ indicated a synergistic interaction, FICI of 1 indicated an additive effect, and $FICI \geq 4$ indicated an antagonistic interaction. The checkerboard assay was designed as follows: row A contained 50 µL of 2-fold serial dilutions of drug A, with 4 times the

MIC as the highest concentration, while column 2 contained the same for drug B. Row A and column 2 were filled with 50 μL of 7H9 Middlebrook medium (Figure 2.2). Subsequently, drug A and drug B were combined in a checkerboard pattern starting from well B3, where the highest concentration of each drug was combined, followed by dilutions in subsequent wells. Within the BSL3 facility, 100 μL of *M. tuberculosis* strain H37Rv was added to wells from A3 - A10 to G2 - G10. The plate was then sealed with breathable, sterile plate seals and incubated for 6 days at 37° C. On day 6, 30 μL of resazurin solution (0.01 % m / v) was added to each appropriate well to facilitate colour change observation and interpretation.

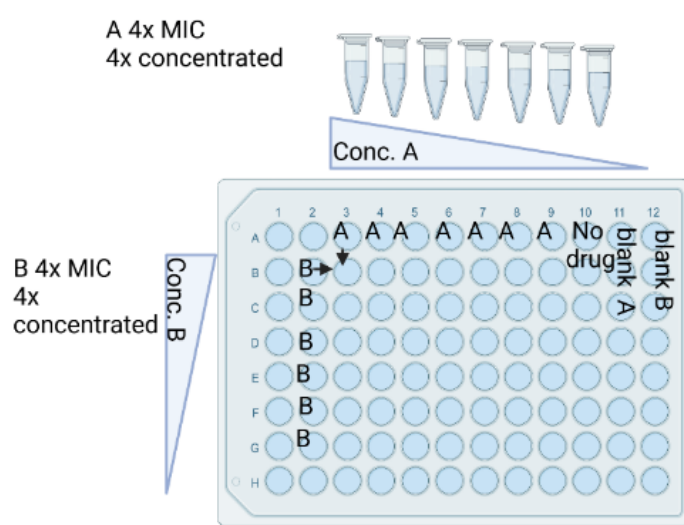


Figure 2.2: 2D Checkerboard assay for drug interaction. The checkerboard assay is used to determine synergy and antagonism along with the minimum inhibitory concentration (MIC). The set-up was the following: column 3 - 9 contained 2x serial dilution of drug A. Row B - G contained a 2x serial dilution of drug B. Row A contains solely a serial dilution of drug A. Column 2 contains a serial dilution of drug B. Here, the MIC for each drug can be determined. Row 10 = positive control (no antibiotic), row 11 and 12 are blanks (medium only).

2.2.3 Evolutionary informed assays

2.2.3.1 Establishing a sequential treatment and time kill assay for *M. tuberculosis*

The parameters for the sequential treatment to investigate hysteresis were adapted from *P. aeruginosa* to *M. tuberculosis* H37Rv, based on the design by Roemhild et al. (103). First, the MIC for wild-type H37Rv was determined using the REMA assay (Chapter 2.2.2.1). Subsequently, subinhibitory MIC concentrations (75 %, 50 %, 25 %, and 12 % of the MIC) were tested in combination for pre-exposure and main exposure. The pre-exposure was carried out in square bottles and the main-exposure carried out in 96-well microtiter plates, measuring bacterial growth by optical density (OD₆₀₀) taken via a BAC-TEC plate reader, allowing for high-throughput testing of parameters (Appendix B Table 0.1).

Three days prior to the experiment, the starter culture was initiated from a frozen stock of *M. tuberculosis* H37Rv. The frozen stock was thawed at room temperature (RT), and 1 mL was passed through

a 26G Braun needle 10 times to disperse the bacterial cells. The suspension was then transferred to 10 mL of 7H9 culture medium consisting of Middlebrook 7H9 broth supplemented with 10 % OADC, 0.05 % Tween 80, and 0.2 % glycerol in a 60 mL square bottle. The culture was incubated at 37° C without CO₂, shaking at 50 rpm. A sample was taken for sterility testing as described in Chapter 2.2.1.4.

On day 2, the optical density at 600 nm (OD₆₀₀) was measured. Once the culture reached an OD₆₀₀ of 0.3–0.6, incubation continued at 37° C until the following day. On the third day, the experimental procedure began with another OD₆₀₀ measurement, and if necessary, the culture was diluted with fresh 7H9 medium to achieve an OD₆₀₀ between 0.3 and 0.6.

Antibiotic Preparation:

A stock solution of 1 mg / mL was prepared for each antibiotic, using an appropriate solvent (DMSO for EMB and RIF). The solutions were sterile-filtered (Millex®-LG hydrophilic Fluoropore™ 0.2 µm PTFE / LCR). Pre-exposure concentrations of each antibiotic were calculated for a final volume of 10 mL. For example, three different pre-exposure concentrations were tested for each antibiotic: 75 %, 50 %, and 25 % of the MIC. A control condition without antibiotics (7H9 medium only) was also included.

Pre-exposure:

The bacterial culture (OD₆₀₀ ~ 0.4) was added to the antibiotic-containing or control medium in a 1:5 ratio. The cultures were incubated at 37° C, standing and protected from light (wrapped in tin foil for RIF) for varying pre-treatment durations (e.g., 2h, 5h and 8h).

Main-exposure:

For the main exposure phase, a single antibiotic (e.g., RIF) was tested at varying concentrations (75 %, 50 %, and 25 % MIC) per experiment, prepared for a final volume of 10 mL in screw-cap tubes. From each condition, 100 µL was dispensed into a 96-well plate following the schema provided in Table 2.2.

After incubation the whole volume from the square bottle was transferred to a 15 mL screw cap tube and centrifugate at 3 500 rpm, for 10 min at RT. The supernatant was carefully removed by pouring or pipetting, and the pellet was resuspended in 1 mL of pre-warmed 7H9 medium. The volume was topped up with an additional 1.5 mL of 7H9 medium and briefly vortexed. The resuspension was transferred to separate reservoirs for each condition, and 100 µL was dispensed into the 96-well plates as outlined in Table 2.2. The final volume per well was then 200 µL (1:2 pre-exposure to main-exposure ratio).

The plates were sealed (adhesive film Breath®easy) and were measured using a plate reader (day 0 = antibiotic switch day). The plates were then returned to the incubator at 37° C, standing for the remainder of the experiment, with periodic measurements taken as required.

Table 2.2: Plating scheme for main-exposure in 96-well plate set up. High throughput parameter test for Hysteresis in MTBC. Pre-75 % EMB = Pre-exposure 75 % the MIC of EMB, Main-75 % = Main-exposure 75 % MIC RIF. The Main-exposure in this example was only RIF (75 %, 50 %, 25 % the MIC) compared with different pre-exposure concentrations of EMB and RIF or no-antibiotic, as depicted on the top. Pre-exposure was dispensed according to the shades of grey.

Pre-75 % EMB				Pre-50 % EMB				Pre-25 % EMB				No-an- tibiotic				Pre-75 % RIF				Pre-50 % RIF				Pre-25 % RIF			
b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b
Main-75 %				Main-75 %				Main-75 %				no-an-tibiotic				Main-75 %				Main-75 %				Main-75 %			
Main-75 %				Main-75 %				Main-75 %				no-an-tibiotic				Main-75 %				Main-75 %				Main-75 %			
Main-75 %				Main-75 %				Main-75 %				no-an-tibiotic				Main-75 %				Main-75 %				Main-75 %			
Main-50 %				Main-50 %				Main-50 %				no-an-tibiotic				Main-50 %				Main-50 %				Main-50 %			
Main-50 %				Main-50 %				Main-50 %				no-an-tibiotic				Main-50 %				Main-50 %				Main-50 %			
Main-50 %				Main-50 %				Main-50 %				no-an-tibiotic				Main-50 %				Main-50 %				Main-50 %			
Main-50 %				Main-50 %				Main-50 %				no-an-tibiotic				Main-50 %				Main-50 %				Main-50 %			
b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b

b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b
Main-25 %				Main-25 %				Main-25 %				no-an-tibiotic				Main-25 %				Main-25 %				Main-25 %			
Main-25 %				Main-25 %				Main-25 %				no-an-tibiotic				Main-25 %				Main-25 %				Main-25 %			
Main-25 %				Main-25 %				Main-25 %				no-an-tibiotic				Main-25 %				Main-25 %				Main-25 %			
No-anti-biotic				No-anti-biotic				No-anti-biotic				No-an-tibiotic				No-anti-biotic				No-anti-biotic				No-anti-biotic			
No-anti-biotic				No-anti-biotic				No-anti-biotic				No-an-tibiotic				No-anti-biotic				No-anti-biotic				No-anti-biotic			
No-anti-biotic				No-anti-biotic				No-anti-biotic				No-an-tibiotic				No-anti-biotic				No-anti-biotic				No-anti-biotic			
b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b	b

b = blank = 7H9 culture medium or braun H₂O for evaporation controle

With this set up various pre-exposure durations were evaluated (drug 1 application), including 1.5, 2, 5, 8, 10 and 13 h. Also, different pre-exposure and main-exposure concentrations were tested (75 %, 50 %, 25 % and 15 % of the MIC). Bacterial growth was measured after drug 2 application across multiple schedules: day 1, 2, 3, 4, 7, 10, 14, 21, and 28; another schedule tested daily sampling until day 6, then days 9 to 18; and a third comparison examined timepoints at days 0, 2, 5, 6, 10, and 14.

2.2.3.2 Sequential treatment and time kill assay

The established sequential treatment followed by a 14-day time kill assay was utilized to evaluate if sub-lethal pre-exposure to one drug induces a treatment-enhancing effect on the second drug, leading to negative cellular hysteresis. The CFU / mL was determined as explained in Chapter 2.2.1.1.

First, a starting culture was made of *M. tuberculosis* strain H37Rv (titre of 6.73×10^7) from frozen stock. To achieve a final concentration of approximately 1×10^5 CFU / mL, 60 μ L of the thawed culture, from frozen stock was directly transferred into a roller bottle containing 200 mL of 7H9 Middlebrook medium supplemented with 0.2 % glycerol, 0.05 % Tween80, and 10 % OADC (Formular 4). The start culture was then incubated standing for 3 days at 37° C.

(4)

$$\frac{\left(\text{Final } \frac{\text{CFU}}{\text{mL}} \text{ to Start} \right) \times (\text{Final volume mL})}{\text{Titer of stock } \left(\frac{\text{CFU}}{\text{mL}} \right)} = \text{mL}$$

The day prior to the experiment, sterility controls of the culture were conducted by inoculating blood agar, Brain Heart Infusion (BHI) medium, and Luria-Bertani (LB) medium. At the initiation of the experiment, OD₆₀₀ was measured. When OD₆₀₀ was about 0.01, then 10 mL of the culture was aliquoted into 50 mL screw cap tubes. Antibiotic stocks were prepared from powder and sterile filtered using a Mi lley® 2 μ L hydrophilic Fluoropore filter unit. A final concentration of 25 % of the MIC of the pre-exposure antibiotic was applied. Bacterial tubes were incubated for 8 h at 37° C, at a 45° angle. After incubation, the bacterial tubes were centrifuged at 3 500 rpm for 10 min at room temperature. The pelleted samples were then resuspended with fresh medium containing or lacking antibiotics for the main-exposure (Figure 2.3).

To ensure quality control for cell loss during the pre-exposure period, CFU / mL was measured at three timepoints: after inoculation with the antibiotic, after 8 h of incubation, and after centrifugation (Figure 2.4).

The “main exposure” was measured as a classic time-kill assay. The initial CFU / mL was measured on day 0, directly after the antibiotic switch and resuspension in fresh medium. Following CFUs were determined on days 1, 2, 3, 5, 10, and 14. The CFU was plated in 10 μ L drop described in Chapter 2.2.1.1. CFU / mL enumeration was assessed on 14, 21, and 28 days after the plating.

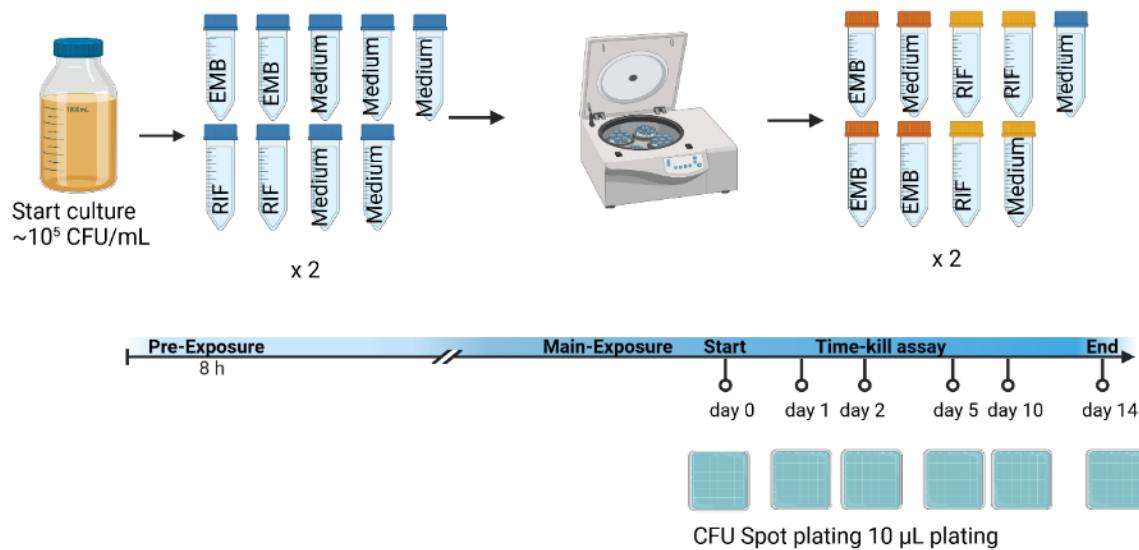


Figure 2.3: Final experimental set up for sequential exposure to two first-line drugs of *M. tuberculosis*. Simplified workflow showing the procedure for one set of biological replicates. The CFU was spotted in triplicates.

The data was visualized using box plots and spaghetti plots utilizing ggplot package in R Studio. Statistics were performed using the multiple comparisons using the Turkey procedure `mctp` function in R using the following command: `comp <- mctp (CFU ~ condition, data = data_set, asy.method = "fisher", type = "Tukey", alternative = "two.sided")`. This test is used to perform a multiple comparison for the means of different groups, specifically comparing CFUs across various conditions. The p-values were calculated using the asymptotic Fisher comparison method, which combines p-values of different tests to produce an overall statistic test. As post-hoc analysis the Tukey's Honest Significant Difference (HSD) method was used, to compare all possible pairs of means while controlling for type I error rate (more conservative than a pairwise t-test). Additionally, it was specified that the alternative hypothesis is two-sided (greater or less than).

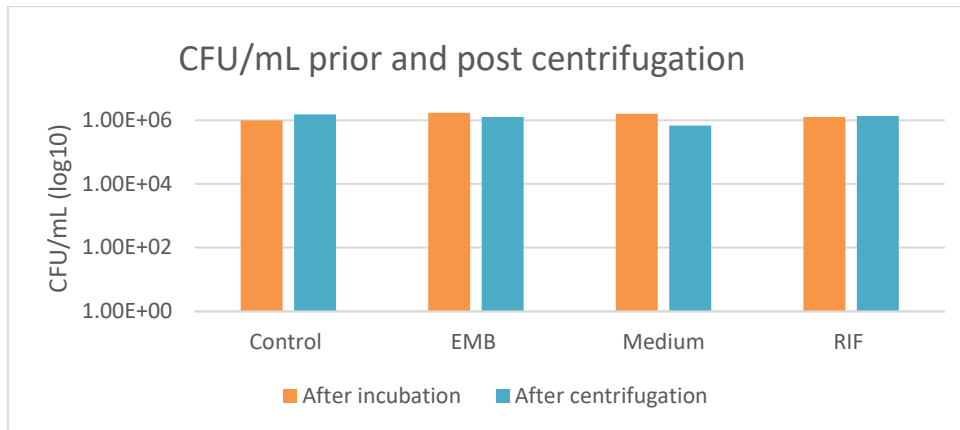


Figure 2.4: Determination of CFU / mL loss by centrifugation. To account for cell loss during centrifugation, samples of the culture were collected both after the 8 h incubation and before centrifugation. Following centrifugation, the cells were resuspended in fresh medium, and these samples were also analysed. N = 3.

2.2.3.3 Transcriptomic analysis of cellular hysteresis inducing drug switch

To assess the transcriptomic landscape and investigate differentially expressed genes (DEGs) during negative (no enhanced efficacy) or positive cellular (enhanced efficacy) hysteresis, RNA sequencing (RNAseq) was employed. Five replicates were performed for each condition, and backups were saved in the - 80° C freezer.

RNA was extracted from the bacteria at the timepoints as follows: TP0, representing only medium directly after aliquoting; TP1, after 8 h of incubation at 37° C with only EMB or RIF, or bacterial culture only in (medium-only) control; TP2, 0.5 h incubation after the main-exposure drug switch (EMB->RIF, RIF->EMB, or Medium->Medium transitions); and TP3, representing 48 h incubation after the main-exposure drug switch (Figure 2.5).

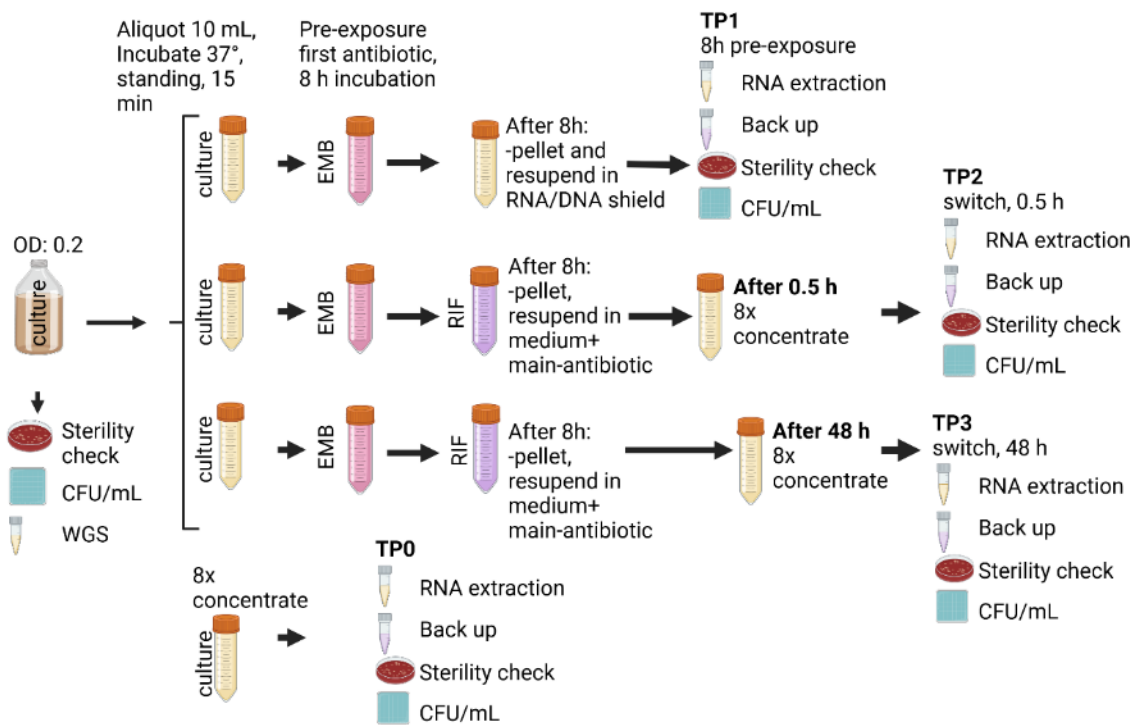


Figure 2.5: Experimental design for RNA sequencing of sequential treatment. Experimental design for condition of ethambutol (EMB) pre-exposure and rifampicin (RIF) main-treatment. When culture was grown to OD₆₀₀ 0.3, 10 mL were aliquoted in 50 mL screw cap tubes, and let sit for 15 min at 37° C before exposure to antibiotics. A medium only control was included in all time-points (TP). TP0 is the culture without antibiotic, no incubation time. TP1 RIF or EMB pre-treatment incubation 8 h, 37° C standing. TP2 after 8h pre-exposure switched to main-treatment incubation for 0.5 h. TP3 after antibiotic switch and 48 h incubation in main-treatment. At every TP, samples were 8x condensed and resuspended in RNA-DNA Shield®. RNA extraction and sequencing was conducted as described in Chapter 2.2.4.1. Additionally, after every TP CFU / mL and sterility controls were obtained. Figure created with BioRender.com.

The sequential treatment experiment was adapted for RNAseq. A starting culture was grown to an OD₆₀₀ = 0.2 in 7H9 Middlebrook medium with 10 % OADC and 0.2 % glycerol. Sterility checks were performed using blood agar plates, as previously described. Following each timepoint, the bacteria were centrifuged for 10 min at 4 000 rpm at 4° C. The supernatant was discarded, and the bacteria were resuspended in 0.6 mL of RNA / DNA Shield®, and topped off with additional 0.6 mL of RNA / DNA Shield®. Samples were stored in the -80° C freezer until extraction steps.

RNA extraction was performed as described in Chapter 2.2.4.5 using the TRIzol® method with cycle time of fastprep (bead beating) adjusted to 40 s, in this single-step method (TRIzol®, spin) the RNeasy Mini Kit (Qiagen) was used for RNA isolation according to the manufacturer's instructions. RNA purification and concentration were carried out using the RNA Clean & Concentrator Kit (Zymo), following manufactures instructions and as described in Chapter 2.2.4.3.

2.2.4 Genomic sequencing and analysis

2.2.4.1 CTAB DNA isolation of *M. tuberculosis*

For safety reasons and to handle bacterial cultures outside the BSL3 facility, bacteria need to be deactivated (killed). First, 5-10 mL of the bacterial culture were transferred to a 15 mL screw-cap tube, followed by centrifugation at 4 000x g for 10 min at room temperature (RT). After centrifugation, the supernatant was carefully discarded, and the pellet was resuspended in 400 µL of sterile distilled water (Braun). The suspension was then transferred to a new 1.5 mL screw-cap tube. The sample was heat killed by placing the tube in a water bath set to 80° C for 20 min. The temperature was closely monitored using a thermometer to ensure a stable temperature of 80° C throughout the process. After the heat-killing, samples were further processed outside of the BSL3 facility.

Subsequently, the cultures were centrifuged at 13 000 relative centrifugal force (rcf) for 3 min at room temperature. The supernatant was carefully removed using a pipette, and the pellet was resuspended in 400 µL of AE Buffer. Following resuspension, 50 µL of 10 mg / mL lysozyme was added to each sample, gently vortexed for 5 s, and then incubated overnight in a water-bath at 37° C. To prevent potential contamination, centrifugation was performed for 3 s to remove any droplets from the lid, prior to proceeding with further steps.

A CTAB / NaCl mixture was prepared and preheated to 65° C. Then, 100 µL of 5M NaCl and 100 µL of the preheated CTAB / NaCl mixture were added to each sample and vortexed until a homogeneous milky mixture was achieved. The samples were then incubated for 10 min at 65° C in a water-bath. Next, 750 µL of Chloroform / Isoamyl alcohol mixture (25:1) was added to each sample, vortexed for 10 s, and subjected to centrifugation at 13 000 rcf for 15 min at room temperature. The resulting aqueous supernatant was carefully transferred to a fresh, labelled 1.5 mL RNase / DNase-free Eppendorf tube. To precipitate nucleic acids, approximately 0.6 volumes (~450 µL) of isopropanol were gently mixed into each sample and incubated at -20° C for 2 h. After incubation, samples were centrifuged at 13 000 rcf for 5 min at room temperature, and the resulting supernatant was discarded. Any remaining supernatant was carefully pipetted off, leaving the nucleic acid pellet. The uncovered samples were fully dried at 60° C for 20 min in a heating block. Subsequently, 40 µL of 10 mM AE Buffer was added to each sample and briefly vortexed or flipped to ensure proper resuspension. The pellet was then diluted over 20 min at 60° C in the heating block to facilitate dissolution. To assess the quality and quantity of the extracted nucleic acids, 1.5 µL of each sample was measured using the DeNovix DS-11FX Spectrophotometer.

2.2.4.2 Whole genome sequencing

Quality control of MTBC wild-type and mutant strains was performed using whole genome sequencing (WGS). DNA was isolated by the CTAB method as described previously and DNA quality and quantity was measured subsequently using NanoDrop (Thermo Fisher Scientific) and Qubit (Thermo Fisher Scientific). The libraries were prepared using Baym2015 (Illumina) (for detailed list of components see 2.1.4 Kits) and Celero EZ (Tecan) kits according to the manufacturer's instruction. Quality of DNA libraries was verified on Fragment Analyzer using the HS NGS Fragment Analysis kit (Aligent) for all samples $< 5 \text{ ng} / \mu\text{L}$ and all samples $> 5 \text{ ng} / \mu\text{L}$ with the SS NGS Kit, according to manufacturer instructions. Samples were sequenced on the NextSeq500, NextSeq1000 or NextSeq2000 then further processed and analysed using the workflow outlined in Chapter 2.2.5.1.

2.2.4.3 Experimental design for RNA sequencing of resistant clones in the presence or absence of their target drug

A bacterial culture from frozen stock was grown in 7H9 Middlebrook medium supplemented with 10 % OADC, 0.5 % glycerol, and 0.025 % Tyloxapol in 30 mL-square bottles at 37° C (Figure 2.6). Turbidity was monitored, and once the culture reached an OD_{600} of 0.15, it was transferred to 500 mL roller bottles and adjusted to a final volume of 100 mL. Subsequently, the culture was incubated, standing at 37° C until reaching an OD_{600} of 0.4 - 0.5. Then, the culture was diluted with 110 mL of fresh medium, which halves the OD_{600} and 10 mL aliquots were prepared in 30 mL square bottles the night before the experiment. The following morning, the treatment samples were exposed to 10x the MIC of BDQ ($1.2 \mu\text{g} / \text{mL}$), or DMSO (the antibiotic solvent). Five replicates were included for each condition. After the incubation, which lasted either 30 min or 4 h, a drop from each culture was spotted onto a blood agar plate as a sterility control. Additionally, 1 mL of each culture was preserved in low-binding Eppendorf tubes at -80° C as a backup. Then, 10 mL of each sample was processed according to the RNA isolation protocol from liquid culture (Chapter 2.2.4.4).

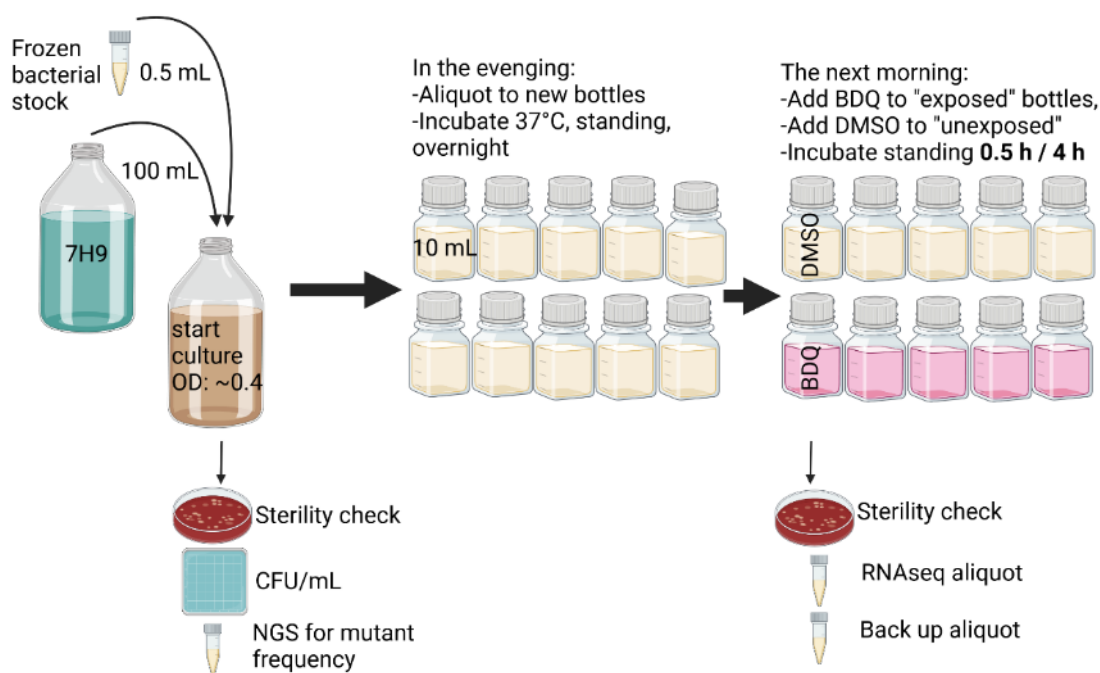


Figure 2.6: Experimental design for RNA sequencing of mutants with and without exposure to their target drug. Culture from frozen stock was grown to OD₆₀₀ 0.4. Sterility and quality controls were conducted of the culture. The evening before the experiment the culture was diluted to OD₆₀₀ = 0.3 and 10 mL was transferred into square bottles. The next day, either 1.2 µg / mL BDQ or DMSO only was added to the culture bottles and incubated for 0.5 h and 4 h. Subsequently, blood agar sterility control was done of every sample. The sample were condensed and RNA extraction was conducted. Created with BioRender.com.

2.2.4.4 Establishing RNA isolation from liquid culture of *M. tuberculosis*

Initially, different RNA extraction protocols were evaluated to achieve a high yield (> 20 ng / µL) of sterile and high-quality RNA (RQN > 4) (Table 2.3). Several sterilization procedures, including chemical treatments and sterile filtration, were evaluated. These methods were tested in collaboration with Silvia Maass, Jana Schönfeld, Larissa Mohr, and Lindsay Sonnenkalb, as detailed in Table 2.3. The protocols included different combinations of reagents and mechanical disruption settings. First, the effectiveness of TRIzol® was assessed using cultures grown in media containing 0.05 % Tween80 or 0.025 % Tyloxapol. Two sets of FastPrep settings were tested: a harsh setting (5 circles of 45 s at 6.5 m / s) and a milder setting (3 circles of 25 s at 4.5 m / s). Then the influence of growth medium on RNA quality (RQN) was assessed, by comparing Tween80 and TRIzol® reagent to Tyloxapol combined with both FastPrep settings (harsh or mild). Simultaneously DNA / RNA Shield™ and RNA protect were tested across all conditions. As the *M. tuberculosis* samples were not sterile after DNA / RNA Shield™ application alone, additional sterilisation parameters were examined. Heat sterilisation for standard (80° C for 20 min) and extended temperature (100° C for 15 min) and also, different sterile filtration methods were tested, using Spin-X tubes and Millipore Ultrafree-MC Filter. Last, the reagent TRIzol® was used

for sterilisation of the *M. tuberculosis*. To stabilize the quality of the RNA, two protocols were compared, a combined protocol utilizing the chloroform precipitation method and TRIzol®. Compared to samples incubation in DNA / RNA Shield™ prior to FastPrep (5 circles of 40 s at 6.5 m / s) for cell disruption, followed by sterilization with TRIzol®. The optimized protocol, combining DNA / RNA Shield™ with the sterilizing capacity of TRIzol®, provided a reliable method for obtaining high-quality (RQN > 8) and sterile RNA suitable for RNAseq.

Table 2.3: Procedures tested for good quality and sterile RNA extraction of *in vitro* cultures.

Procedure	Protocol	Sterility	Yield (ng / μL)	Quality (RQN)
TRIzol®	The culture was pelleted and resuspended in TRIzol®, incubated 5 min room temperature and added to Lysing Matrix B tubes. After FastPrep24 procedure, Lysing Matrix B tubes was spined down and supernatant transferred into fresh Eppendorf tube 1.5 mL (= mechanical cell disruption).	Not sterile	High (> 50)	High (> 7)
RNAprotect	The culture was pelleted and resuspended in RNA protect befor mechanical cell disruption by Fastprep.	Sterile	Medium (< 50)	Low (< 5)
DNA / RNA shield™	The culture was pelleted and resuspended in DNA / RNA shield™ incubated 5 min room temperature, befor mechanical cell disruption by Fastprep.	Sterile	High (> 50)	Low -mid (1 - 6)
Heat kill	The culture was pelleted and resuspended in DNA / RNA shield™ incubated 5 min room temperature, mechanical cell disruption by FastPrep, sample spin down transferred in H ₂ O and heat killed either at 80° C for 20 min or 100° C for 15 min	Sterile	Low (> 30)	Low -mid (1 - 6)
DNA / RNA shield™ + Sterile filtration with Spin-X tubes®	The culture was pelleted and resuspended in DNA / RNA shield™, incubation 5 min room temrea-ture, after mechanical cell disruption by Fastprep. transferred into Spin-X tubes, centrifuge at 500x g for 10 min at 4° C. Transfer supernatants into fresh spin-X tube and centrifuge at 2,000 x g for 10 min at 4° C. Store at -80 ° C.	Sterile	High (> 50)	Low-mid (1 - 6)
DNA / RNA shield™ + Sterile filtration with Millipore Ultrafree-MC Filter	The culture was pelleted and resuspended in DNA / RNA shield™, incubation 5 min room temperature, after mechanical cell disruption by Fastprep. transferred into filter column, centrifuge 4,000 rpm 2 min at 10° C. Repeat filtration with new filter / tube.	Sterile	Medium (< 50)	Low (< 5)
DNA / RNA shield™ + TRIzol®	The culture was pelleted and resuspended in DNA / RNA shield™, incubated 5 min room temperature, after mechanical cell disruption by Fastprep. transferred into 2 mL of TRIzol®, incubated 5 min room temperature.	Sterile	Medium (< 50)	High (> 7)

DNA / RNA shield™ + TRIZol® half volume	The culture was pelleted and resuspended in DNA / RNA shield™, after mechanical cell disruption by Fastprep. transferred into 1 mL of TRIZol®, incubated 5 min room temperature.	Sterile	Medium (< 50)	High (> 7)
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All RNA samples underwent sequencing at the Research Center Borstel, Molecular and experimental mycobacteriology department. Final RNA isolation of samples was conducted using a combination of the single-step method with TRIZol® and the RNeasy mini kit (Qiagen), following the manufacturer's instructions. To ensure high-quality RNA extraction, bacterial cultures of mid or exponential growth phase were mixed with 600 µL of DNA / RNA shield™ (Zymo) per 10 mL of culture. Adjustments were made for higher bacterial loads, with 1.2 mL of DNA / RNA shield™ per 1×10^7 bacterial cells, according to manufacturer's recommendation.

Mechanical cell disruption and lysis followed palletisation at 3,500 rpm centrifugation for 20 min at 4° C, subsequently the pellet was resuspended in 600 µL of RNA / DNA shield incubated 5 min at room temperature and transferred to Lysing Matrix B tubes containing 0.1 mm silica beads. Followed by mechanical disruption, as previously described. The rotor was paused for 5 min between cycles to allow cooling, during this time the samples were placed on ice. Beads were then separated from the lysate by centrifugation at 12 000 x g for 15 min, and the supernatant was transferred to new RNase-free tubes.

For inactivating the bacteria, 1 mL of TRIZol® was added to the samples and incubated for 5 min at room temperature. Outside the BSL3 facility, RNA precipitation was achieved by adding one volume of 70 % ethanol to the sample under a fume hood. The RNeasy mini kit (Qiagen) was employed for RNA purification following manufacture instructions with modifications: on-column digestion was omitted, and an additional centrifugation step in a fresh collection tube at full speed was included. RNA was eluted with 50 µL of RNase-free water and stored at $\leq -70^\circ$ C.

Quality and RNA concentration were initially checked using the Nanodrop (Thermo Fisher Scientific) before the RNA digestion step, followed by verification with the Qubit (Thermo Fisher Scientific) later in the protocol. DNase I from the RNase-free DNase set (Qiagen) was utilized for DNA digestion according to the manufacturer's instructions and the concentrations for the reaction in Table 2.4, by gentle inversion and incubation at room temperature for 10 min.

The RNA Clean & Concentrator™ Kit (Zymo) was employed for purification and concentration following the manufacturer's protocol. Aliquots (2 µL each) were taken for Fragment Analyzer analysis RNA quality control and PCR to verify effective DNA digestion. Control PCR was performed using Primer-pair

#415 (rrs F (+1202), rrs R (+1565)) and the master mix for the reaction according to Table 2.5. Visualization of PCR products on a 1.5 % agarose gel confirmed absence of DNA contamination. RNA quantity was measured using the Qubit® 2.0 Fluorometer. Qubit® Working Solution was prepared by diluting Qubit® RNA Reagent 1:200 in Qubit® RNA Buffer. After calibration with standards, samples were measured for RNA concentration. Additionally, samples were subjected to quality assessment using the Fragment Analyzer.

Table 2.4: Reaction set up for DNA-free RNA reaction with DNase I.

Sample (volume adjusted with water)	50 µL
DNase I	2.5 µL
Buffer RDD	10 µL
Water	adjust to 100 µL

Table 2.5: Master mix for PCR to control RNA digestion.

Component	Master mix [µL]
10 x PCR buffer (with MgCl ₂)	2.5
MgCl ₂	-
dNTPs	0.5
Primer F (6,25 µM)	0.5
Primer R (6,25 µM)	0.5
DMSO	-
H ₂ O	18.875
Taq	0.125
DNA (1:1000 to 1:80 000)	2

2.2.5 Computational Analysis

2.2.5.1 WGS processing and bioinformatic analysis

DNA was extracted from both the MTBC wild-type strain H37Rv (ATCC27294) and the *Rv0678* mutant strains (inversion, A99V, ins274A) to confirm the wild-type sequence and the presence and frequency of specific mutations. DNA isolation was conducted using CTAB method as described in Chapter 2.2.4.1. The extracted DNA samples were then sequenced at Research Center Borstel. Library preparation followed the protocol outlined in Chapter 2.2.4.2 and sequencing was performed using the Illumina NextSeq500 / 1000 or 2000 machines (Illumina, San Diego, CA, USA). The resulting WGS data was analysed using the MTBseq pipeline v1.0.3 for sample identification and resistance profiling (142). Reads were mapped to *M. tuberculosis* H37Rv reference genome (GenBank ID:NC_000962.3) using Burrows-Wheeler Aligner (BWA)-MEM algorithm, with SAMtools employed for mapping procedure (123,146). Further refinement of the alignment file was performed using the Genome Analysis Toolkit (GATK) and SAMtools (147). Variant calling was performed with mpileup tool from SAMtools to obtain coverage information at base-pair level applying coverage and base quality thresholds. Variants including single

nucleotide polymorphisms (SNPs) and insertions / deletions (InDels), were identified with criteria requiring two reads mapped in both forward and reverse direction. A detailed report was generated for each sample, annotating the observed SNPs and InDels along with metadata, and link it to antibiotic resistance

2.2.5.2 RNA sequencing and bioinformatic data analysis

RNA samples obtained from *in vitro* experiments were sequenced at Research Center Borstel, following the experimental workflow outlined in Figure 2.7. RNA was extracted according to the RNA extraction protocol described in Chapter 2.2.4.4, with RNA integrity assessed using Fragment Analyzer and the HS RNA Kit (Agilent). Only samples with an RNA Quality Number (RQN) of 5 or higher were selected for sequencing (Appendix A Table 0.1).

RNAseq libraries were constructed using the Zymo-Seq Ribo free Total RNA Library Kit v1.3.0, used for the initial experiment and v2.0.0 for the main experiment. The quality of NGS libraries was verified using a Fragment Analyzer with HS / SS NGS Fragment kits (Agilent), according to manufacturer's instructions. Sequencing was conducted on the NextSeq500 / 1000 / 2000 (Illumina) platforms according to the manufacturer's instructions.

For raw FASTQ data analysis, custom scripts were used. Reads were mapped to the *M. tuberculosis* H37Rv reference (NCBI accession: NC_000962.3) using bowtie2 v2.3.5.1 and the resulting alignments were processed with sambamba v0.6.1. Mapping statistics were collected with samtools flagstat v1.9, and the number of uniquely mapped reads was determined using RSeQC v4.0.0 'bam_stat.py'. Mapped reads overlapping with gene features (GTF file of NC_000962.3) were quantified with featureCounts v1.5.0, while read pairs were only counted once (Appendix A Figure 0.7). Quality controls such as gene body coverage, and coding region coverage were integrated into the workflow, using RSeQC (Appendix Figure 0.9). Samples with less than 20 % rRNA content, as determined from the featureCounts table, were selected for further analysis (Appendix A Table 0.2, Table 0.3). Additional quality metrics, including sequencing yield above 8 million reads, homogeneous gene body coverage, and mapping efficiency above 80 %, were monitored throughout the experiments (Appendix A Figure 0.6).

Differential expression analysis of genes (DEGs) was performed using the DESeq2 package v1.42.0 in RStudio v23.09.1+494 using R v4.3.2 (148). Genes with at least 30 reads in all samples were included in downstream analysis. Statistical significance was determined by an adjusted p-value (*p*_{adj}) ≤ 0.05 and a log₂ fold change (log₂FC) ≥ 1 or ≤ -1. For comparative analysis, Venn diagrams were generated using the online tool Venny 2.1 (149).

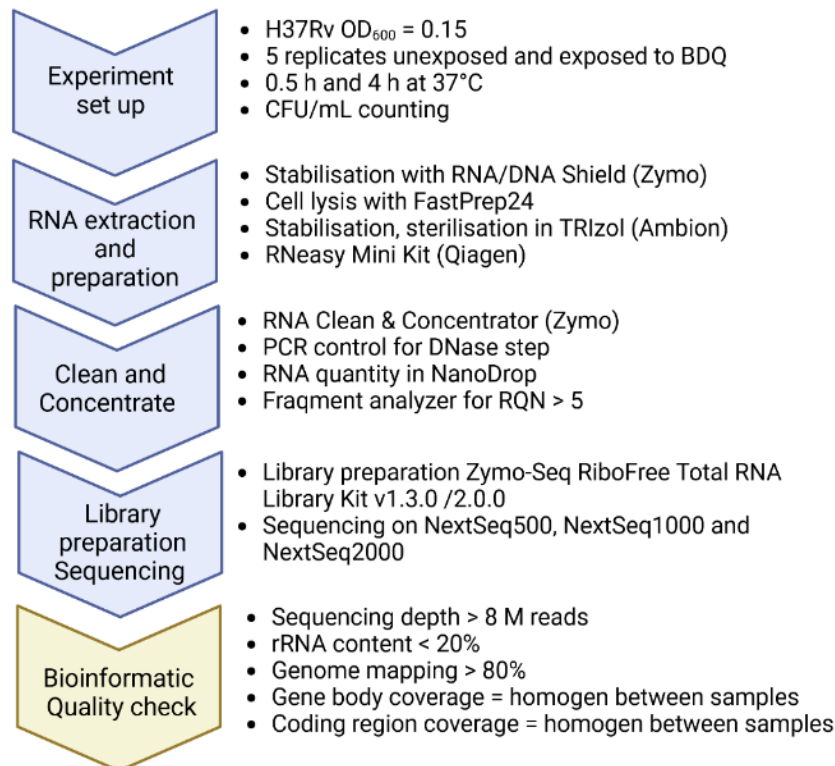


Figure 2.7: RNA sequencing workflow and quality controls. From wet lab to in silico analysis including different quality controls throughout the sample processing. Created with BioRender.com

Differentially gene expression data interpretation

A short excursion on the interpretation of gene expression levels. In this thesis, gene expression will be visualized mainly using volcano plots or tables, indicating $\log_2$ of the fold change ($\log_2FC$), which reflects the ratio of expression between two conditions, specifically expressed as (expression condition 1) / (expression condition 2) (150). To convert $\log_2FC$ values to fold change (FC) values, the calculation is performed as follows: $FC = 2^{\log_2FC \text{ value}}$ (151,152). For example, if $\log_2FC = 1$ (which equals 2^1), this indicates that the expression in condition A is twice as high as in condition B so the $FC = 2$ (152).

2.2.5.3 GO enrichment and KEGG pathway analysis

To investigate the biological functions involved in MTBC with respect to drug exposure, a list of the up- or downregulated DEGs was imported into the DAVID platform (<https://david.ncifcrf.gov>) and analysed with default settings for Gene Ontology (GO) and biological pathways via Kyoto Encyclopaedia of Genes and Genomes (KEGG) (153–156). GO analysis was utilized to screen biological processes (BP), cellular components (CC), and molecular functions (MF). Additionally, KEGG enrichment analysis was employed to identify crucial signalling pathways involved in the biological processes.

3 Results

3.1 Transcriptomics of bedaquiline resistant *Mycobacterium tuberculosis* clones

To combat antibiotic resistance in *M. tuberculosis* and protect new and existing regimes, it is crucial to investigate the mechanisms behind resistance evolution (157). BDQ, a recently introduced drug for TB therapy, is essential in treatment regimens against MDR-TB (158,159). However, MTBC strains rapidly developed resistance to BDQ through mechanisms such as mutations in its target, *atpE*, and the *Rv0678* gene (160). This thesis focuses on *Rv0678*, a transcriptional regulator also regulating the MmpS5-MmpL5 efflux pump system, which is responsible for nearly all BDQ resistances in clinical MTBC strains (65,117,118,161–163). *Rv0678* binds the promoter region of the *mmpS5-mmpL5* operon, blocking transcription of the efflux pump system (117,118). Mutations in *Rv0678* result in a malfunctional regulator, upregulating efflux pump genes and resulting in low level resistance to BDQ (118). To elucidate the spectrum of resistance mechanisms in *M. tuberculosis* clones with mutations in *Rv0678*, RNAseq technology was used to analyse resistance phenotypes, comparing the wild-type H37Rv clone of *de novo* lab generated mutants with an *atpE* mutation or *Rv0678* mutation. The transcriptome was further evaluated after exposure to BDQ or incubation without BDQ for 0.5 h and 4 h, assessing dynamic changes over time, as detailed in Chapter 2.2.4.3.

For this project, the RNAseq workflow was established mainly by S. Maas, J. Schönfeld and L. Mohr, testing different parameters to obtain a good yield of sterile (removal of live bacteria), high-quality RNA (Chapter 2.2.4.4, Table 3.1). The RNA yield was checked using Qubit, an acceptable yield for library preparation was reached with > 12 ng / μ L. High-quality RNA refers to the integrity of the RNA, this was checked with a fragment analyser (Advanced Analytica). The fragment analyser gives an RQN based on electrophoresis with a score between 1 and 10, 1 is degraded and 10 of the highest quality. Only samples with an RQN > 4 were further processed for Illumina sequencing. Extensive tests were conducted comparing different RNA preparation systems using RNeasy Protect (Qiagen), DNA / RNA Shield™ (Zymo Research), or TRIzol® (Ambion), with different growth medium components (Tween⁸⁰ / Tyloxapol) and FastPrep™-24 conditions (Table 3.1). The use of DNA / RNA Shield™ resulted in an overall high-quality RNA (RQN > 8). However, previous work in this lab found that despite manufacturer claims, the use of DNA / RNA Shield™ buffer did not sterilize *M. tuberculosis* samples. Thus, DNA / RNA Shield™ treated samples were tested with heat kill inactivation, which resulted in very low RQN (RQN < 5). Given the superior performance in preserving RNA integrity, DNA / RNA Shield™ was subject to sterile filtration using two systems. Despite both systems yielded high-quality RNA (RQN > 7), the RNA yield remained below 10 ng / μ L. The use of TRIzol® sterilized the samples but RNA quality remained

low (RQN < 5). Finally, the combination of DNA / RNA Shield™ and TRIzol® was tested, which resulted in a good RNA yield (> 12 ng / μ L) and high-quality RNA (RQN > 7) (Table 3.1, Appendix A Table 0.1).

Table 3.1: Test parameters for good yield and high-quality RNA.

Procedure	Sterility	Yield (ng / μ L)	Quality (RQN)
RNAprotect	Sterile	High (> 100)	Low -mid (1 - 6)
DNA / RNA Shield™	Not sterile	High (> 100)	High (> 7)
TRIzol®	Sterile	High (> 100)	Low-mid (1 - 6)
DNA / RNA Shield™ + Heat kill	Sterile	Medium (< 50)	Low (< 5)
DNA / RNA Shield™ + Sterile filtration with Spin-X tubes®	Sterile	Very low (< 5)	High (> 7)
DNA / RNA Shield™ + Sterile filtration with Millipore Ultrafree-MC Filter	Sterile	Very low (< 10)	High (> 7)
DNA / RNA Shield™ + TRIzol® (2 mL)	Sterile	Mid-high (> 80)	High (> 7)
DNA / RNA Shield™ + TRIzol® (1 mL)	Sterile	Mid-high (> 80)	High (> 7)

Next, the experimental set-up was designed in which *M. tuberculosis* BDQ resistant mutants and the ancestral wild-type clones were exposed to BDQ for 0.5 h (Chapter 2.2.4.3). This protocol was tested using two mutants, one with an IS6110 insertion in *Rv0678* (total loss-of-function mutation) and one with the *atpE* A63P variant, alongside the wild-type H37Rv ancestor clone. Four biological replicates were performed for each experimental condition. The established RNA extraction protocol for this experiment resulted in moderate-to-high RNA concentrations (> 20 ng / μ L), as the RQN varied from 4 - 9, and high-quality library preparation as the non-rRNA reached > 70 % for almost all samples (Figure 3.1, Appendix A Table 0.2 Figure 0.3). However, after Fragment Analyzer analysis, one sample was excluded due to RQN below 3 and two more samples were excluded after sequencing as due to non-rRNA < 70 % (an indication of sub-optimal library preparation).

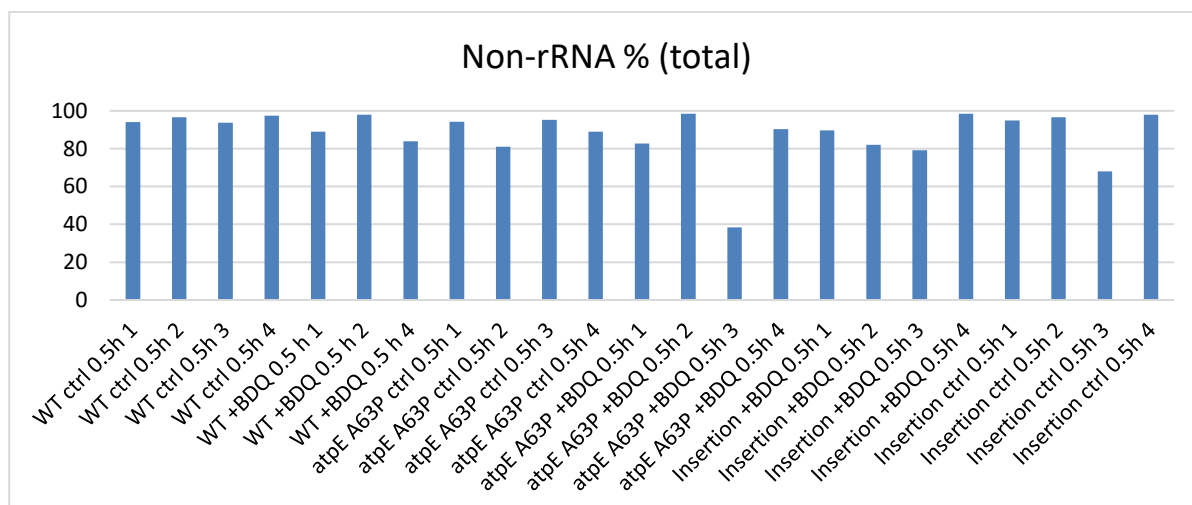


Figure 3.1: Quality check observing non-rRNA % content of total reads. Quality control of the library preparation, the y- axis depicts percentage of the total reads and x-axis the different replicates.

To assess the quality after Illumina™ sequencing, rRNA quantity was assessed using a combination of quality control analysis utilizing “Feature Counts” to check the uniquely mapped reads, “Bowtie2” for analysis of alignment scores and “RSeQC” to assess the gene body coverage of read coverage over gene bodies, visualized MultiQC v1.13. dev0 (Chapter 2.2.5.2, Appendix A Figure 0.1, Figure 0.2, Figure 0.4). To get a first insight into the relationship between gene expression profiles of samples and initial group assessment, a Principal Component Analysis (PCA) was conducted (Figure 3.2). This analysis was used to determine variability between the different experimental conditions and between technical or biological replicates (inter- and intragroup variability) and can be utilized for outliers identification (164). The principal component 1 (PC1) explains 76 % of the variance in the dataset and PC2 adding 11 %, which was sufficient for preliminary interpretation of the data. The four technical replicates of one condition, were split on the PC2 plane (y-axis), forming two separate groups related to a minor batch effect. This sequencing variability resulted as the samples were analysed on two different sequencing runs. Notably, the replicates of all samples on one run grouped together, so intragroup variability was low among unexposed condition, and increased between samples exposed to BDQ. While all tested clones group together when not exposed to BDQ, the *Rv0678* mutant and the wild-type samples exposed to BDQ show a clear shift compared to non-exposed, but are clearly separated of each other. The *atpE* mutant groups with the unexposed samples, regardless of BDQ exposure. These results indicated a strong BDQ effect on the transcriptome of only the *Rv0678* mutant and wild-type ancestor.

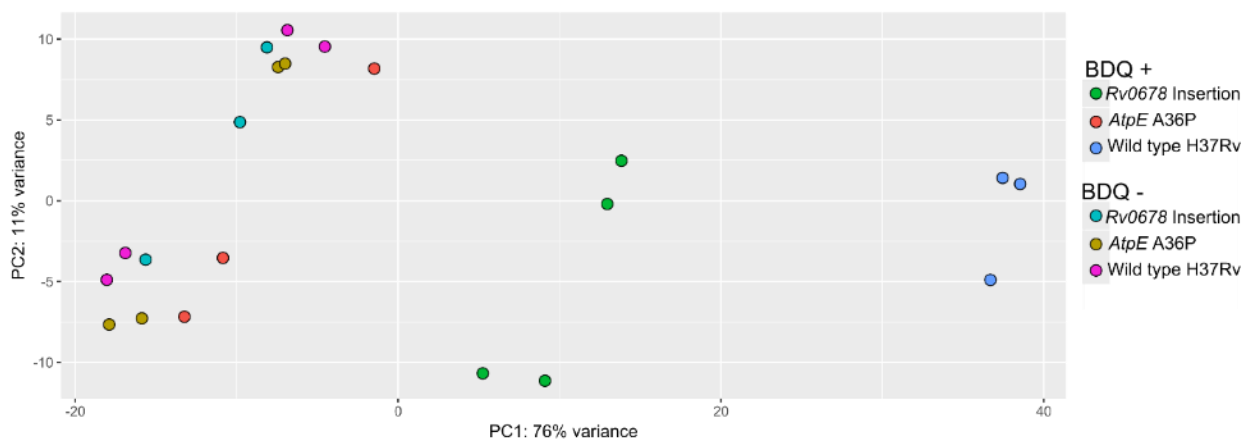


Figure 3.2: PCA plot of resistant and wild-type clones with or without exposure to bedaquiline. RNA sequencing of the first *in vitro* experiment on bedaquiline (BDQ) resistant mutants and the ancestral wild-type. Each dot represents a technical replicate coloured according to bacterial clone and BDQ exposure. The intragroup variability is related to sequencing variability of two separated sequencing runs, as two of four replicates were on separate runs. PC = Principal Component, BDQ+ = with BDQ exposure, BDQ- = without BDQ exposure, n= 1.

Next, a heatmap with hierarchical clustering of the 20 most differentially expressed genes was performed to assess data similarity and dissimilarity (Figure 3.3). Clustering means that the distance between the samples of the count matrix was assessed, this showed the counts for a specific gene which are similar, then clusters were generated according to this similarity. This observation reflected the observations of the PCA plot, where clones exposed to BDQ group together but separated from each other and not exposed clones grouped together, except the *atpE* A63P mutant which shows less differential expression in the *mmpS5-mmpL5* genes regardless of the presence or absence of BDQ. Investigations of the efflux pump genes *mmpS5-mmpL5* (*Rv0677c*, *Rv0676c*), which were described to be key players in the resistance mechanism of *Rv0678* mutants, were up-regulated under BDQ exposure and unexposed in the *Rv0678* mutant. Interestingly, the ancestral wild-type also exhibited an upregulation of the efflux pump genes under BDQ exposure but not without. This experiment proved that the experimental set-up and 0.5 h exposure time worked as expected. Therefore, a follow-up experiment was designed.

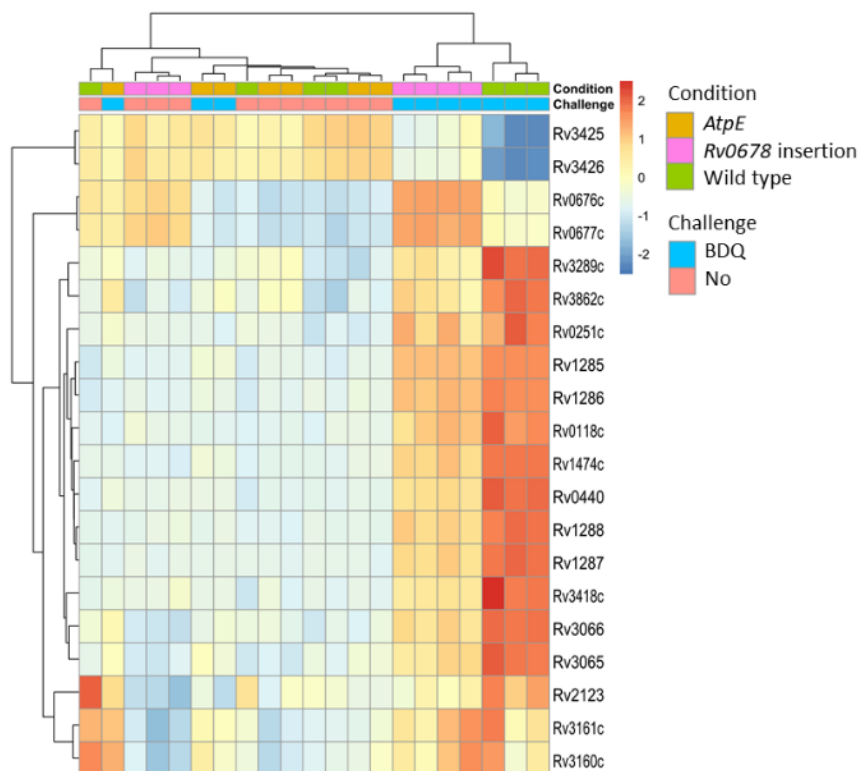


Figure 3.3: Heatmap of the 20 most variable expressed genes with and without bedaquiline exposure for 0.5 h. This heatmap was generated from the count matrix of four replicates per experimental conditions, except the wild-type (green) exposed to BDQ which had three replicates. Clones with a mutation in *atpE* (yellow), *Rv0678* (pink) and the wild-type ancestor (green) were included. The challenge is indicated on top of the heatmap with blue for 0.5 h BDQ exposure and with red for no BDQ exposure. The colour scale indicates the expression of a single gene varying over the samples, with up-regulation depicted in red and down-regulation in blue. One sample was excluded prior to sequencing due to low RQN, two samples due to non-rRNA % < 70 %, replicates = 4, n = 1.

The next experiment was designed to address several questions: What is the expression profile of the *mmpS5-mmpL5* efflux pump system in *Rv0678* mutants, both with and without exposure to BDQ? Is there a shared gene expression profile among *Rv0678* mutants linked to resistance mechanisms other than the *mmpS5-mmpL5* efflux pump system? Can an analysis of 0.5 h and 4 h transcriptional response gives valuable insights into the resistance mechanisms of *Rv0678* mutants? This second experiment which will further be referred to as the “main experiment”, included additional *Rv0678* mutants harbouring clinically relevant resistance mutations and a second exposure timepoint of 4 h to capture later response to BDQ (Figure 3.4).

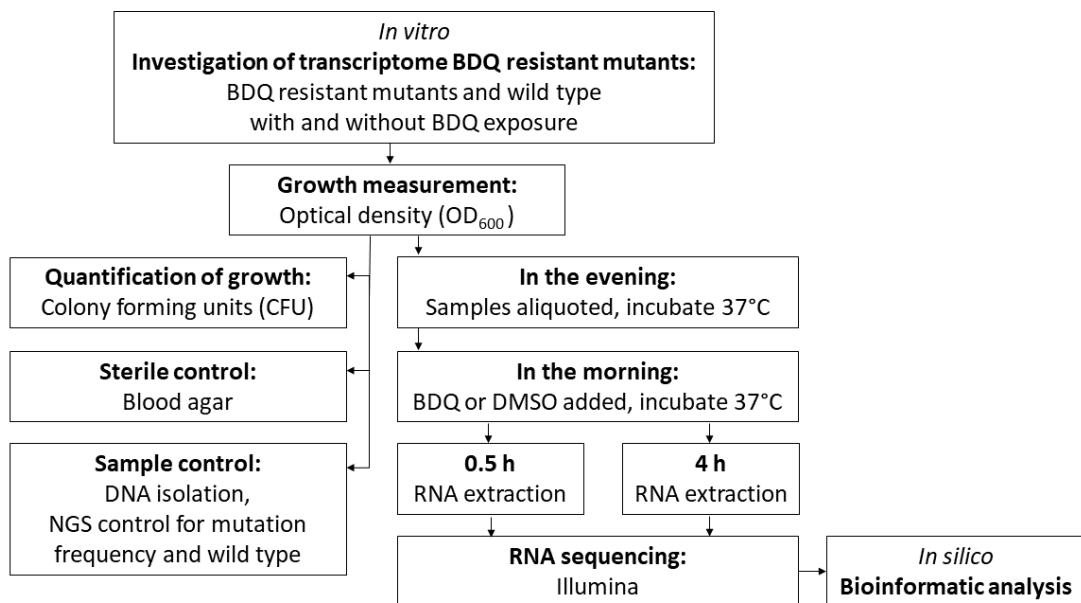


Figure 3.4: Experimental procedure of main experiment for transcriptomic investigation of bedaquiline resistant mutants exposed to bedaquiline or not. Including *M. tuberculosis* clones harbouring *Rv0678* resistance-associated mutations: Inversion, A99V or 274insA. A clone with the mutation in *atpE* A63P and the ancestral wild-type H37Rv were included as controls. The start culture from frozen stock aliquoted at exponential phase in the evening before starting the experiment, on the next morning the bacteria were exposed to 10x the MIC bedaquiline (BDQ) or DMSO (the antibiotic solvent) and incubated for 0.5 h or 4 h at 37° C. RNA was extracted after incubation and all samples with four replicates were sequenced on one sequencing run.

For the main experiment, three clones with different *Rv0678* mutations: inversion, A99V and 274insA, one *atpE* A63P mutant and the wild-type, were exposed to 1.2 µg / mL BDQ (10x MIC) or left untreated (DMSO only). High quality RNA was extracted from all samples with an RQN > 8. The main experiment was analysed separately for each timepoint to assess transcriptional responses after 0.5 h and 4 h BDQ exposure. Each PCA plot explains about 80 % variance of the data. The identified groups correspond to samples exposed to BDQ or not exposed as the PC1 along the x-axis explains 73 % of variance at 0.5 h and 88 % at 4 h. The *atpE* mutants grouped together with the untreated samples, indicating not much variation in the gene expression profile between them (Figure 3.5).

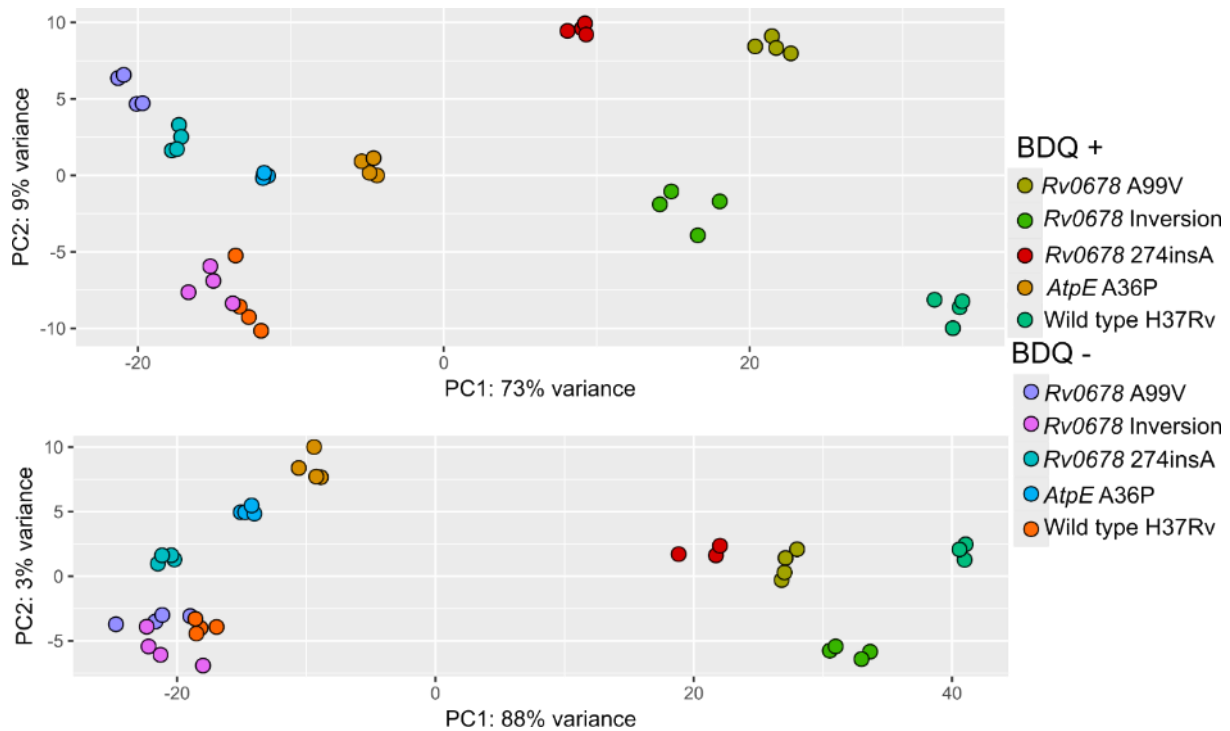
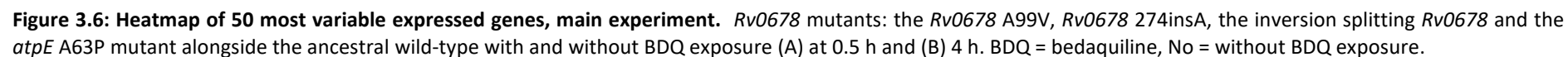


Figure 3.5: Main experiment Principal Component Analysis (PCA) plot. PCA Plot of the wild-type and mutant clones. Top plot: 0.5 h and bottom plot: 4 h after BDQ or no drug exposure. Each dot represents a technical replicate coloured according to bacterial clone and BDQ exposure. The x-axis stands for the PC1 and the y-axis for the PC2. PC = Principal Component, n = 1.

Within the BDQ exposed samples, the ancestral wild-type H37Rv grouped furthest away and similarly in both timepoints. Moreover, the intergroup variability among BDQ exposed samples was low. In unexposed conditions, at the 0.5 h timepoint, the ancestral wild-type and *Rv0678* inversion sample grouped together, while the two other *Rv0678* mutants with the mutations 274insA and A99V grouped distant (Figure 3.5). Overall, the PCA plot and further quality controls, including homogeneous gene body coverage, consistent high mapping quality, with over 80 % of reads uniquely mapped across almost all samples, and a library strandedness below 10 %, generated meaningful results (Appendix A Table 0.2). Two samples were excluded from downstream analysis due to inhomogeneous gene body coverage; sequencing depth < 8 M reads and non-rRNA < 70 % (Appendix A Table 0.2, Table 0.3). After quality control assessment, further analysis focused on investigating the expression pattern of *mmpS5-mmpL5* efflux pump system in *Rv0678* mutants with and without BDQ exposure. A heatmap of the 50 most variable genes, with hierarchical clustering, supported the PCA plot observation as clones exposed to BDQ were clearly separated in the heatmap from unexposed, whereas the *atpE* mutant grouped with the unexposed samples, and all replicates group together as well (Figure 3.6). The investigation of different exposure timepoints as 0.5 h (Figure 3.6A) and after 4 h of BDQ exposure (Figure 3.6B), revealed different gene expression profile in the 50 most variable expressed genes. Notably, the ESX conserved component *eccD1* an ESX-1 type VII secretion system protein (*Rv3877*), was upregulated

solely in the *atpE* A63P mutant after 0.5 h regardless of BDQ exposure. The quality control validated the RNAseq protocol and run. Key results are detailed in the following sections.



3.1.1 Efflux pump genes *mmpS5-mmpL5* expression with and without bedaquiline exposure of *Rv0678* mutants associated with bedaquiline resistance

This section aims to answer the question: What is the expression profile of the *mmpS5-mmpL5* efflux pump system in *Rv0678* mutants, both with and without exposure to BDQ? Therefore, the data validated in the previous section was analysed in depth.

The specific impact of different mutation types on the expression of this efflux pump remains unclear, and whether these mutations consistently drive efflux pump expression in the mutants. Upregulation of the *MmpS5-MmpL5* efflux pump system has been identified as a resistance mechanism in BDQ-resistant clones harbouring *Rv0678* mutations (117,118,162,165,166). However, it is hypothesized that loss-of-function mutations in *Rv0678* disrupt its DNA-binding ability in the *Rv0678* regulator. For example, a frameshift mutation could impair *Rv0678* transcription regulator function, whereas an amino acid polymorphism might result in only a partially functional protein (117,118,165,167). To test the hypothesis that efflux pump expression is upregulated in *Rv0678* mutants, regardless of BDQ exposure, the established RNAseq protocol was employed. The *mmpS5* and *mmpL5* and efflux pump genes and their regulator *Rv0678* were investigated after 4 h unexposed or exposed with high dose BDQ (1.2 µg / mL, i.e., 10x MIC).

This analysis is performed on the previously mentioned four BDQ resistant clones with one of each mutation: the missense mutation *Rv0678* A99V, the frame shift mutation *Rv0678* 274insA, an inversion (genome rearrangement) splitting *Rv0678* and the missense mutation in *atpE* A63P (Figure 3.6). Each mutant was compared to the ancestral wild-type H37Rv without BDQ exposure (Figure 3.7A). The efflux gene expression levels for the *Rv0678* A99V mutant were $\log_2FC$ of 0.86 ($padj = 8.64E-26$) for *mmpL5* and $\log_2FC$ of 1.04 ($padj = 2.12E-14$) for *mmpS5* (Figure 3.7B). The *Rv0678* inversion mutant had a $\log_2FC$ of 0.64 ($padj = 5.58E-15$) for *mmpL5* and $\log_2FC$ of 0.61 ($padj = 1.06E-05$) for *mmpS5*, similar to the frame shift mutant *Rv0678* 274insA with a $\log_2FC$ of 0.62 ($padj = 2.85E-14$) for *mmpL5* and $\log_2FC$ 0.69 ($padj = 2.18E-07$) for *mmpS5*. The *atpE* A63P harbouring mutant had a $\log_2FC$ of 0.30 ($padj = 3.54E-04$) for *mmpL5* and $\log_2FC = 0.44$ ($padj = 1.65E-03$) for *mmpS5*. This comparison revealed moderate (about one $\log_2FC$) upregulation of the efflux pump genes across the *Rv0678* mutants compared to the wild-type H37Rv (Figure 3.7). Notably, the missense mutant *Rv0678* A99V exhibited a significant increase in *mmpS5* and *mmpL5* expression levels, when compared to both clones harbouring a frameshift mutation in *Rv0678* (274insA and the inversion) at baseline level (Appendix A Table 0.4).

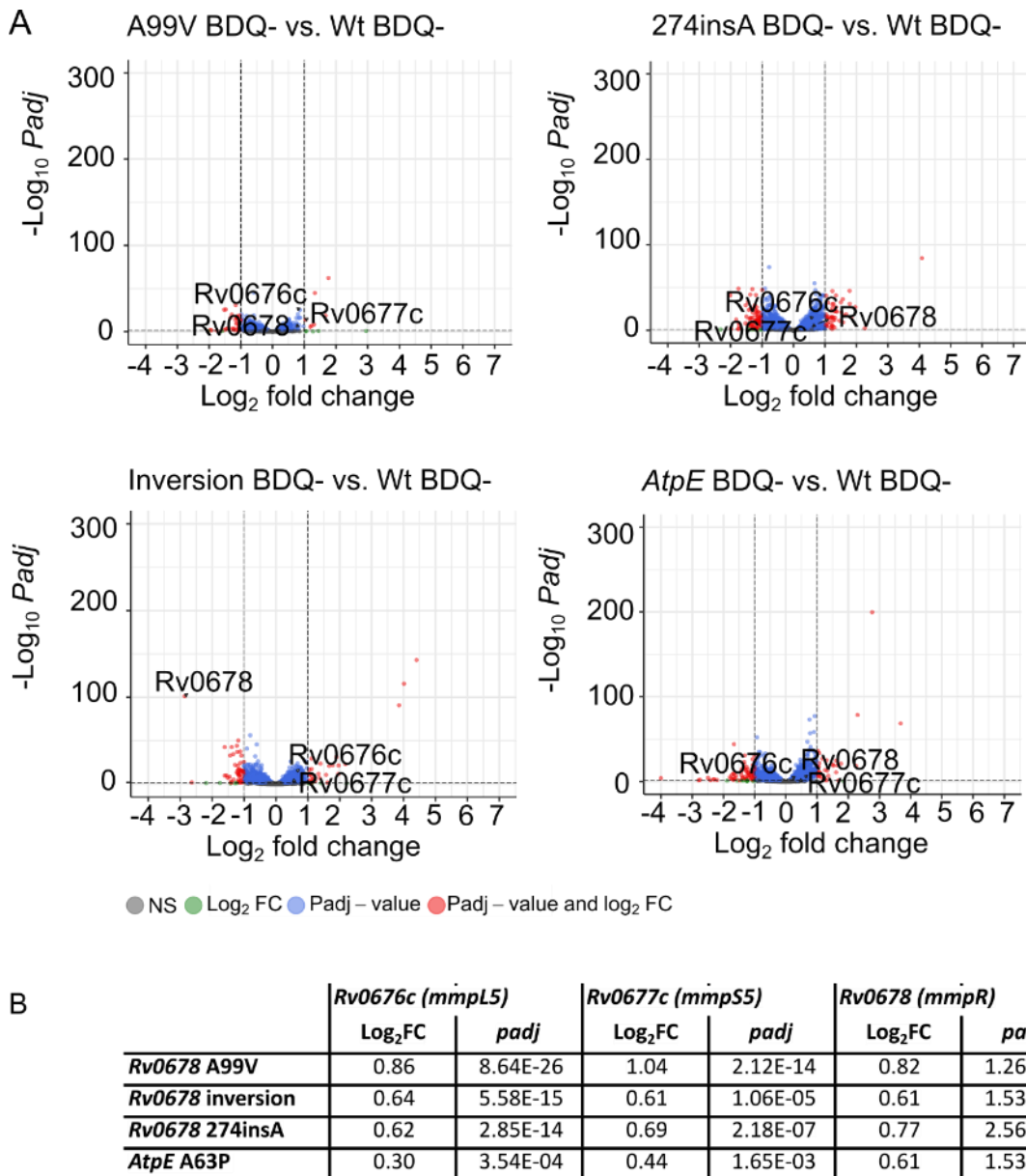


Figure 3.7: Gene expression profile of *Rv0678* mutants and *atpE* mutants. Mutants with a *Rv0678* mutation (A99V, 274insA and inversion) or an *atpE* mutation (A63P) compared to the H37Rv wild-type (Wt) in untreated conditions. *Rv0676c* = *mmpL5*, *Rv0677c* = *mmpS5*, *Rv0678* = *mmpR*. (A) Volcano plots indicating differentially expressed genes, statistically significantly differentially expressed ($p < 0.05$) shown in red, NS = not significant in grey. (B) Table comparing the log₂FC and *padj* of the *Rv0678* (*mmpR*), and efflux pump genes (*mmpS5*-*mmpL5*). BDQ- = BDQ unexposed, FC = fold change, *padj* = adjusted p-value, n = 1, replicates = 4.

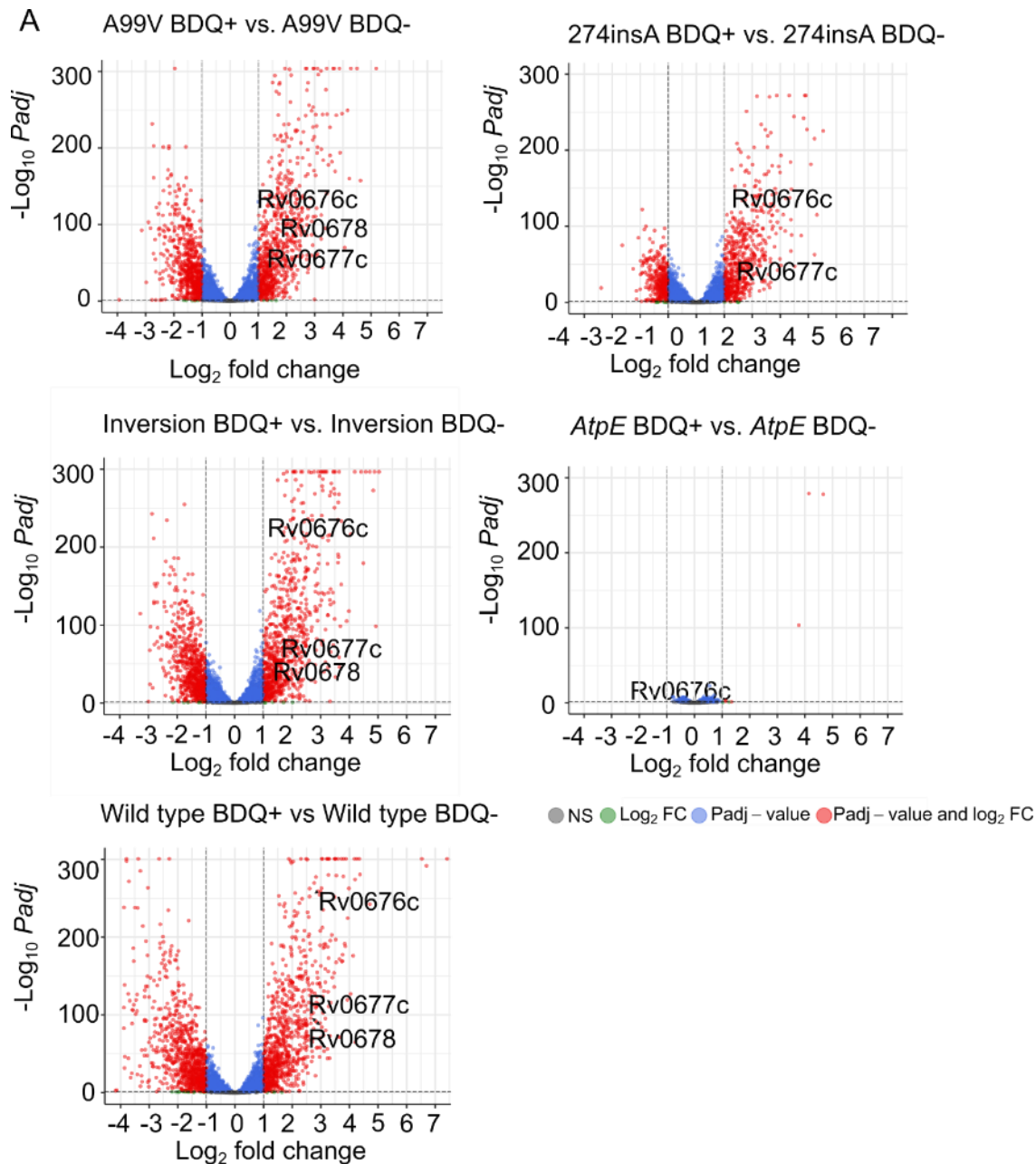
Then, clones with different *Rv0678* mutations were compared to the ancestral wild-type H37Rv under BDQ exposure (Table 3.2, Appendix A Table 0.4). The gene expression levels in the *Rv0678* A99V mutant for *mmpL5* were decreased by a log₂FC of -0.06 and low for *mmpS5* with a log₂FC of 0.19, the *Rv0678* inversion mutant exhibited also a moderate log₂FC of 0.30 for *mmpL5* and a decreased log₂FC of -0.10 for *mmpS5*, whereas the *Rv0678* 274insA had a decreased expression of both genes with a log₂FC of -0.23 for *mmpL5* and log₂FC -0.29 for *mmpS5* and about 4-fold lower expression of efflux pump genes in the *atpE* A63P mutant vs. the wild-type with a log₂FC of -2.53 for *mmpL5* and log₂FC =

-2.11 for *mmpS5*. The expression level of the *mmpS5-mmpL5* efflux pump genes was low in *Rv0678* mutants compared to wild-type under BDQ exposure indicating no difference in efflux pump expression between the mutant and wild-type under BDQ exposure.

Table 3.2: Efflux pump gene expression in *Rv0678* mutants and *atpE* mutants vs. wild-type H37Rv, bedaquiline exposed 4 h. Mutants with an *Rv0678* mutation (A99V, 274insA and Inversion) or an *atpE* mutation (A63P) compared to the H37Rv wild-type (Wt) under BDQ exposure. *Rv0676c* = *mmpL5*, *Rv0677c* = *mmpS5*, *Rv0678* = *mmpR5*. Statistically significantly expressed ($p < 0.05$) Table comparing the $\log_2FC$ and *padj*. BDQ- = BDQ unexposed, FC = fold change, *padj* = adjusted p-value, n = 1, replicates = 4.

<i>Rv0678</i> mutants BDQ+ vs. WT BDQ+						
	<i>Rv0676c (mmpL5)</i>		<i>Rv0677c (mmpS5)</i>		<i>Rv0678 (mmpR)</i>	
	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>
<i>Rv0678</i> A99V	-0.06	5.71E-01	0.19	2.54E-01	0.43	6.37E-03
<i>Rv0678</i> inversion	0.30	1.16E-03	-0.10	2.74E-01	-3.93	3.09E-167
<i>Rv0678</i> 274insA	-0.23	1.96E-02	-0.29	8.04E-02	-0.40	1.48E-02
<i>AtpE</i> A63P	-2.53	1.07E-198	-2.11	4.92E-53	-1.72	7.15E-34

To further understand the low expression level of *Rv0678* mutants in contrast to the wild-type, the efflux pump gene expression levels of each clone with *Rv0678* mutations were examined BDQ exposed vs. unexposed. According to the general hypothesis, the MTBC bacteria harbouring an *Rv0678* mutation have elevated *mmpS5* / *L5* expression regardless of BDQ exposure (117,118). Across the mutants with different *Rv0678* mutations, a significant upregulation of the *mmpS5* and *mmpL5* efflux pump genes was observed under BDQ exposure compared to themselves without BDQ (Figure 3.8A). The missense mutation in *Rv0678* A99V revealed increase of $\log_2FC$ of 1.98 for *mmpL5* and $\log_2FC$ of 1.96 for *mmpS5* (Figure 3.8B). The clone harbouring the mutation *Rv0678* inversion had a $\log_2FC$ of 2.56 for *mmpL5* and a $\log_2FC$ of 2.39 for *mmpS5*. Finally, the mutant *Rv0678* 274insA showed a $\log_2FC$ of 2.05 for *mmpL5* and $\log_2FC$ of 1.83 for *mmpS5*. The *atpE* A63P mutant had almost no differential regulation of *mmpS5* and *mmpL5* efflux pump genes, as for *mmpL5* $\log_2FC$ = 0.07 and *mmpS5* $\log_2FC$ = 0.27 under BDQ exposure. This analysis revealed that the *Rv0678* mutants exhibit significant upregulation of the *mmpS5* and *mmpL5* efflux pump genes under BDQ exposure as compared to unexposed, in contrast to the *atpE* A63P mutant which showed minimal differential regulation of these genes.

**B**

	<i>Rv0676c</i> (<i>mmpL5</i>)		<i>Rv0677c</i> (<i>mmpS5</i>)		<i>Rv0678</i> (<i>mmpR</i>)	
	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>
<i>Rv0678</i> A99V	1.98	3.57E-142	1.96	2.99E-53	2.46	6.91E-79
<i>Rv0678</i> inversion	2.56	3.36E-237	2.39	1.51E-78	1.78	5.60E-41
<i>Rv0678</i> 274insA	2.05	6.12E-131	1.83	4.17E-40	1.69	1.44E-32
<i>AtpE</i> A63P	0.07	5.93E-01	0.27	1.26E-01	0.53	1.29E-03
Wild type (ancestral)	2.90	1.88E-260	2.81	3.37E-93	2.86	6.42E-91

Figure 3.8: Gene expression profile of *Rv0678* mutants, *atpE* mutant and wild-type after 4 h, bedaquiline exposure vs. unexposed. Mutants (*Rv0678* A99V, *Rv0678* 274insA and *Rv0678* inversion or *atpE* A63P) and Wt transcriptome expression between exposed vs. unexposed to BDQ. (A) Volcano plots indicating differentially expressed genes, statistically significantly differentially expressed ($p < 0.05$) shown in red, NS = not significant in grey. (B) Table comparing the log₂FC and *padj* of the *Rv0678* (*mmpR*), and efflux pump genes (*mmpS5* and *mmpL5*). BDQ+ = BDQ exposed, BDQ- = BDQ unexposed, FC = fold change, *padj* = adjusted p-value, $n = 1$, replicates = 4.

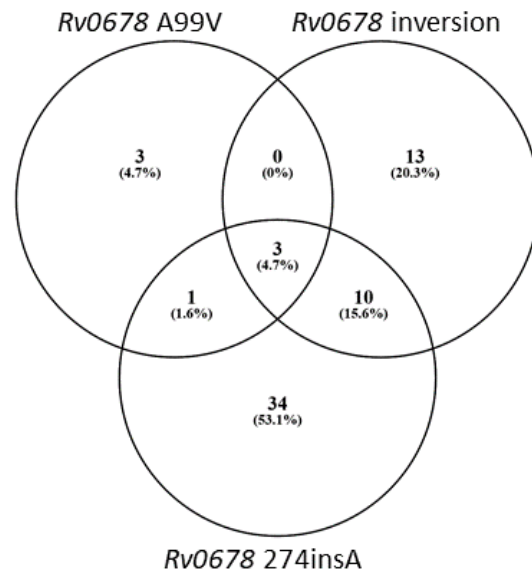
These observations were in concordance with the hypothesis of enhanced *mmpS5* and *mmpL5* efflux pump gene expression in *Rv0678* mutants regardless of BDQ exposure, and supported the mechanism described in the literature (118). Notably, the susceptible ancestral wild-type strain H37Rv exhibited also strong expression of the efflux pump genes of $\log_2FC$ of 2.90 ($padj = 1.88E-260$) for *mmpL5* and $\log_2FC$ of 2.81 ($padj = 3.37E-93$) for *mmpS5* under BDQ exposure (Figure 3.8). This was in concordance with the first experiment; however, the magnitude in which the expression levels of both pump genes were observed was not expected to have the same magnitude as the *Rv0678* mutants. As this observation questions the described resistance mechanism of being solely enhanced efflux pump expression in the *Rv0678* mutants, further investigation of the transcriptome of *Rv0678* mutants was performed to explore commonly shared DEGs other than the *mmpS5*-*mmpL5* efflux pump genes and additional resistance conferring mechanism explaining the low-level resistance of these *Rv0678* mutants.

3.1.2 Clones harbouring resistance-associated mutations in *Rv0678* reveal different transcriptional profiles

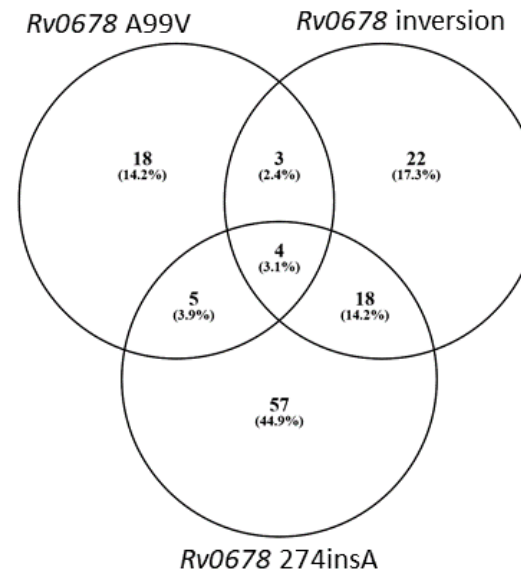
The gene expression profiles of clones harbouring different *Rv0678* mutations revealed an upregulation of the *mmpS5* and *mmpL5* efflux system after 4 h BDQ exposure, in accordance with existing literature (61,118,161,167). Surprisingly, a similar gene expression pattern was observed in the susceptible wild-type H37Rv ancestor. This observation raised the question, if further mechanisms were shared among *Rv0678* mutants leading to BDQ resistance, other than the investigated efflux pump system. To address this question, shared DEGs among the *Rv0678* mutants were investigated.

First, DEGs between the *Rv0678* mutants and the wild-type, were assessed after 4 h without BDQ exposure. Then, the significantly DEGs ($\log_2FC > 1$, $padj < 0.05$) were compared (Figure 3.9A). In total, 37 genes were significantly differentially expressed for the *Rv0678* A99V mutant, 73 genes in the *Rv0678* inversion mutant and 132 DEGs for *Rv0678* 274insA mutant compared to the wild-type H37Rv. The observed genes were sorted in up- and downregulated genes (Figure 3.9, Appendix A Table 0.7). Databases from david.ncifcrf.gov and mycobrowser.epfl.ch were used to catalogue gene products and functional categories of the DEGs (153,154).

A Upregulated DEGs vs. Wt BDQ-



Downregulated DEGs vs. Wt BDQ-



B

	Locus	Gene Name	Functional category	Rv0678 A99V		Rv0678 inversion		Rv0678 274insA	
				Log ₂ FC	padj	Log ₂ FC	padj	Log ₂ FC	padj
Upregulated	Rv2875	Major secreted immunogenic protein Mpt70	Cell wall and cell processes	1.67	2.71E-19	1.57	2.83E-11	1.97	8.68E-28
	Rv0575c	Possible oxidoreductase	Intermediary metabolism and respiration	1.23	1.89E-07	2.18	1.84E-23	4.08	5.10E-85
	Rvnt38	NA		1.19	2.41E-06	1.57	2.83E-11	1.84	1.42E-15
Downregulated	Rv2485c	Probable carboxylesterase LipQ	Intermediary metabolism and respiration	-1.33	1.47E-16	-1.19	1.55E-13	-1.13	1.99E-12
	Rv1286	CysN/CysC: sulfate adenylyltransferase	Intermediary metabolism and respiration	-1.14	1.07E-17	-1.28	8.83E-23	-1.36	6.50E-26
	RVnc0001	NA		-1.07	5.73E-05	-1.51	6.47E-08	-1.75	1.67E-09
	Rv3742c	Possible oxidoreductase	Intermediary metabolism and respiration	-1.02	7.30E-13	-1.19	4.83E-17	-2.01	8.00E-42

Figure 3.9: Significant DEGs shared between mutants with *Rv0678* variants. (A) The significant DEGs between *Rv0678* mutants and ancestral wild-type after 4 h without BDQ exposure (BDQ-). The data represents up- and downregulated genes, between the *Rv0678* mutants. (B) Table of the shared significantly up- and downregulated genes of *Rv0678* mutants, including functional category and log₂FC. BDQ = bedaquiline, DEGs = Differentially Expressed Genes, Log₂FC = Log₂ Fold change, *padj* = adjusted p-value, NA = not available.

Three DEGs were commonly upregulated and four were downregulated (Figure 3.9B). Among the up-regulated genes were: *Rv0575* encoding a possible oxidoreductase involved in metabolism and respiration and *mpt70* (*Rv2875*), encoding a major secreted immunogenic protein involved in cell wall and cell function. The *rvnt38* gene was also upregulated, encoding a small RNA with undetermined function. Notably, both genes were significantly upregulated in all *Rv0678* mutants ($p_{adj} < 0.05$). For the gene encoding an oxidoreductase (*Rv0575c*), the *Rv0678* A99V mutant revealed a $\log_2FC = 1.23$, the *Rv0678* inversion mutant a $\log_2FC = 2.18$ and the *Rv0678* 274insA mutant a $\log_2FC = 4.08$ (Figure 3.9B). The gene encoding Mpt70 exhibited upregulation for the *Rv0678* A99V mutant with a $\log_2FC = 1.67$, for *Rv0678* inversion a $\log_2FC = 1.57$ and for *Rv0678* 274insA a $\log_2FC = 1.97$. Of the 4 downregulated genes three were involved in intermediary metabolism and respiration. *LipQ* (*Rv2485c*), encoding an immune-modulatory lipase, was significantly downregulated in the *Rv0678* mutants: the *Rv0678* A99V had a $\log_2FC = -1.33$, the *Rv0678* inversion had a $\log_2FC = -1.19$, and the *Rv0678* 274insA a $\log_2FC = -1.13$. *CysN / cysC* (*Rv1286*), which encodes a stress response gene, was downregulated with a $\log_2FC = -1.14$ for *Rv0678* A99V, a $\log_2FC = -1.28$ for *Rv0678* inversion and a $\log_2FC = -1.36$ for *Rv0678* 274insA. The gene *Rv3742c* encoding a possible oxidoreductase had a $\log_2FC = -1.02$ for *Rv0678* A99V, a $\log_2FC = -1.19$ for *Rv0678* inversion and for *Rv0678* 274insA a $\log_2FC = -2.01$. The gene encoding *Rvnc0001*, a small RNA with unknown function was also significantly downregulated in all *Rv0678* mutants.

Taken together, the analysis of gene expression profiles for shared resistance-involved mechanisms in the *Rv0678* mutants, without exposure to BDQ, reveal a common upregulation of three genes. Of those, two were involved in cell wall and cell processes and intermediary metabolism and respiration, and one was of unknown function. Four genes were commonly downregulated, three of which were involved in intermediary metabolism and respiration.

3.1.3 Chronological response of *Rv0678* mutants to bedaquiline exposure

In the previous chapter, clones harbouring different *Rv0678* mutations shared only 7 DEGs without exposure to BDQ. All downregulated genes were involved in intermediary metabolism and respiration. The observations made by the previous analysis alone was insufficient to conclude a common mechanism of resistance among the *Rv0678* mutants, beyond the *mmpS5-mmpL5* efflux pump system. Therefore, the question remained whether there is a shared gene expression profile among the *Rv0678* mutants pointing to resistance mechanisms in addition to the efflux pump system. To address this question, significantly DEGs were assessed by comparing the *Rv0678* mutants (with variants *Rv0678* A99V, *Rv0678* 274insA, and an inversion) against the wild-type H37Rv ancestor under BDQ exposure (Figure 3.10). The significantly up- and downregulated DEGs were then compared between the clones harbouring different *Rv0678* mutations to identify genes shared among the mutants under BDQ exposure.

This analysis explores resistance mechanisms in *M. tuberculosis* H37Rv, emphasizing its metabolic plasticity as detailed by Koul et al. (168). *M. tuberculosis* can switch energy sources and metabolic modes based on environmental conditions (66). Additionally, it utilizes alternative pathways like the methylcitrate cycle and methylmalonyl pathway to metabolize propionyl-CoA, a side product of lipid metabolism. By examining DEGs over time, the study aims to reveal gene regulation patterns in key pathways during 0.5 to 4 h of BDQ exposure (Figure 3.10, Appendix A Table 0.8).

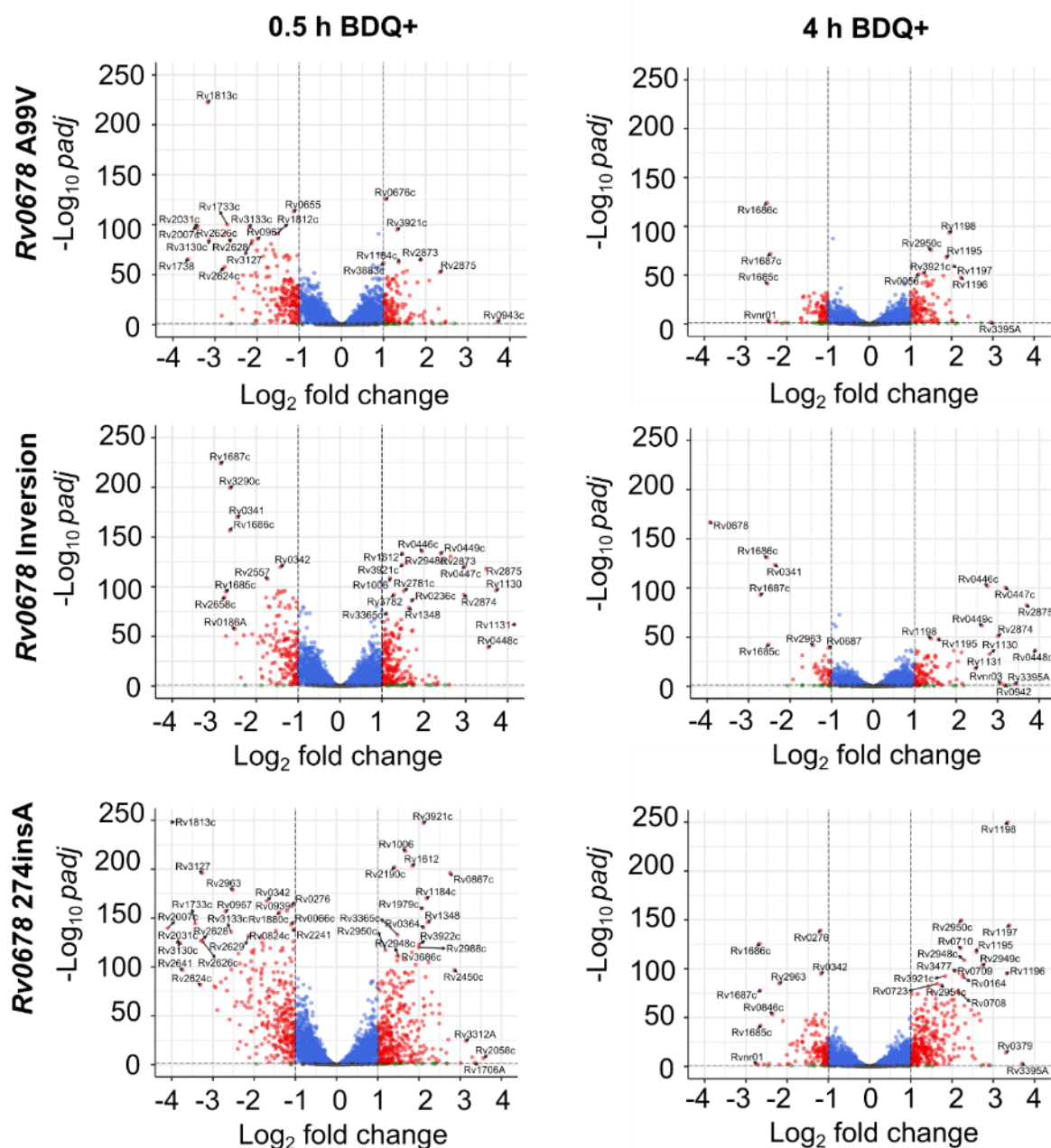


Figure 3.10: Global *Rv0678* mutant response on bedaquiline exposure after 0.5 h and 4 h. The differentially expressed genes (DEGs) were assessed by comparing the *Rv0678* mutants against the ancestral wild-type H37Rv under BDQ exposure. Volcano plot depicting the DEGs of *Rv0678* mutants vs. wild-type under BDQ exposure. BDQ + = exposed to bedaquiline, BDQ - = without exposure to bedaquiline.

In response to BDQ exposure after 0.5 h were in all *Rv0678* variants 77 shared genes upregulated and 86 shared genes downregulated (Figure 3.11). The *Rv0678* variants share almost 11 % (77 / 703) of the individually upregulated genes and 9 % (86 / 665) of the individually downregulated genes in response to 0.5 h BDQ exposure. After 4 h were in all *Rv0678* variants 60 shared genes upregulated and 16 shared genes downregulated (Figure 3.12). This are almost 12 % (60 / 510) which are shared between the *Rv0678* variants of the individually upregulated genes and about 6 % (16 / 272) of individually downregulated genes.

Gene regulation after 0.5 h bedaquiline exposure

The genes differentially regulated after 0.5 h exposure to BDQ compared to wild-type H37Rv were primarily involved in cell wall and cell processes (27 genes upregulated and 15 genes downregulated), also intermediary metabolism and respiration (10 genes upregulated and 24 genes downregulated). However, a substantial amount conserved hypothetical genes (18 genes upregulated and 26 genes downregulated) were also differentially expressed (Figure 3.11).

After 0.5 h of BDQ exposure, 27 genes involved in cell wall and cell processes were upregulated (Appendix A Table 0.9). Among them, there were the *mpt70* (*Rv2875*) immunogenic gene and *mpt83* (*Rv2873*), encoding a lipoprotein, as well as other lipoprotein-genes like *lppi* (*Rv2046*) and *lpqZ* (*Rv1244*). Also, *esxT* (*Rv3444c*) encoding an ESAT-6 like protein, and genes belonging to the ABC transporter family (*Rv2564*, *Rv2563*), as well as other transporter encoding genes such as *irtA* (*Rv1348*), *arsB2* (*Rv3578*) and *mgtE* (*Rv0362*) were upregulated. One of the resuscitation-promoting factor genes, *rpfA* (*Rv0867c*), which promotes resuscitation of dormant *M. tuberculosis*, was also upregulated.

Ten genes that were involved in intermediary metabolism and respiration were upregulated, such as *prpC* (*Rv1131*) and *prpD* (*Rv1130*) involved in methylcytrat cycle, *dipZ* (*Rv2874*), encoding a C-type cytochrome located in an operon with *mtp83* and *mpt70*, and *LeuD* (*Rv2987c*), encoding an isopropylmalate isomerase (Appendix A Table 0.9). Additionally, seven genes involved in information pathways, including *spoU* (*Rv3366*), encoding an RNA methyltransferase, two genes encoding ribosomal proteins (*rpmB2* (*Rv2058c*) and *rpmF* (*Rv0979A*)), and a gene encoding anti-anti-sigma factor (*Rv0516c*) were upregulated. Five genes involved in lipid metabolism, three genes encoding regulatory proteins (*Rv3840*, *Rv1353c*, *Rv2989*), one PE family gene (*PE18*, *Rv1788*) and two virulence-associated genes (*Rv0662c*, *Rv3922c*) were also upregulated.

The downregulation of crucial pathways is a strategy of *M. tuberculosis* to evade antimicrobial pressure by entering a dormant state (80). Among these pathways, genes involved in intermediary metabolism, such as *Rv0575c* (a member of *dosR* regulon), and iron regulator genes like *fdxA* (*Rv2007c*), *fdxD*

(*Rv3503c*) and *bfrB* (*Rv3841*) a bacterioferritin, were downregulated. Additionally, 15 genes related to cell wall and cell processes, including two ABC transporter family genes (*Rv1686c* and *Rv1687c*), *espF* (*Rv3865*), *espA* (*Rv3616c*), *espD* (*Rv3614c*) and the *esxJ* (*Rv1038c*) which encodes an ESAT-6 like protein, were downregulated. The genes *espA* and *espD* are part of co-transcribed gene cluster with genes of ESX-1, and *espF* is also a secretion associate gene involved in virulence. Furthermore, nine down-regulated genes were associated with virulence, detoxification and adaption: *mymT* (*Rv0186A*), *hspX* (*Rv2031c*) a heat shock protein, *Rv2624c* a universal stress family protein, genes belonging to TA-system families such as, the VapBC family (*vapB22* (*Rv2830c*), *vapB28* (*Rv0608*), *vapB36* (*Rv1982A*)) as well as an MazEF family gene (*mazE6* (*Rv1991A*)) and the regulatory gene *trcR* (*Rv1033c*) which is also a part of a TA-system. Additionally, regulatory genes involved in metal homeostasis, including *csorR* (*Rv0967*) and *ctpV* (*Rv0969*), which are essential for copper regulation, and *kmtR* (*Rv0827c*), encoding a transcriptional repressor from the ArsR-SmtB family, were also downregulated.

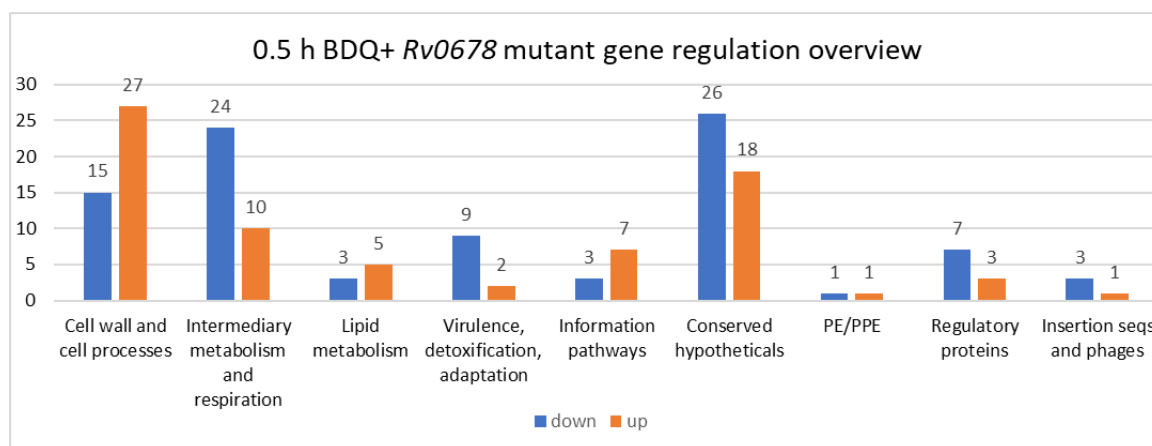


Figure 3.11: Category of DEGs shared between all *Rv0678* mutants after 0.5 h bedaquiline exposure. The DEGs were assessed by comparing the *Rv0678* mutants against the ancestral wild-type under BDQ exposure (BDQ+). Numbers above bars indicate number of genes in the respective category. Categories from mycobrowser.epfl.ch., up = upregulated genes (orange), down = downregulated genes (blue).

Gene regulation after 4 h bedaquiline exposure

After 4 h BDQ exposure compared to no exposure in *Rv0678* mutants, 16 genes attributed to cell wall and cell processes, including two ESAT-6 like genes *esxK* (*Rv1197*) and *esxL* (*Rv1198*), along with conserved hypothetical genes (14 genes), were upregulated in all *Rv0678* mutants (Figure 3.12, Appendix A Table 0.8, Table 0.10). Additionally, 11 upregulated genes were involved in intermediary metabolism and respiration, including *leuC* (*Rv2988*) and *leuD* (*Rv2987*), *prpC* (*Rv1130*), *prpD* (*Rv1131*), which also overlap with the upregulation observed after 0.5 h BDQ exposure.

Sixteen genes were downregulated after 4 h BDQ exposure in *Rv0678* mutants (Figure 3.12, Appendix A Table 0.8, Table 0.10). Among these, four were genes involved in cell-wall and cell processes, including two ABC transporter genes (*Rv1687c*, *Rv1686c*) and *IniB* (*Rv0341*). Also, virulence genes like the anti-toxin (AT) system genes *higB* (*Rv1955*) and *higA* (*Rv1956*), as well as genes involved in defence against oxidative stress and CLZ, *cysK2* were downregulated, at both timepoints.

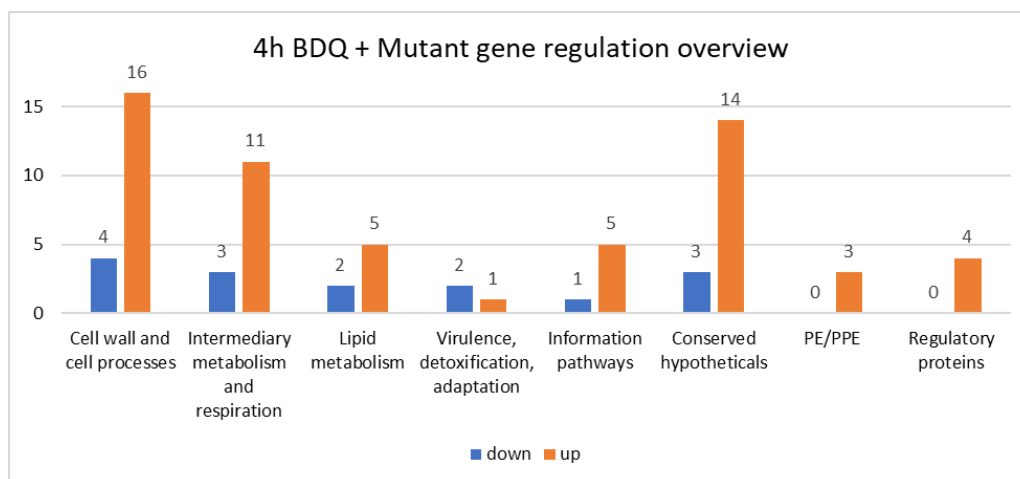


Figure 3.12: Category of DEGs shared between all *Rv0678* mutants after 4 h bedaquiline exposure. The DEGs were assessed by comparing the *Rv0678* mutants against the ancestral wild-type under BDQ exposure (BDQ+). Numbers above bars indicate number of genes in the respective category. Categories from mycobrowser.epfl.ch. Up = upregulated genes (orange), down = downregulated genes (blue).

Commonly regulated genes of clones with different *Rv0678* mutations at both timepoints

In the *Rv0678* mutants between the 0.5 h and 4 h timepoints, 18 DEGs were consistently upregulated under BDQ exposure. These included *prpC* and *prpD* (*Rv1130*, *Rv1131*), and *Rv0516c* encoding an anti-anti sigma factor (Appendix A Table 0.8). Genes associated with transition from dormancy to growth such as *rpfA* (*Rv0867c*), were also upregulated. Furthermore, the gene encoding the transcription regulators *Rv2989* and *leuD* (*Rv2987c*) were upregulated. Additionally, genes involved in cell-wall biosynthesis, such as *embA* (*Rv3794*) and *ufaA1* (*Rv0447c*), and also *spoU* (*Rv3366*) a gene of the information pathways, were upregulated. Furthermore, *dipZ* (*Rv2874*), involved in intermediary metabolism and respiration located in the membrane, and *mpt70* (*Rv2875*) a gene associated with cell wall and cell processes, were upregulated at both timepoints.

Between the timepoints of 0.5 h and 4 h, eight genes were consistently downregulated. Four of these were involved in cell-wall and cell processes, including ABC transporter genes (*Rv1687c*, *Rv1686c*) and a lipoprotein *lpqS* (*Rv0847*). Additionally, three genes (*Rv2913c*, *Rv0848*, *Rv0846c*) involved in intermediary metabolism and respiration were also downregulated.

In this chapter, mechanisms contributing to low-level resistant phenotypes, which lead to enhanced MIC of the mutants were investigated. Therefore, the DEGs of the *Rv0678* mutants compared to the

ancestral wild-type H37Rv were assessed under exposure to BDQ. Furthermore, to explore the changes in the transcriptomic landscape in regard to a metabolism utilizing metabolic plasticity, different timepoints after BDQ exposure were explored to gather a chronological view on the response of *Rv0678* mutants (66,68,169). Taken together, commonly upregulated genes regardless of timepoint were involved in intermediary metabolism, respiration and cell wall and cell processes. Common downregulated genes were involved in cell wall and cell processes. Additional, other genes involved in intermediate metabolism were downregulated. Notably, one gene belonging to the *dosR* dormancy regulon and other transcriptional regulators was also downregulated.

Here, a very dynamic process heavily influenced by the drug exposure and its duration was observed, this multi angled insights should add further to understand response of resistant mutants towards its target drug. The thesis then focused on exploring a novel treatment approach: sequential antibiotic exposure, which demonstrates the principle of negative cellular hysteresis in *M. tuberculosis*. Negative cellular hysteresis describes the reduced capacity of bacterial cells to adapt to rapidly changing environments, especially when exposed to sub-lethal drug concentrations. By strategically switching the sequence of drug exposure, this approach can destabilize the adaptive mechanisms of *M. tuberculosis*, rendering the bacteria more vulnerable to the compounds. This method has the potential to significantly enhance the efficacy of existing antibiotics by potentially preventing the development of resistance, thereby supporting more robust and effective treatment outcomes.

3.2 Sequential exposure approach to explore negative cellular hysteresis in *M. tuberculosis*

As observed in the previous chapter, most resistance mechanisms still need further investigation to be fully understood. Furthermore, with the advanced technique of RNAseq, novel targets for tailored treatment strategies could be wholly investigated. To define such targets, it is important to fully understand the response of the pathogen under drug exposure. As observed by Koul et al., genes involved in alternative energy creating mechanisms and dormancy related expression suggest an metabolic remodelling under drug exposure (specifically BDQ) (168). The observations in this study could be used to tailor alternative treatment strategies using refurbished drugs in an evolutionary informed approach.

R. Roemhild's discovery of hysteresis in *P. aeruginosa* has inspired the exploration of this sequential treatment inducible phenomenon in *M. tuberculosis* (103,106). These evolutionary-informed treatment strategies which utilized sequential administration of two drugs aim to enhance treatment efficacy and potentially slow down or circumvent resistance evolution. This phenomenon, known as “negative cellular hysteresis”, improves treatment outcome by exploiting the enhanced susceptibility of bacteria to the second drug following the exposure to the first (103).

In this thesis, the concept of cellular hysteresis was applied to *M. tuberculosis* using EMB and RIF. An *in vitro* model was first developed where the *M. tuberculosis* H37Rv (ATCC27294) strain was exposed to sub-lethal concentrations of these antibiotics, followed by a time-kill assay, making the first application of this method in *M. tuberculosis*. Finally, to understand the mechanism involved, the initial and late transcriptomic response to the sequential treatment was investigated using RNAseq.

It was hypothesized that the sequential administration of EMB followed by RIF induces negative cellular hysteresis in *M. tuberculosis*, thereby enhancing treatment efficacy. The mechanism behind that phenomenon is thought to be of physiological nature where the pre-exposure to EMB bridges the cell wall and facilitates entrance of RIF.

3.2.1 Establishment of sequential antibiotic exposure designed for *M. tuberculosis*

Initially, the optimal drug concentrations and exposure timeframe for *M. tuberculosis* was determined (103). The antibiotics EMB and RIF, chosen for their different modes of action, were tested to determine MICs for *M. tuberculosis* H37Rv (ATCC27294) using REMA, as described in the Chapter 2.2.2.1. Three independent experiments resulted in an MIC of 0.008 µg / mL for RIF and 2 µg / mL for EMB. Furthermore, the interaction profile of EMB and RIF was investigated for synergy or antagonism using

a checkerboard assay (Figure 3.13) (100). A synergistic interaction between EMB and RIF was observed, with a MIC of 0.002 $\mu\text{g} / \text{mL}$ for RIF and 0.5 $\mu\text{g} / \text{mL}$ for EMB when tested separately, the combined MIC was for RIF 0.001 $\mu\text{g} / \text{mL}$ and for EMB 0.062 $\mu\text{g} / \text{mL}$, using the formular described in Chapter 2.2.2.2, a FIC Index of 0.324 was calculated indicating synergy of the drug combination (Synergy: FICI ≤ 0.5).

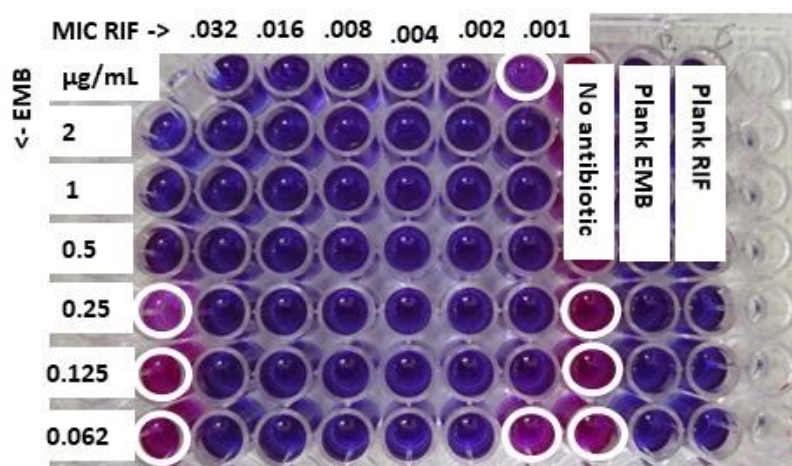


Figure 3.13: Checkerboard assay of ethambutol and rifampicin on *M. tuberculosis* H37Rv. Representative image of a checkerboard assay. Growth was indicated by white circles, no antibiotic growth control and plank = no bacteria, but antibiotic was included. Fluorescence readout at 540 / 35 nm and 590 / 20 nm. The MIC of single drug for *M. tuberculosis* was determined by the lowest concentration preventing growth on the first row and column for ethambutol (EMB) and rifampicin (RIF) respective. The combined MIC was determined by well where the combination of both inhibits growth.

To assess the inducibility of cellular hysteresis by sequential exposure to EMB or RIF, a high throughput setting using 96-well plate microtiter broth dilution was developed, with growth measured by OD_{600} . Testing conditions included: EMB pre-exposure followed by RIF main-exposure, RIF pre-exposure followed by RIF main-exposure and no pre-exposure with RIF main-exposure alongside a medium only growth control. The duration of the time-kill assay was initially explored over a 10-day period. Next, various concentrations were tested for the main treatment. Since, it was known that sub-inhibitory antibiotic concentrations could induce hysteresis, sub-lethal concentrations: 75 % MIC, 50 % MIC, and 25 % MIC were tested. Finally, the optimal antibiotic pre-exposure time was also determined. As the generation time for this H37Rv was 16 h (time for one binary fission), pre-exposure time shorter than the generation time (such as 2 h, 5 h and 8 h) were compared.

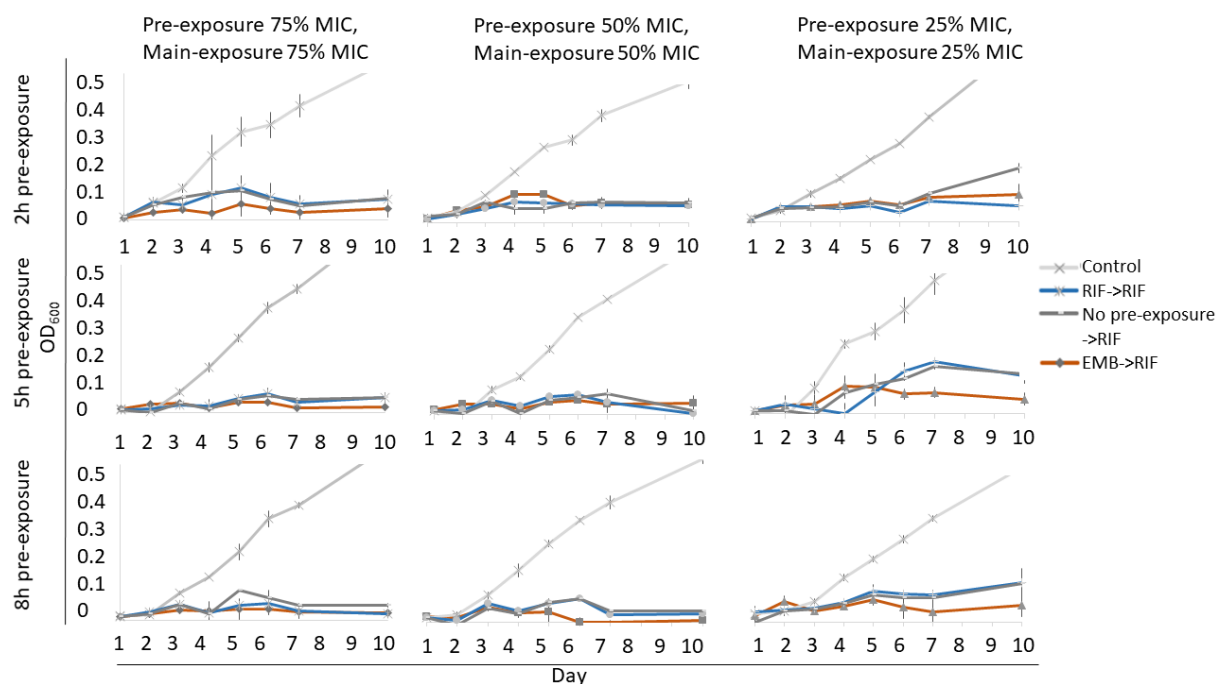


Figure 3.14: High-throughput assay using OD_{600} for initial investigation on hysteresis. A range of sub-lethal concentrations for pre-exposure and main-exposure were tested, simultaneously with different pre-exposure durations. The y-axis depicts the OD_{600} and the x-axis the duration in days after administering the main treatment. Included were the following conditions: sequential treatment of ethambutol (EMB) pre-exposure and rifampicin (RIF) as main-exposure depicted by an orange graph stands, medium control in grey with a cross, the RIF pre-exposure and RIF main-exposure as blue graph and no pre-exposure with RIF main-exposure, $n = 3$.

The high-throughput assay indicated first and foremost an enhanced killing effect of EMB pre-exposure with RIF main-exposure, as compared to treating only with RIF (Figure 3.14). A decrease in OD was observable after 5 h and 8 h pre-exposure with 25 % EMB and 25 % RIF. Exposure to 50 % or 75 % MIC of the drugs suppressed growth too strongly to observe the EMB->RIF interaction. Although 5 h and 8 h preexposure showed similar results, considering the 16 h doubling time of the laboratory reference strain H37Rv, the 8 h exposure time was chosen for future experiments.

Since OD_{600} measurement alone does not distinguish between viable and non-viable cells, further experiments using CFU / mL enumeration were conducted. For the CFU enumeration assay, the culture volume was increased up to 10 mL, and 10 μ L spots were plated to determine CFU / mL. Concentrations between 12 % and 50 % of the MIC for EMB and RIF were tested with 8 h pre-exposure time and 15 days observation after main-exposure (Figure 3.15). Again, when applying 50 % of the MIC as the main-exposure concentration for RIF, the concentration was highly effective, reducing CFU / mL below the detection limit. However, to assess differences between conditions, CFU / mL needed to be detectable. When using 25 % or 12 % of the MIC as main-exposure concentration, differences between conditions were detectable by CFU / mL plating. It was concluded, that the optimal concentration for these experiments were 25 % of the MIC (25 % MIC for EMB = 0.5 μ g / mL and RIF = 0.002 μ g / mL) for both

pre-exposure and main-exposure, as this was the highest concentration where CFU / mL were detectable.

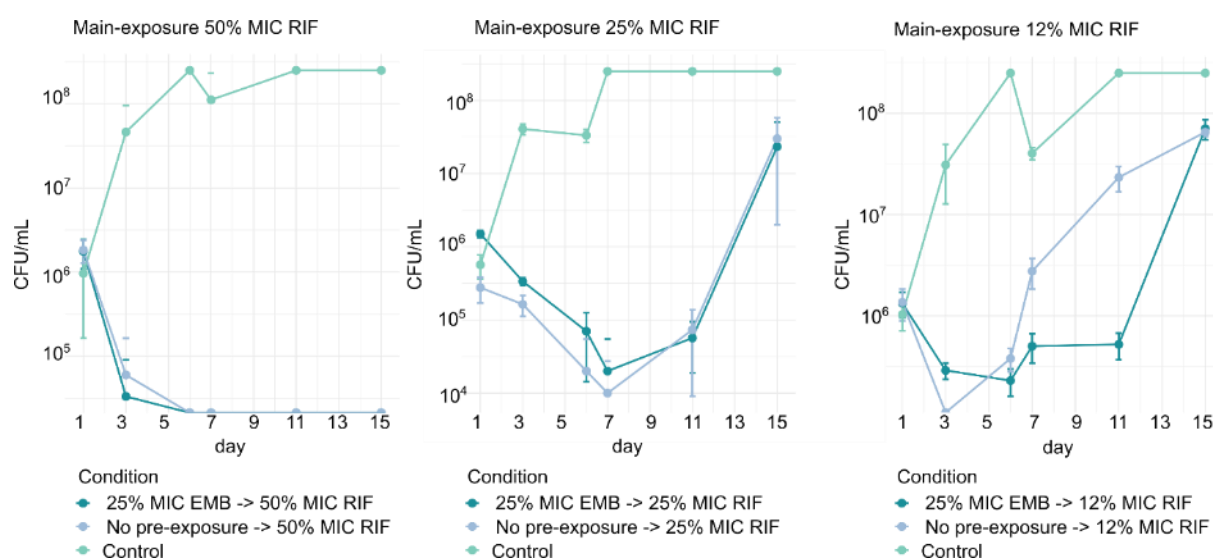


Figure 3.15: Time-kill assay to determine optimal main-exposure concentration following 8 h pre-exposure using CFU enumeration. The x-axis represents time (days) post main-exposure and the y-axis depicts CFU / mL. EMB pre-exposure switched to RIF main-exposure (petrol), RIF treatment without pre-exposure (light blue) and medium control (mint). The CFU / mL was measured at day 1, 3, 6, 7, 11 and 15. CFUs were enumerated after 14 and 28 days. EMB = ethambutol, RIF = rifampicin, CFU = Colony forming units. Error bars represent standard deviation (SD), n = 2, technical triplicates.

3.2.2 Enhanced killing effect of first-line drugs against *M. tuberculosis* is sequence dependent

The previous section described how the sequential antibiotic treatment was adapted and optimized for *M. tuberculosis*, by testing multiple parameters to assess cellular hysteresis detection (103). The final workflow comprised a starter culture from frozen stock, grown and adjusted to about 10^5 CFU / mL, and 10 mL culture was transferred to 50 mL screw cap tubes (Figure 3.16, Chapter 2.2.3.2). The antibiotic for pre-exposure was added to the samples which were incubated for 8 h at 37° C, standing at a 45° angle. Control CFU counts were conducted at three steps: directly from the original culture bottle, from the aliquoted sample, after 8 h incubation and after centrifugation, to determine any cell loss due to antibiotic pre-exposure or centrifugation. Following the drug switch, the initial CFU (day 0) was taken, and a time-kill assay was conducted over 14 days without further medium change. The CFU / mL was measured on days 0, 1, 2, 5, 10 and 14.

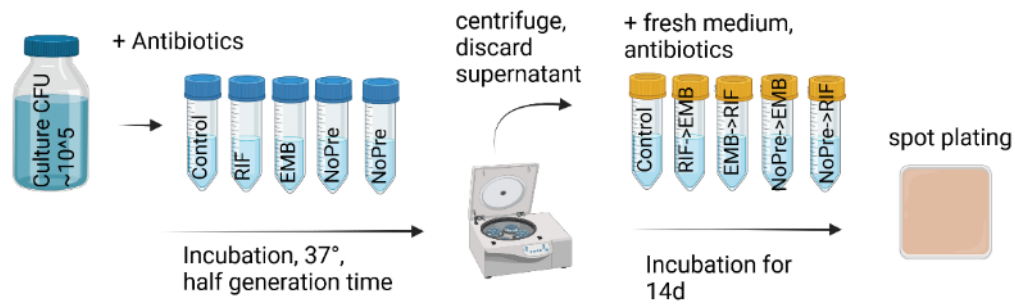


Figure 3.16: Experimental setup for sequential treatment in *M. tuberculosis*. Pre-culture from frozen stock was grown and adjusted to a CFU of 10^5 / mL. Subsequently, 10 mL aliquots were taken and pre-exposed to the first antibiotic for 8 h at 37° C while standing at a 45°angle. After 8 h of exposure, pelleting was performed at 3 500 rpm for 10 min, followed by discarding of the old medium. The cells were then resuspended in fresh 7H9 medium and the second antibiotic “main-exposure” was applied. The cultures were then incubated at 37° C for 14 days. CFU counts conducted by spot plating at days 0, 1, 2, 5, 10 and 14. Sub-lethal concentrations of 25 % MIC EMB or RIF were utilized. D = days, EMB = ethambutol, RIF = rifampicin, NoPre = No pre-exposure (no antibiotic), control = growth control (no antibiotic). Image created with BioRender.com.

The final experimental setup included the following conditions: a no-treatment control with each sample set, EMB pre-exposure for 8 h followed by a switch to RIF (EMB->RIF) and the reverse order (RIF->EMB), and no pre-exposure (medium only) followed by RIF or EMB as main-exposure (No pre-exposure->RIF, No pre-exposure->EMB). Bacterial killing was assessed by reduction in CFU / mL. The following analysis combined data from three independent experiments, where each data point was assessed in triplicate (Figure 3.17). The medium control (yellow) exhibited normal growth behaviour, exponential growth phase from day 2 and entering the stationary phase on day 10.

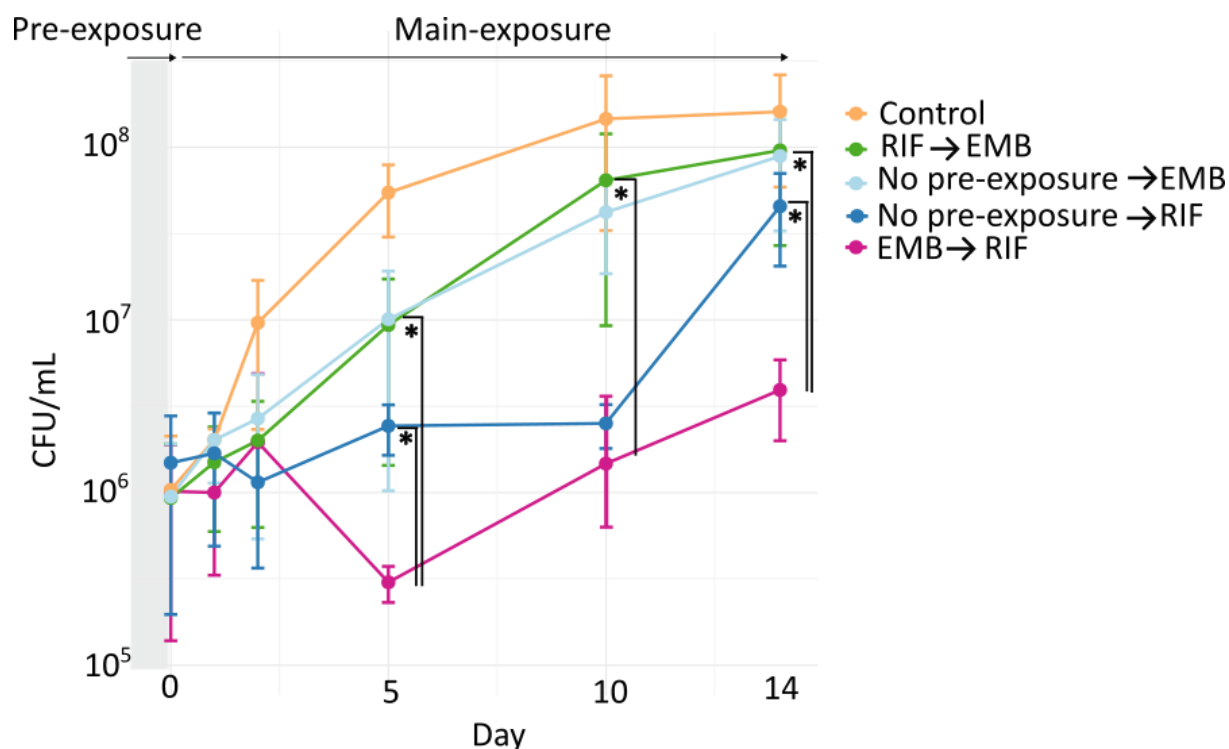


Figure 3.17: Sequential exposure to ethambutol → rifampicin induced killing effect in *M. tuberculosis* H37Rv.

Time-kill assays were performed using laboratory strain H37Rv exposed to different combinations of ethambutol (EMB) or rifampicin (RIF). The x-axis depicts time (days) post main-exposure and the y-axis represents CFU / mL. The grey zone horizontal to the y-axis indicates the 8 h pre-exposure of the first antibiotic with a CFU around 1×10^6 per mL. The switch to the main antibiotic was conducted on day 0. CFU was determined on days 0, 1, 2, 5, 10 and 14. The medium control (yellow), RIF pre-exposure to EMB main-exposure (green), EMB treatment without pre-exposure (light blue), RIF treatment without pre-exposure (dark blue) and EMB pre-exposure switched to RIF main-exposure (pink). Error bars represent standard deviation (SD), $n = 3$, technical triplicates. * $P < 0.001$ denotes significant difference by multiple comparison test using Turkey procedure for p-values post-hoc analysis and Fisher's Combination method.

The condition with no pre-exposure and EMB main-exposure (light blue) and RIF pre-exposure EMB main exposure (green) displayed with half a log-fold growth suppression compared to the control condition (yellow) (Figure 3.17). Comparing both conditions, no significant differences ($p > 0.05$) in CFU / mL counts were observed; these graphs were almost identical. In contrast, as response to RIF main-exposure without pre-exposure (dark blue), the CFU / mL were steady between 10^6 and $10^{6.5}$ till day 5, entered a plateau over five days, then the population rebounded exponentially from day 10 onwards. Finally, when the *M. tuberculosis* culture was pre-exposed to EMB followed by RIF main-exposure (pink), a reduction of about half a log fold (1×10^6 to $1 \times 10^{5.5}$ CFU) in the bacterial population was observed from day 2 until day 5, when the population remained a gradually growth.

The enhancing effect of the treatment is highlighted by comparing the condition with no pre-exposure to RIF main-exposure (dark blue), to EMB then RIF (pink) which resulted in a significant reduction of CFU / mL by one-fold ($p < 0.001$) at days 5 and 14. This effect suppressed the overall population rebound compared to only RIF main-exposure (dark blue) by one-fold change at day 14. It is clear that

EMB pre-exposure then RIF main-exposure resulted in the strongest suppressive effect, compared to all other exposure conditions (Figure 3.17).

This work demonstrated that exposing *M. tuberculosis* first to EMB and subsequently to RIF significantly enhanced bacterial killing compared to other treatment sequences. Initial exposure to EMB is hypothesized to facilitate entry of RIF, leading to an increased reduction in CFU / mL around day two and suppressing population rebound compared to RIF alone. Since the hysteresis effect was observed under sequential exposure of EMB followed by RIF, further investigations on the transcription profile of the sequential treatment were conducted to elucidate the underlying mechanism.

3.2.3 Transcriptomic landscape of cellular hysteresis in *M. tuberculosis*

The previous chapter demonstrated that the sequential administration of EMB followed by RIF induces negative cellular hysteresis effect in *M. tuberculosis* H37Rv. To further investigate the hypothesis that this effect is due to changes in cell wall composition and facilitated drug entry, DEGs were analysed after the drug switch using Illumina RNAseq technology. This approach examines initial and late responses after the sequential treatment in sub-inhibitory concentrations (25 % MIC). The sequential treatment was conducted as previously described, followed by a shortened time-kill assay over two days. The samples for RNAseq were collected at timepoint 0 (TP0), which was used as baseline control. TP1 collected after 8 h exposure to either RIF, EMB or medium only. TP2 was taken post main-exposure or medium switch after 0.5 h rest, and TP3 after 48 h (Figure 3.18).

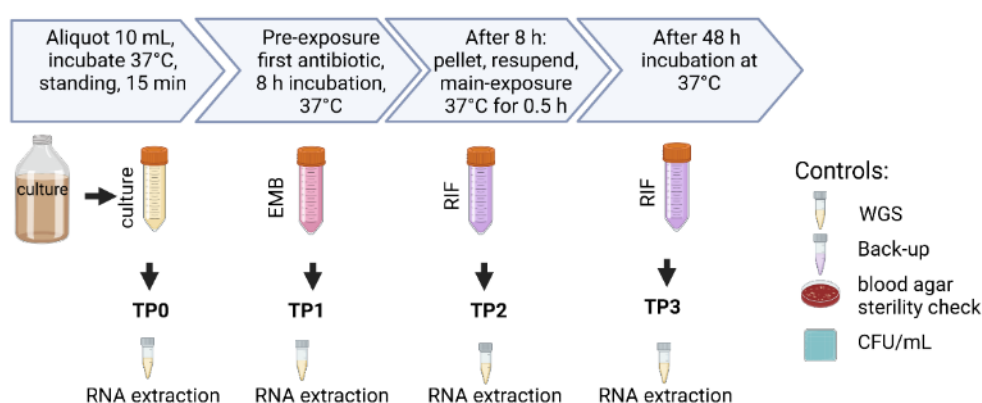


Figure 3.18: Experimental procedure for RNA sequencing of sequential treatment. Sampling for RNA sequencing was conducted at timepoint 0 (TP0) directly of culture after 15 min rest, TP1 after 8 h pre-exposure, TP2 after 0.5 h main-exposure, and TP3 after 48 h main-exposure. And the same for the sequence starting with RIF only. Quality controls such as whole genome sequencing (WGS) validate the strain, a back-up sample, blood agar for sterility check and CFU/mL for growth and viability check. EMB = ethambutol, RIF= rifampicin. Created with Bio-Render.com.

The established RNAseq protocol yielded >12 ng / μ L and high-quality RNA ($RQN \geq 9$) (Chapter 2.2.4.3). However, samples were excluded when the non-rRNA % was below 60 % as this indicated sub-optimal library preparation (Figure 3.19), (Appendix B, Table 0.3). Further quality controls of the run, lead to exclusion of two additional samples from the DESeq analysis, due to inhomogeneous gene body coverage and read counts below 10 million, respectively (Appendix B Table 0.3, Figure 0.1, Figure 0.2, Figure 0.4, Figure 0.5).

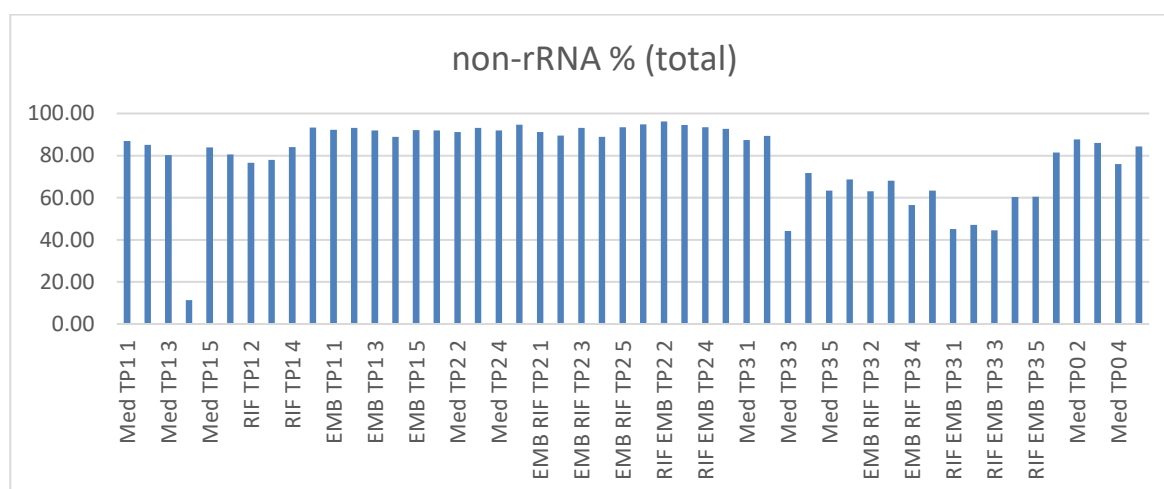


Figure 3.19: Check of non-rRNA content of hysteresis sequencing run. Graph shows the percentage of total non-rRNA content on the y-axis. On the x-axis, all replicates of each sample are shown. EMB = ethambutol, Med = medium only (e.g., no antibiotic), RIF = rifampicin, TP = timepoint.

The PCA plot analysis indicated low variation within the replicates, as evidenced by their close proximity to each other (Figure 3.20). The intragroup variability was minimal across different conditions, indicating less pronounced differences in gene expression (Figure 3.20, Figure 3.21). The samples grouped along the y-axis according to time, with PC1 (74 %) and PC2 (8 %) together explaining a significant amount of the variance in this dataset (Figure 3.20). At TP0, the medium control clusters separately. At TP1, samples exposed to RIF grouped together, except for two replicates that clustered with the EMB samples. However, the variance was minimal (about 5 %) thus these replicates were not excluded from downstream analysis. Notably, at TP2, the drug sequence EMB→RIF, which induced negative cellular hysteresis, grouped closely with the reverse sequence, RIF→EMB. The low variance in these clusters indicates less variation of their gene expression patterns. Furthermore, TP3, collected 48 h after the drug switch, formed a distant group separate from the earlier timepoints, although the different conditions within TP3 clustered all together.

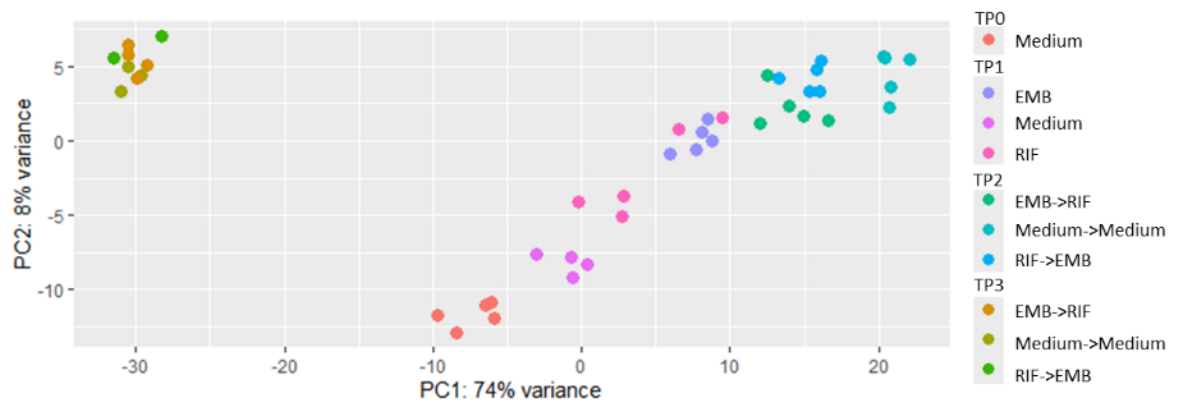


Figure 3.20: The PCA Plot of hysteresis transcriptomic analysis. The *in vitro* experiment on H37Rv utilized ethambutol (EMB) and rifampicin (RIF) which previously induced negative cellular hysteresis, followed by a short time-kill assay. The samples group according to timepoints (TP). TP0 refers to medium only at the start of experiment, TP1 includes all samples after 8 h incubation, TP2 highlights samples of drug switch with 0.5 h incubation in the new drug and TP3 includes samples 48 h past the antibiotic switch.

To investigate the transcriptomic landscape of the sequential exposure, the focus was on three key timepoints: 8 h of exposure to one drug (TP1), shortly after the drug switch with 15 min rest period (TP2) and 48 h after the drug switch (TP3). Initially, the DEGs at each timepoint were identified by comparing the respective sample to a medium control of the same timepoint. First investigation of the functional categories across the 50 most variable genes was conducted to get an overview of the transcriptome. This revealed that 40 % of the expressed genes belonged to conserved hypotheticals, 21 % were involved in intermediary metabolism and respiration, and 13 % were related to cell wall and cell functions (Figure 3.21A, Appendix B Table 0.3, Table 0.4). Notably, other categories including regulatory proteins, lipid metabolism, insertion sequences, and phage together made up the remaining 26 % of DEGs.

A Functional categories of 50 most variable genes
0.5 h and 48 h after BDQ+ exposure

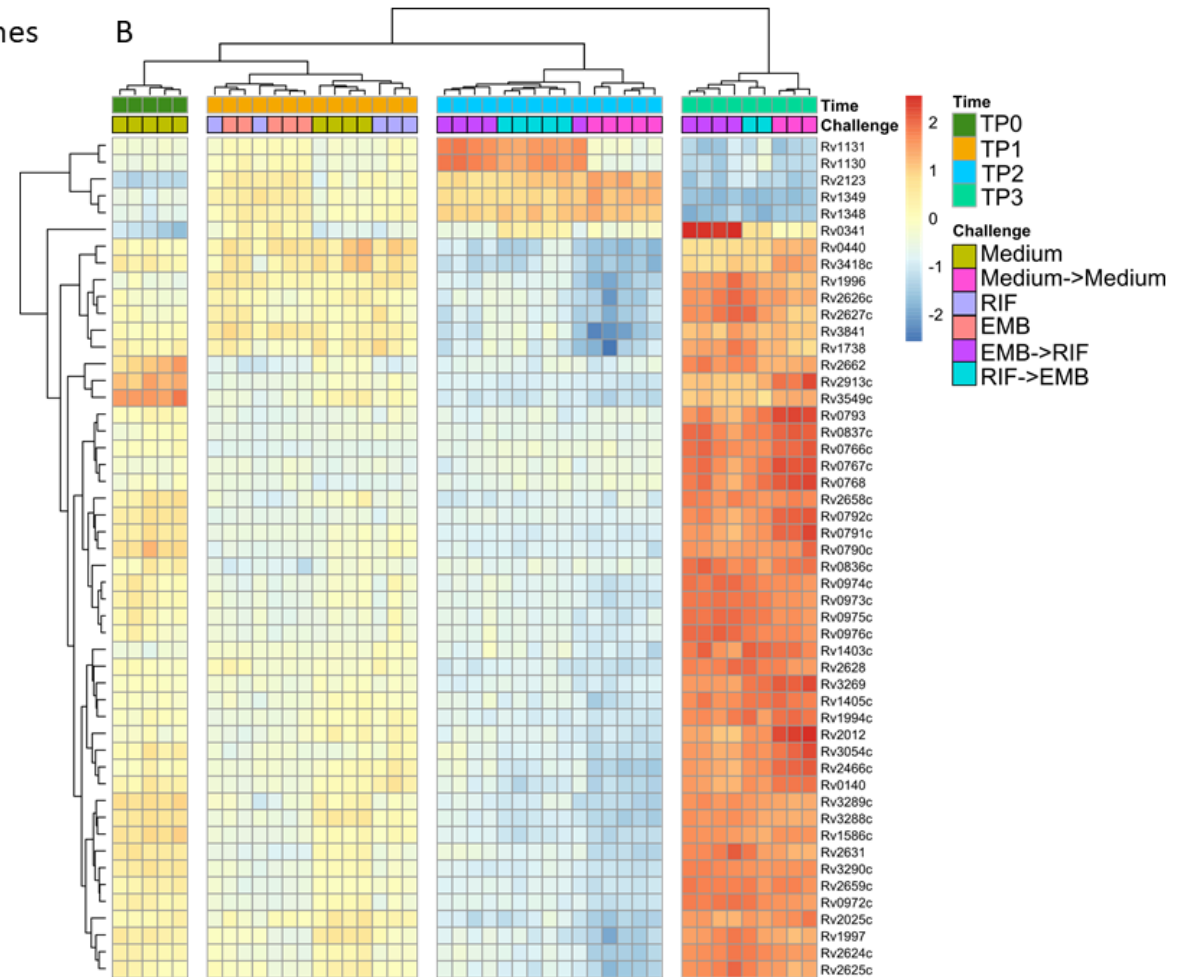
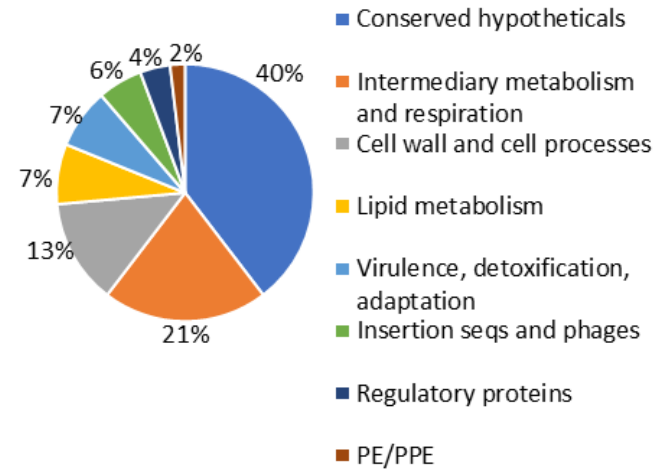


Figure 3.21: The 50 most variable expressed genes of all samples and functional categories. (A) Overview of functional categories across the 50 most variable expressed genes displayed in the heatmap. (B) Heatmap of the 50 most variable genes expressed at timepoint 0 (TP0) baseline culture transcriptome, TP1 or after 8 h pre-exposure to first drug, TP2 0.5 h after drug switch and TP3 or 48 h after drug switch. The scale bar represents a z-score of relative expression from mean, with red indicating upregulated and blue downregulation. Hierarchical clustering of rows and columns. EMB = ethambutol, RIF = rifampicin.

The initial response to antibiotic switch and primary exposure predominantly involved genes associated with metabolism and respiration followed by cell wall and cell functions (Figure 3.21A). The samples of TP1, mono drug exposure for 8 h, had similar expression profiles as genes in TP0, except the upregulation of a set of three genes, a D-amino acid aminohydrolase (*Rv2913c*), a dehydrogenase / reductase (*Rv3549c*) and a hypothetical gene (*Rv2662*) involved in intermediary metabolism and respiration (Figure 3.21B). These three genes were downregulated at TP2, 15 min after antibiotic switch followed by upregulation at TP3, 48 h after the drug switch. Most genes were downregulated at TP2, besides five genes, *prpC* (*Rv1131*) and *prpD* (*Rv1130*) involved in intermediary metabolism and respiration, PPE37 (*Rv2123*) belonging to PE / PPE family, both iron regulated transporter genes *irtB* (*Rv1349*) and *IrtA* (*Rv1348*) involved in cell wall and cell processes, were upregulated for both EMB->RIF (violet) and RIF->EMB (turquoise), unlike the medium control. Interestingly, at TP3 most genes in the heatmap (45 genes) were upregulated in both sequential treatment samples. Notably, of *iniB* (*Rv0341*) expression increased significantly by 2-fold in the EMB->RIF sample (violet), this gene is linked to virulence, reduce cell wall permeability, and stress response in *M. tuberculosis*. Taken together, the 50 most variable genes shared between all timepoints and samples indicated differences mainly between 8 h exposure and 48 h of exposure and downregulation after the drug switch of most genes.

In order to shed light on the dynamic regulation of DEGs at different timepoints during sequential exposure, to understand mechanisms involved in cellular hysteresis, a detailed gene expression analysis per sample at each respective timepoint was conducted. This would allow to determine specific gene expression patterns for the different conditions. This involved assessing all DEGs at each sample to the respective medium control at each timepoint, visualized by volcano plots (Figure 3.22). After EMB exposure (TP1), 172 DEGs (93 upregulated, 79 downregulated) were observed. After the EMB->RIF switch 0.5 h exposure (TP2), 81 DEGs (77 upregulated, 4 downregulated) were detected, and 43 DEGs (19 upregulated, 24 downregulated) were observed 48 h after switch at TP3 (Figure 3.22A). For the RIF treatment (TP1), 50 DEGs were identified after 0.5 h (38 upregulated, 12 downregulated) (Figure 3.22B). For the TP2 RIF->EMB switch, 35 DEGs (34 upregulated, 1 downregulated) were expressed, and 60 DEGs (43 upregulated, 17 downregulated) were observed after 48 h of exposure (TP3).

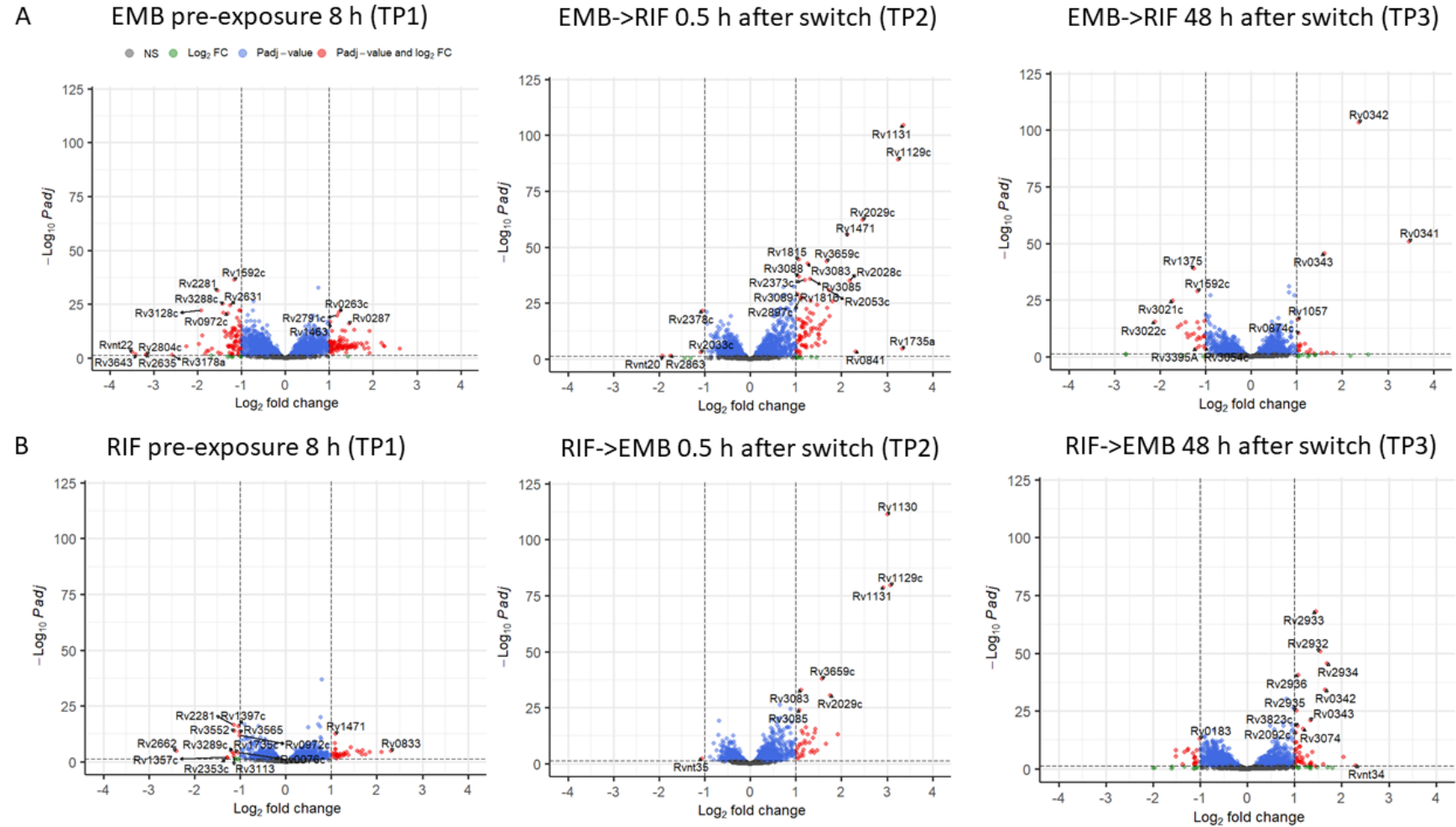


Figure 3.22: Transcriptome under sequential exposure comparing exposure condition and unexposed control. Each volcano plot shows differentially expressed genes (DEGs) relative to the medium control of the specific timepoint. (A) EMB pre-exposure for 8 h, EMB followed by RIF 0.5 h and 48 h. (B) RIF pre-exposure for 8 h, RIF followed by EMB 0.5 h and 48 h. For each graph the y-axis indicates the adjusted p-value ($padj$) and the x-axis displays the log₂ fold change ($\log_2$ FC) of DEG. Grey dots indicated not significance, green dots indicate non-significance and $\log_2$ FC > 1, blue dots indicated $padj < 0.05$ and red dots indicate significant values with $\log_2$ FC > 1 and $padj < 0.05$. Sample indicated with the locus were ranked according to highest $padj$ value, followed by highest or lowest $\log_2$ FC. EMB = ethambutol, RIF = rifampicin, TP = timepoint.

Next, only the significant ($\log_2FC > 1$, $padj < 0.05$) DEGs were compared to the medium control of the respective timepoint (e.g., EMB TP1 vs. medium TP1). The genes marked in Figure 3.22 were filtered as most significant genes in respect to the highest $padj$ value, followed by the most upregulated or most downregulated $\log_2FC$. Additionally, the DAVID platform (david.ncifcrf.gov), was employed to carry out GO and KEGG enrichment analysis. In brief, GO analysis identifies the most prominent biological processes, cellular components, and molecular functions by determining which genes in the provided DEG list are enriched in these categories. KEGG pathway enrichment uses statistical analysis to pinpoint pathways that are significantly represented within the DEG list.

The chronological response to sequential exposure was assessed by first investigating pre-exposure to one drug and then the subsequent drug switch after 0.5 h and 48 h of exposure. The pre-exposure to EMB over 8 h (TP1) induced significant ($\log_2FC > 1$, $padj < 0.05$) upregulation in a set of genes involved in cell wall and cell processes, such as the ESAT-6 like gene, *esxG* (*Rv0287*) ($\log_2FC = 1.45$, $padj = 7.66E-17$) and *Rv1463*, a ABC transporter ($\log_2FC = 1.03$, $padj = 1.91E-17$) (Figure 3.22A) (Appendix B Table 3.3). GO analysis found a cluster of upregulated genes when exposed to EMB 8 h, which are associated with cell surface, involved in virulence, majorly members of PE_PGRS family genes (Appendix B Table 0.5). Notably, biological processes involved in cell wall organization for example peptidoglycan synthesis (*murE*, *murG*, *Rv2719c*) were enriched in the upregulated genes. Downregulation of biological processes such as DNA transposition and molecular functions like transposase activity (*IS1081* (*Rv2512c*)) was also observed (Appendix B Table 0.6). The KEGG enrichment analysis of upregulated genes, revealed an enrichment in porphyrin metabolism and general metabolic pathways (Appendix B Table 0.7). Porphyrins are organic compounds involved in cell processes, and metabolic and respiratory functions. The *pitB* (*Rv2281*) ($\log_2FC = -1.54$, $padj = 5.43E-32$) and conserved hypotheticals were downregulated. Taken together the sample under EMB pre-exposure mainly expressed DEGs involved in cell wall maintenance like peptidoglycan synthesis (*murE*, *murG*).

Next, following the drug switch from EMB to RIF, i.e., main-exposure after 0.5 h exposure (TP2); genes involved in intermediary metabolism and respiration were most significant upregulated. These genes as depicted in Figure 3.22A, were: *prpD* (*Rv1130*) ($\log_2FC = 3.42$, $padj = 1.85E-146$), *prpC* (*Rv1131*) ($\log_2FC = 3.35$, $padj = 3.07E-105$), *trxB1* (*Rv1471*) ($\log_2FC = 2.12$, $padj = 2.20E-56$) and *Rv3085* ($\log_2FC = 1.26$, $padj = 1.05E-36$) (Figure 3.22A, Table 3.3). Additionally, transmembrane proteins, *Rv2053c* ($\log_2FC = 1.72$, $padj = 1.85E-31$) and *Rv0841* ($\log_2FC = 2.30$, $padj = 3.52E-04$), along with genes involved in lipid metabolism such as *Rv3088* ($\log_2FC = 1.06$, $padj = 9.58E-38$), and *Rv3089* ($\log_2FC = 1.04$, $padj = 9.97E-30$) and two transcriptional regulatory proteins, *Rv1129c* ($\log_2FC = 3.24$, $padj = 6.00E-90$) and *Rv1816* ($\log_2FC = 3.24$, $padj = 3.61E-28$), were upregulated. *Rv1816*, is a member of the TetR regulator family and, regulates *mmpL3* and *mmpL11*. Additionally, stress-related genes, including the chaperone *dnaJ2*

(*Rv2373c*) ($\log_2FC = 1.18$, $padj = 3.73E-36$) and *Rv2028c* ($\log_2FC = 2.18$, $padj = 7.87E-36$) were upregulated. By next performing the GO analysis on the whole set of upregulated DEGs the importance of *dnaJ2*, *dnaJ1*, *dnaK*, *sigB*, *hsp* and *hspX* was verified as they were enriched in biological processes such as heat response, protein refolding and chaperone-dependent protein refolding (Appendix B Table 0.3). A second set of genes *Rv3087*, *tgs1* and *tgs4*, was determined to be involved in nitric response, triglyceride biosynthesis and glycerol metabolic processes (Appendix B Table 0.8). In addition, the KEGG analysis revealed that genes involved in glycolipid metabolism ($p\text{-value} < 0.05$) and biosynthesis of unsaturated fatty acids pathways ($p\text{-value} = 0.06$) were among the DEGs, as an important for cell wall maintenance (Appendix B Table 0.9). Altogether, this analysis observed that the switch between antibiotics induced *dnaK*, enhances expression of lipid metabolism crucial for cell wall maintenance, and regulatory proteins.

Finally, looking into 48 h after RIF main-exposure following EMB pre-exposure switch (TP3), genes of *iniBAC* operon: *iniA* (*Rv0342*) ($\log_2FC = 2.36$, $padj = 3.02E-104$), *iniB* (*Rv0341*) ($\log_2FC = 3.46$, $padj = 9.48E-52$) and *iniC* (*Rv0343*) ($\log_2FC = 1.61$, $padj = 2.13E-46$), are upregulated (Figure 3.22A, Table 3.3). In contrast, members of the virulence covering PPE family *PPE47* (*Rv3021c*) ($\log_2FC = -1.73$, $padj = 1.80E-25$) and *PPE48* (*Rv3022c*) ($\log_2FC = -2.11$, $padj = 3.91E-16$), were most significantly ($\log_2FC > 1$, $padj < 0.05$) downregulated. GO and KEGG analysis did not reveal functional clusters. Although the transcriptomic response was not very dynamic at TP3, the increased expression of the *iniBAC* operon suggests that this sequence could promote EMB resistance, given the operon's known role in conferring resistance to cell surface-active drugs.

Table 3.3: Significant DEGs of sequential exposure ethambutol->rifampicin compared to no pre-exposure ethambutol over time. DEGs of each timepoint relative to the medium control of the specific timepoint. Grey highlighting indicates most significance ($\log_2FC > 1$, $p_{adj} < 0.05$) as indicated with locus name in the Figure 3.21. $\log_2FC$ = $\log_2$ Fold Change, p_{adj} = adjusted p-value. EMB = ethambutol, RIF = rifampicin. Gene function and categories asses according to mycobrowser.epfl.ch.

Locus	Gene name	Category	EMB 8 h		EMB->RIF 0.5 h		EMB->RIF 48 h	
			$\log_2FC$	p_{adj}	$\log_2FC$	p_{adj}	$\log_2FC$	p_{adj}
Rv0287	ESAT-6 like protein EsxG	cell wall and cell processes	1.45	7.66E-17	-0.55	1.55E-03	0.47	4.02E-02
Rv1463	Probable ABC transporter	cell wall and cell processes	1.03	1.91E-17	-0.25	4.63E-02	0.10	6.05E-01
Rv2281	Putative phosphate-transport permease PitB	cell wall and cell processes	-1.54	5.43E-32	0.66	1.59E-06	-0.21	2.80E-01
Rv1130	<i>prpD</i>	cell wall and cell processes	0.85	1.88E-08	3.42	1.85E-146	0.09	7.53E-01
Rv1131	<i>prpC</i>	cell wall and cell processes	0.65	2.93E-04	3.35	3.07E-105	0.19	5.97E-01
Rv1471	Probable thioredoxin TrxB1	metabolism and respiration	0.75	2.93E-07	2.12	2.20E-56	-0.88	2.46E-07
Rv3085	Probable short-chain type dehydrogenase / reductase	metabolism and respiration	0.06	6.07E-01	1.26	1.05E-36	-0.38	1.92E-02
Rv2053c	Probable transmembrane protein FxsA	cell wall and cell processes	-0.33	4.49E-02	1.72	1.85E-31	0.00	9.97E-01
Rv0841	Probable conserved transmembrane protein	cell wall and cell processes	0.45	4.98E-01	2.30	3.52E-04	-2.75	8.42E-02
Rv3088	Putative triacylglycerol synthase Tgs4	lipid metabolism	-0.06	5.47E-01	1.06	9.58E-38	0.34	8.29E-04
Rv3089	Probable chain -fatty-acid-CoA ligase FadD13	lipid metabolism	-0.26	8.44E-03	1.04	9.97E-30	0.19	1.35E-01
Rv1129c	Probable transcriptional regulator protein	regulatory proteins	0.65	4.17E-04	3.24	6.00E-90	-0.39	1.75E-01
Rv1816	Possible transcriptional regulatory protein	conserved hypothetical	-0.69	4.40E-09	3.24	3.61E-28	-0.18	3.20E-01
Rv2373c	Probable chaperone protein DnaJ2	virulence, detoxification	0.15	1.58E-01	1.18	3.73E-36	0.13	3.82E-01
Rv2028c	Universal stress protein family protein	virulence, detoxification	-0.85	1.95E-06	2.18	7.87E-36	0.16	5.57E-01
Rv0342	IniA, isoniazid-inducible gene	cell wall and cell processes	0.20	6.38E-02	0.42	2.30E-05	2.36	3.02E-104
Rv0341	IniB, isoniazid-inducible gene	cell wall and cell processes	1.56	8.15E-14	-0.48	2.54E-02	3.46	9.48E-52
Rv0343	IniC, isoniazid-inducible gene	cell wall and cell processes	0.35	1.31E-03	0.37	3.90E-04	1.61	2.13E-46
Rv3021c	<i>PPE47</i>	PE-PPE	-0.43	1.05E-02	0.24	1.65E-01	1.73	1.80E-25
Rv3022c	<i>PPE48</i>	PE-PPE	-0.27	4.50E-01	0.47	1.56E-01	-2.11	3.91E-16

Next, looking into the sequence of RIF pre-exposure followed by EMB main-exposure. First checking the DEGs of 8 h exposure to RIF only against the no-antibiotic control at the same timepoint, focusing on upregulated DEGs.

The most significant ($\log_2FC > 1$, $padj < 0.05$) upregulated genes were involved in virulence like *PE_PGRS13* (*Rv0833*) ($\log_2FC$ 2.30, $padj = 1.51E-05$) and redox reactions like *trxB1* (*Rv1471*) ($\log_2FC$ 1.09, $padj = 3.12E-13$) also involved in thioredoxin system (Figure 3.22B, Table 3.4). The GO analysis on the whole upregulated DEGs, supports this observation as it identified a functional annotation cluster in the biological process of virulence (PE_PGRS Family genes, *esxG*). The KEGG analysis found enrichment of a cofactor biosynthesis pathway (*hemY*, *CobT*, *bioF1*) ($p = 0.07$) (Appendix B Table 0.10). Similar to EMB exposure also *pitB* (*Rv2281*) a phosphate-transport permease was downregulated under RIF only exposure. The set of downregulated DEGs was not enough for GO or KEGG analysis. Overall, pre-exposure to RIF only indicates induction of virulence processes and the redox balancing system.

Now, the transcriptional reaction, on the drug switch from RIF to EMB following 0.5 h incubation (TP2) is investigated and compared to the EMB->RIF antibiotic switch. Genes involved in intermediary metabolism and respiration like after the drug switch of EMB to RIF were prominent. The genes *prpD* (*Rv1130*) ($\log_2FC$ 3.00, $padj = 3.81E-112$) and *prpC* (*Rv1131*) ($\log_2FC$ 2.92, $padj = 1.55E-79$), the transcriptional regulator *Rv1129c* ($\log_2FC$ 3.07, $padj = 1.91E-80$), along with *Rv3083* ($\log_2FC$ 1.11, $padj = 1.31E-33$), *Rv2029* ($\log_2FC$ 1.75, $padj = 3.28E-31$) and *Rv3085* ($\log_2FC$ 1.08, $padj = 1.28E-24$) were most significantly ($\log_2FC > 1$, $padj < 0.05$) upregulated (Figure 3.22B, Table 3.4). GO analysis revealed enrichment of genes belonging to biological processes like responses to heat, stress and protein folding (*grpE*, *narX*, *dnaK*, *hsp*, *hspX*, *SigB*) (Appendix B Table 0.12). This functional annotation cluster was also observed after the switch from EMB to RIF (Appendix B Table 0.8). Notably, the second cluster of the switch RIF->EMB was involved in transcriptional regulation (*Rv0081*, *SigB*, *Rc1129c*). Solely one gene of the significantly DEGs, with unknown function, was downregulated after the antibiotic switch. In conclusion, 0.5 h after both antibiotic switches a similar response was noted.

Finally, 48 h after RIF switch to EMB (TP3), these *M. tuberculosis* bacteria exhibit an increase of the isoniazid-inducible genes *iniA* (*Rv0342*) ($\log_2FC$ 1.64, $padj = 5.74E-35$) and *iniC* (*Rv0343*) ($\log_2FC$ 1.33, $padj = 9.04E-22$), similar as 48 h after EMB->RIF switch (Figure 3.22B, Table 3.4). Notably, genes involved in lipid metabolism, such as *ppsC* (*Rv2933*) ($\log_2FC$ 1.45, $padj = 7.76E-69$), *ppsB* (*Rv2932*) ($\log_2FC$ 1.54, $padj = 1.52E-51$), *ppsD* (*Rv2934*) ($\log_2FC$ 1.68, $padj = 2.51E-46$) and *ppsE* (*Rv2935*) ($\log_2FC$ 1.04, $padj = 5.95E-26$), along with two transporter genes, the ABC transporter *drvA* (*Rv2936*) ($\log_2FC$ 1.08, $padj = 1.86E-41$) and the *mmpL8* transporter *Rv3823c* ($\log_2FC$ 1.06, $padj = 1.87E-19$) were upregulated. GO analysis on all significantly upregulated DEGs, showed the enrichment of a gene cluster (*ppsBCE* and *pks* coding for polyketide synthases), involved in biological processes like DIM / DIP cell wall layer

assembly and fatty acid biosynthesis process, phenolic phthiocerol biosynthetic and phthiocerol biosynthetic process (Appendix B Table 0.13). The GO analysis of downregulated genes showed a small cluster (*clgR*, *hsp*) involved in biological process of heat response (Appendix B Table 0.14). KEGG analysis yielded no results due to an insufficient number of genes. Taken together, the induction of genes involved in lipid metabolism and transporters can be observed as well as the upregulation of *iniA* a resistance associated gene, 48 h after switch from RIF->EMB.

Table 3.4: Significant DEGs of sequential exposure rifampicin->ethambutol compared to no pre-exposure rifampicin, over time. DEGs of each timepoint relative to the medium control of the specific timepoint. Grey highlighting indicates most significance ($\log_2FC = 1$, $p_{adj} < 0.05$) as indicated with locus name in the Figure 3.21. $\log_2FC = \log_2$ Fold Change, p_{adj} = adjusted p-value. EMB = ethambutol, RIF = rifampicin. Gene function and categories asses according to mycobrowser.epfl.ch.

Locus	Gene name	Category	RIF 8 h		RIF->EMB 0.5 h		RIF->EMB 48 h	
			$\log_2FC$	p_{adj}	$\log_2FC$	p_{adj}	$\log_2FC$	p_{adj}
Rv0833	PE-PGRS family protein PE_PGRS13	PE / PPE	2.30	1.51E-05	-0.11	8.80E-01	0.59	4.86E-01
Rv1471	Probable thioredoxin TrxB1	metabolism and respiration	1.09	3.12E-13	1.12	1.66E-15	-0.46	3.87E-02
Rv1130	Possible methylcitrate dehydratase PrpD	metabolism and respiration	0.38	2.91E-02	3.00	3.81E-112	1.10	3.33E-06
Rv1131	Probable prpC, methylcitrate synthase	metabolism and respiration	0.38	6.35E-02	2.92	1.55E-79	1.10	3.82E-04
Rv1129c	Probable transcriptional regulator protein	regulatory proteins	0.14	5.54E-01	3.07	1.91E-80	-0.11	7.98E-01
Rv3083	Probable monooxygenase (hydroxylase)	metabolism and respiration	0.21	6.87E-02	1.11	1.31E-33	-0.49	1.21E-03
Rv2029c	6-phosphofructokinase PfkB	metabolism and respiration	-0.80	1.97E-06	1.75	3.28E-31	0.20	4.50E-01
Rv3085	Probable short-chain type dehydrogenase / reductase	metabolism and respiration	0.10	4.70E-01	1.08	1.28E-24	1.64	5.74E-35
Rv0342	Isoniazid inducible gene protein IniA	cell wall and cell processes	0.13	3.14E-01	0.80	6.19E-17	1.64	5.74E-35
Rv0343	Isoniazid inducible gene protein IniC	cell wall and cell processes	0.19	1.42E-01	0.46	8.80E-06	1.33	9.04E-22
Rv2933	Phenolphthiocerol synthesis type-I polyketide synthase PpsC	lipid metabolism	0.79	1.26E-37	0.31	7.18E-07	1.45	7.76E-69
Rv2932	Phenolphthiocerol synthesis type-I polyketide synthase PpsB	lipid metabolism	0.40	1.66E-06	0.28	3.74E-04	1.54	1.52E-51
Rv2934	Phenolphthiocerol synthesis type-I polyketide synthase PpsD	lipid metabolism	0.75	1.54E-16	0.14	1.72E-01	1.68	2.51E-46
Rv2935	Phenolphthiocerol synthesis type-I polyketide synthase PpsE	lipid metabolism	0.42	5.44E-08	0.03	7.65E-01	1.04	5.95E-26
Rv2936	Daunorubicin-dim-transport ATP-binding protein ABC transporter DrrA	cell wall and cell processes	0.49	4.22E-16	-0.05	4.65E-01	1.08	1.86E-41
Rv3823c	Conserved integral membrane transport protein MmpL8	cell wall and cell processes	0.20	3.98E-02	-0.17	6.55E-02	1.06	1.87E-19

These investigations highlighted how the transcriptomic profiles following exposure to EMB and RIF have similarities but important distinctive differences. Taken together, exposure to EMB without alone upregulated genes involved in cell wall and cell processes. Whereas, RIF exposure induced the upregulation of a PE_PGRS protein family gene. Notably, 48 h after the antibiotic switch, genes of the *iniBAC* operon were as well upregulated after exposure to both EMB or RIF. These results warranted further analysis to uncover the negative cellular hysteresis mechanism by the specific EMB->RIF exposure sequence.

The negative hysteresis effect of ethambutol to rifampicin on the *M. tuberculosis* transcriptome

It was shown in Chapter 3.2.2, that pre-exposure to EMB before RIF (EMB->RIF) resulted in a reduction of CFU / mL over time compared to the same drug administered without pre-exposure, demonstrating a negative hysteresis effect. Likewise, this reduction was drug order dependent as the same effect was not observed when first treating with RIF followed by EMB. The analysis in the previous paragraphs revealed distinct gene expression profiles following exposure to EMB or RIF, with EMB upregulating cell wall-related genes and RIF enhancing a virulence-related gene expression, while both antibiotics initially upregulated methylcitrate cycle genes and later induced the *iniBAC* operon, with EMB also enhancing lipid metabolism genes after 48 h. To shed light on transcriptional differences specifically between the two drug sequences EMB->RIF vs. RIF->EMB, the DEGs at 0.5 h and 48 h after both drug switch were further compared.

An initial response after the drug switch from EMB to RIF compared to RIF->EMB (0.5 h post main-exposure), induced a significant expression of eight genes ($\log_2FC > 1$, $padj < 0.05$) (Figure 3.23A). Of these, six genes were upregulated, including a crucial transcription regulator for cell wall maintenance (*Rv1816*) ($\log_2FC$ 1.15, $padj = 2.76E-27$), a TetR family transcription regulator (*Rv3830c*) ($\log_2FC$ 1.07, $padj = 4.87E-10$) which is known to enhance multi-drug efflux pump expression when upregulated, a virulence-conferring antigenic protein *vapC18* ($\log_2FC$ 1.38, $padj = 1.60E-02$), and *mpt70* ($\log_2FC$ 1.06, $padj = 4.64E-09$) involved in cell wall and cell process. Two genes were significantly downregulated: *IniB* (*Rv0341*) ($\log_2FC$ -1.44, $padj = 2.43E-12$), which belongs to *IniBAC* operon which is induced by cell wall-active antibiotics in response to cell wall stress, and *mymT* (*Rv0186A*) ($\log_2FC$ -1.05, $padj = 1.09E-04$), which protects the cell from copper toxicity (Figure 3.23B).

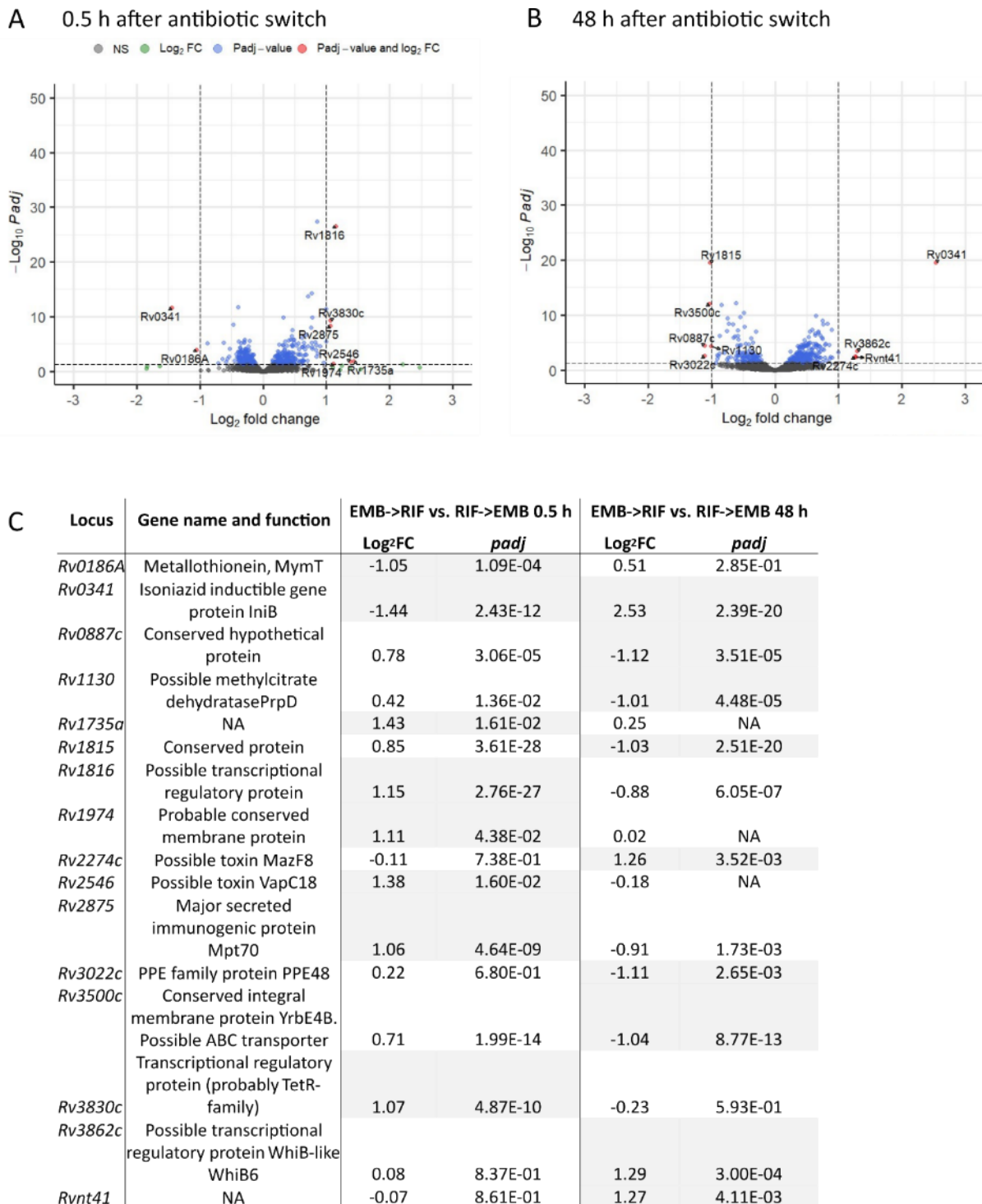


Figure 3.23: Differentially Expressed Genes (DEGs) after switch ethambutol to rifampicin vs. rifampicin to ethambutol. (A) DEGs after 0.5 h of sequential exposure EMB->RIF vs. RIF->EMB. (B) DEGs 48 h after antibiotic switch EMB->RIF vs. RIF->EMB. Genes were ranked based on their *padj* values and absolute log₂-fold change, with significantly differential regulation at each timepoint indicated by red dots in the volcano plots. Genes without significance were depicted in grey dots. Genes with significant log₂-fold change are shown in green, those with a significant adjusted p-value are shown in blue. (C) Table of genes in volcano plot with their gene name and function. Grey highlighting indicates statistical significance in accordance to the timepoint with the threshold *padj* = 0.05, log₂FC = 1, determined using Wald test, corrected for multiple testing with Benjamin Hochberg.

The analysis of DEGs after 48 h main-exposure to RIF with EMB pre-exposure compared to the reverse (RIF->EMB), revealed nine DEGs: *iniB* (*Rv0341*) ($\log_2FC$ 2.53, $padj$ = 2.39E-20), *whiB6* (*Rv1815*) ($\log_2FC$ 1.29, $padj$ = 3.00E-04), and *mazF8* (*Rv3500c*) ($\log_2FC$ 1.26, $padj$ = 3.52E-03) and five genes with undetermined function (Figure 3.23B). Notably, *whiB6* is an important regulator of ESX-1 and Dos regulon. Further, five downregulated DEGs were observed, including *prpD* (*Rv1130*) ($\log_2FC$ -1.01, $padj$ = 4.48E-05), *PPE48* (*Rv3022c*) ($\log_2FC$ -1.11, $padj$ = 2.65E-03), and conserved hypothetical genes.

In conclusion, pre-exposure to EMB switch to RIF significantly (1-fold change) reduced CFU / mL over time, an effect depended on the order of drug administration. Investigating transcriptional differences between the order of RIF or EMB exposure, revealed that for EMB->RIF, 8 DEGs at 0.5 h post drug switch and 9 DEGs after 48 h, were expressed related to cell wall function, virulence, detoxification, adaptation and regulatory mechanisms. The dynamics in the transcription profile during sequential exposure is reflected by the various functional categories involved, highlighting induction of important regulatory genes such as *whiB6*, upregulated 48 h after EMB->RIF switch.

4 Discussion

The persistent challenge of drug resistant and MDR-TB continues to undermine current treatment strategies, with over 400 000 new RR / MDR-TB cases annually (170). Several factors including the long-term, complicated and often toxic treatment regimens, delayed diagnostics, and pharmacokinetic interactions facilitate the selection of drug resistant MTBC strains, which then can be transmitted further (43,171–173). Despite the success of a novel shortened all-oral MDR-TB treatment regimen, the observation of rapid BDQ resistance evolution, a pivotal component of this regimen, is of concern (174). Accordingly, a precise understanding of resistance mechanisms is key for effective diagnostics and treatment (61). Rapid resistance evolution demands an exploration for alternative treatment strategies, refurbishing antibiotics with the potential to circumvent or slow down resistance evolution (87).

This thesis adopts a multilateral approach to address the current knowledge gaps in understanding the resistance mechanisms to BDQ, the most novel cornerstone drug for the treatment of MDR-TB. Nearly all BDQ resistance observed in clinical MTBC strains is linked to variants in the *Rv0678* gene, despite these mutants exhibiting only moderate resistance (162,162,175,176). To better understand the impact of different *Rv0678* mutations on *M. tuberculosis* clones, the transcriptomic landscape of laboratory-generated mutants was analysed both in the presence and absence of BDQ.

As part of the effort to address resistance evolution, this thesis also aimed to demonstrate the principle of sequential treatment to enhance therapeutic efficacy and prevent the development of resistance. This approach was novel in the context of *M. tuberculosis* and was tested using the susceptible laboratory strain H37Rv along with the well-studied first-line drugs EMB and RIF. Finally, RNAseq was employed to investigate the potential molecular mechanisms underlying the observed effects.

4.1 Transcriptomic analysis of bedaquiline resistant mutants with and without bedaquiline exposure

Understanding the mechanisms of resistance is essential for improving treatment strategies and mitigating resistance evolution in the ongoing fight against drug resistant MTBC strains. Despite the careful and gradual introduction of new drugs, the rapid emergence of resistance continues to be a major concern, contributing to the public health crisis posed by MDR-TB (114,157,175). BDQ, approved by the US Food and Drug Administration (FDA) in 2012, has become a cornerstone in MDR-TB treatment (177). However, BDQ resistant MTBC strains already emerged (22). Although BDQ targets the ATP synthase, encoded by the *atpE* gene, MTBC strains harbouring a mutation in this gene are rarely detected in the field (61,118,160,175,178). Interestingly, nearly all clinically detected resistant strains and subsequent treatment failure have been linked specifically to variants in the *Rv0678* gene (22,65,118,178).

These off-target mutations have been observed throughout the *Rv0678* gene, which encodes a MarR-like transcriptional regulator that is believed to control the expression of the *mmpS5* and *mmpL5* efflux pump genes (179–181). The transcription regulator *Rv0678* binds to the intergenic region between *Rv0678* and *mmpS5* (*Rv0677c*) (118,166). This binding prevents transcription, resulting in diminished expression of *mmpS5* and *mmpL5* (117,118,166). It has been postulated that depending on the mutation type in the *Rv0678* gene, this will lead to structural changes in the regulator, prohibiting DNA binding. As a result, transcription of efflux pump genes is increased, even without the exposure to BDQ. It is still not known whether the types of mutation, i.e. frameshift mutation or an amino acid polymorphism, have varying impacts on protein functionality (117,118,165,167).

This work aims to understand the impact of different *Rv0678* mutations on the expression of the efflux pump genes *mmpS5-mmpL5*, through transcriptome analysis using Illumina™ RNAseq. This was achieved by exposing resistant mutants harbouring different *Rv0678* mutation to high concentration of BDQ (10x MIC). Which revealed differences in the mutant's expression profile of the *mmpS5-mmpL5* efflux pumps in regard to the *Rv0678* variant.

4.1.1 Varying efflux pump expression levels between clones with different *Rv0678* mutations regardless of bedaquiline exposure

The expression profiles of *mmpL5*, *mmpS5*, and *Rv0678* were investigated by RNAseq in three clones harbouring different *Rv0678* mutations (i.e., A99V, inversion or 274insA) after 4 h exposure to BDQ or unexposed. First, the efflux pump gene expression was analysed by comparing the mutants to the ancestral wild-type H37Rv in the absence of BDQ, to establish baseline gene expression levels (Chapter 3.1.1 Figure 3.7). The hypothesis was that in clones with different *Rv0678* mutations, the defective transcription regulator would result in an increased expression of the *mmpL5-mmpS5* efflux pump system, regardless of BDQ exposure (166). Additionally, it was expected that the type of mutation would influence the expression levels of these efflux pumps (65,182).

Efflux pump expression without antibiotic exposure revealed different expression profiles between *Rv0678* mutants

As hypothesized, clones with different *Rv0678* mutations exhibited higher expression levels of the *mmpS5* and *mmpL5* genes compared to the wild-type, even without BDQ exposure. Moderate increase in expression levels (between $\log_2\text{FC} = 0.6\text{--}1$) of both efflux pump genes were observed across all *Rv0678* mutants when compared to the wild-type, without BDQ (Chapter 3.1.1 Figure 3.7). Notably, the degree of *mmpS5-mmpL5* expression differed among *Rv0678* mutants. Furthermore, mutations that severely disrupted the *Rv0678* gene, such as frameshift mutations or insertions, were associated with a higher MIC (4–8x higher) in previous studies, whereas single amino acid changes resulted in a

broader range including moderately increased and high MIC (2 - 8x higher) (61,182,183). This suggests that the mutation type influences the expression levels of *mmpS5* and *mmpL5*, and the BDQ resistance level.

Interestingly, expression of *mmpS5* and *mmpL5* at baseline level was highest in the *Rv0678* missense mutant A99V, with a $\log_2FC$ of 1.04 for *mmpS5* and a $\log_2FC$ of 0.86 for *mmpL5* (Chapter 3.1.1 Figure 3.7, Appendix A Table 0.5). Both of the mutant clones with a frameshift mutation revealed similar expression levels, 274insA mutant had a $\log_2FC$ of 0.62 for *mmpL5* and $\log_2FC$ 0.69 for *mmpS5* and the inversion mutant had a $\log_2FC$ of 0.64 and 0.61 for *mmpL5* and *mmpS5*, respectively.

Generally, it is thought that frameshift mutations cause a loss of function of the protein as the sequence changes drastically (184). Thus, the observations obtained here are somehow in contrast to this understanding as the missense mutation (A99V) resulted in a significantly enhanced efflux pump expression (about 1-fold) compared to the frameshift mutations in *Rv0678* at baseline level only (Appendix A Table 0.5). However, recent studies reported that frameshift mutation might not influence protein function, and can result in similar protein function as within the wild-type H37Rv (185,186). Such reports could explain the variation between the A99V missense and the frameshift mutations in this study, thus the effect on protein function needs to be further investigated. Additionally, a paper in preprint on *Rv0678* mutations in *M. tuberculosis*, proposes that frameshift mutations may not necessarily lead to a resistant phenotype ($MIC \geq 1 \mu g / mL$), if they are in a specific position which is not important for gene function (182). However, in this work the clone harbouring the 274insA mutation displayed a MIC of $1 \mu g / mL$, indicating that this non-synonymous SNP could result in a reduced function of the protein but not in a complete loss of function. This could explain the slightly lower *mmpS5* expression levels in the clone harbouring the 274insA mutation compared to the clone harbouring the A99V mutation, without BDQ exposure (Chapter 3.1.1 Figure 3.7, Appendix A Table 0.5). However, under BDQ exposure the clone harbouring 274insA mutation had similar *mmpS5*-*mmpL5* expression levels as the clone harbouring A99V mutation.

In the end, the enhanced MIC of the frameshift mutants in this work might indicate an additional resistance mechanism. Since no other resistance-related mutation was detected (in *atpE* or additional clones with different *Rv0678* mutations), these findings further reinforce the possibility of an alternative resistance mechanism linked to *Rv0678* mutations. These results highlight the complexity in understanding the functional pathways of resistance-conferring mutations and emphasize the need for further investigation, as supported by findings in other studies (120,187).

As expected, the clone with the *atpE* A63P mutation exhibited minimal changes in expression levels, with a $\log_2FC$ of 0.30 for *mmpL5* and $\log_2FC = 0.44$ for *mmpS5*. This aligns with expectations, as the

mutation alters the drug target, effectively abolishing the drug's action, and *atpE* mutations are not reported to influence Rv0678 protein function.

Exposure to bedaquiline induces *mmpS5-mmpL5* efflux pump gene expression regardless of *Rv0678* mutation

To better understand the effect of *Rv0678* mutations on *mmpL5-mmpS5* efflux pump genes and their expression levels, the transcriptional profile of clones harbouring different *Rv0678* mutations were compared to the ancestor wild-type H37Rv when exposed to BDQ for 4 h or not. The first comparison of *Rv0678* mutants under BDQ exposure vs. unexposed revealed an enhanced *mmpL5-mmpS5* efflux pump gene expression (Chapter 3.1.1 Figure 3.8B).

All clones with different *Rv0678* mutations had a similar increase in efflux pump expression levels when exposed to BDQ vs. unexposed. The clones with an *Rv0678* missense mutation (A99V) exhibited almost four times increased expression levels for *mmpL5* ($\log_2FC = 1.98$) and also for *mmpS5* ($\log_2FC = 1.96$) when exposed to BDQ (Chapter 3.1.1 Figure 3.8). Both frameshift mutations in *Rv0678*, the inversion and the 274insA, showed enhanced expression of *mmpL5* ($FC > 4$) and *mmpS5* ($FC > 2$) efflux pump genes when exposed to BDQ vs. unexposed.

Notably, when comparing the ancestral wild-type H37Rv exposed to BDQ vs. unexposed, both *mmpL5* and *mmpS5* efflux pump genes, displayed an increased expression level, to a magnitude comparable to the *Rv0678* mutants (*mmpL5* = $\log_2FC$ of 2.90, and *mmpS5* = $\log_2FC$ of 2.81) (Chapter 3.1.1 Figure 3.8). As expected, the *atpE* A63P mutant clone did not exhibit increased *mmpS5-mmpL5* expression levels in response to BDQ exposure compared to the unexposed condition ($\log_2FC = 0.07$ for *mmpL5* and $\log_2FC = 0.27$ for *mmpS5*), because BDQ is ineffective against this clone, as the mutation alters the BDQ target. However, it was surprising that the ancestral wild-type H37Rv strain exhibited also an enhanced *mmpS5-mmpL5* expression when exposed to BDQ vs. unexposed to a similar level as observed for the clones with different *Rv0678* mutations (Chapter 3.1.1 Figure 3.8).

In order to better understand the increase in *mmpL5* and *mmpS5* expression levels of the ancestral wild-type H37Rv, the *Rv0678* mutants were compared to the wild-type under BDQ exposure as outlined in Chapter 3.1.1 Figure 3.8 (Appendix A Table 0.4). The observed decrease of efflux pump genes *mmpL5* and *mmpS5* expression levels in *Rv0678* mutants compared to ancestral wild-type H37Rv under BDQ exposure, indicates that there was no statistically significant difference in efflux pump expression between *Rv0678* mutants and wild-type H37Rv under exposure to BDQ. In contrast, when looking at the *atpE* mutant (A63P) compared to the ancestral wild-type H37Rv under BDQ exposure, the efflux pump expression levels for both efflux pump genes were almost not enhanced, as it would be expected due to the different target of *atpE* mutation.

This observation challenges the previously held assumption that a dysregulated *mmpS5-mmpL5* efflux pump system is the primary resistance mechanism in MTBC strains with *Rv0678* mutations, and points towards additional mechanisms beyond *Rv0678* efflux pump regulation such as the induction of additional resistance conferring pathway.

Indeed, the BDQ-induced expression of *mmpS5-mmpL5* genes in H37Rv likely represents a normal physiological response to antibiotic-induced stress. It is also plausible that additional regulatory mechanisms may enhance *mmpS5-mmpL5* expression (116). For example, a recent study on siderophores and their transport systems (*mmpS4-mmpL4* / *mmpS5-mmpL5*) suggested that *Rv0455c*, a gene encoding a previously unidentified secreted core protein in MTBC, might also facilitate the activity of MmpL efflux systems like MmpL4-MmpS4 and MmpL5-MmpS5 by potentially linking them to an outer membrane channel (Appendix A Table 0.5) (188). However, *Rv0455c* is unlikely to play a role in this study, as it was not significantly differentially regulated in *Rv0678* mutants compared to the wild-type, both with and without BDQ exposure, nor when comparing BDQ-exposed and unexposed *Rv0678* mutants (Appendix A Table 0.6). Furthermore, under normal conditions, *Rv0678* suppresses transcription through DNA binding. MarR-family regulators, such as *Rv0678*, are generally known to detect stress signals and are activated by factors like antibiotics or oxidative stress (166). Crystallographic resolution of *Rv0678* protein revealed a complex with an isomer of 2-stearoylglycerol (166). In the presence of this isomer, *Rv0678* was unable to bind the target promotor region (166). This could indicate that a compound or substrate produced during BDQ exposure could potentially induce a conformational shift in the DNA-binding side or block *Rv0678*, preventing it from binding to DNA and inhibiting transcription, thus leading to expression of *mmpS5-mmpL5* in the wild-type H37Rv. Looking to other effects caused by *Rv0678* mutations, it was reported that the *Rv0678* regulator is able to bind other promotor regions of *mmpS2-mmpL2*, *mmpS4-mmpL4*, and *Rv0991-Rv0992* (166). Therefore, mutations in *Rv0678* may not only affect *mmpL5-mmpS5* expression, but also other genes / operons potentially involved in resistance. Still, the exact mechanisms regulating *Rv0678* have not yet been defined.

To gain deeper insight into the response to BDQ in clones with different *Rv0678* mutations compared to the wild-type, the RNAseq data were further explored to determine additional resistance mechanisms. First, as the obtained observations were contradictory to the current consensus of the scientific community, the samples underwent additional quality controls to validate the assumed variants. As a first step, the bam files were visualized with IGV v7.12.7 genome browser and inspected for the respective mutation. The expected mutations were all confirmed, the magnitude of the *mmpL5* and *mmpS5* expression was checked using wiggle files for visualisation. This specific format displays continuous data as probability score, equalized in size making it possible to compare samples with different read number, for magnitude of gene expression reflected as changes in height.

Subsequently, literature research revealed a study from 2015, which investigated efflux pump expression via qRT-PCR in response to sub-inhibitory concentrations of RIF, EMB and streptomycin. They reported elevated efflux pump expression of *mmpL5* (*Rv0676c*) in the H37Rv wild-type (115). Although, BDQ was not investigated, these finding suggests the possibility of another resistance mechanism that may accompany the intrinsic efflux pump resistance mechanism. Supporting this, Koul et al. observed in a study from 2014 using micro arrays, a reshaping of the metabolism in *M. tuberculosis* under BDQ exposure (168). Furthermore, it was observed that overexpression of efflux pump genes might be as well induced in response to cell stress, which could further explain the observed upregulation in the susceptible wild-type under BDQ exposure (189).

To provide a deeper understanding of the potential pathways involved in BDQ resistance, the full transcriptome of *Rv0678* mutants was analysed in the absence of BDQ. This allowed for a comprehensive assessment of the *Rv0678* mutation's global impact on gene expression, aiming to identify additional resistance mechanisms. The analysis is presented in separate chapters, each focusing on different functional categories.

4.1.2 The transcriptomic profile is diverse among clones with different *Rv0678* mutations in absence of bedaquiline

The previous section demonstrated that clones with various *Rv0678* mutations and the ancestral wild-type H37Rv show a similar enhanced *mmpL5-mmpS5* gene expression under BDQ exposure, leading to the conclusion that other mechanisms are potentially involved in the resistance of the clones with *Rv0678* mutations to BDQ. To identify molecular patterns or pathways potentially involved in resistance, the full transcriptome of the different *Rv0678* mutants was first compared to the ancestral wild-type H37Rv without BDQ exposure (Chapter 3.1.2 Figure 3.9). The DEGs in the *Rv0678* mutants were then analysed, and the significantly DEGs were further assessed.

A total of 242 DEGs were assessed, of which 7 DEGs were commonly expressed in the investigated *Rv0678* mutants at baseline level (Chapter 3.1.2 Figure 3.9, Appendix A Table 0.7). The three upregulated genes were *Rv0575c* (an oxidoreductase), *Rv2875* (*mpt70*, major secreted immunogenic protein) and a gene of unknown function. The four downregulated genes commonly expressed in *Rv0678* mutants without exposure to BDQ were: *Rv2485c* (*LipQ*, an immunogenic lipase), *Rv1286* (*cysN* / *cysC*, a stress response gene), *Rv3742c* (encoding a possible oxidoreductase) and a gene with unknown function. The low number of shared DEGs without BDQ exposure reveals a diverse transcriptome of clones with different *Rv0678* mutations at baseline. Since the genes in this analysis are involved in different functional categories such as intermediary metabolism / respiration and cell wall / cell processes, their interplay and effect can only be hypothesized based on the transcription data.

The enhanced expression of certain genes in the *Rv0678* mutants against the wild-type, such as the oxidoreductase *Rv0575c*, has been reported in response to hypoxic stress (80,190,191). Oxidoreductases catalyse redox reactions by transferring electrons between molecules. Given that BDQ targets components of the electron transport chain, the expression of the oxidoreductase *Rv0575c* at baseline level may represent an alternative energy transport mechanism improving the performance of the *Rv0678* mutants in the event of BDQ exposure. *Mpt70* is thought to be involved in cell wall and cell function and was reported to be induced in response to harsh environmental conditions like hypoxia (192,193). In MTBC strains, *mpt70* is located in an operon together with *mpt83*, and with *dipZ* located between these genes (194). This operon is typically regulated by *sigK*, encoding for a sigma factor that reacts upon environmental stress. In this thesis, the entire *mpt70-dipZ-mpt83* operon was expressed, suggesting the involvement of a less well-characterized stress response system.

LipQ, which encodes for an immunogenic lipase that regulates cytokine production, was downregulated in all *Rv0678* mutants compared to the ancestral H37Rv wild-type at baseline level (195,196). Since these lipases, including *lipQ*, are also regulated by sigma factors (*sigF* regulates *lipQ*) and are likely induced by environmental stress, its downregulation at baseline level might only indicate the absence of such stressors (194). Furthermore, *Rv1286* (*cysN*, bifunctional enzyme involved in the sulphate assimilation pathway), was also downregulated at baseline. Typically upregulated under stress, *cysN* may only be induced in such conditions (197). Additionally, *cysN* contains also an HLA-E motif, recognised by cytotoxic T-cells. Thus, its downregulation could hypothetically provide a fitness advantage for *Rv0678* mutants (198).

Conversely, *Rv3742c*, which codes for another oxidoreductase, was downregulated in clones with different *Rv0678* mutations compared to ancestral wild-type without BDQ exposure. A similar downregulation was observed in transcriptomic data of MDR-TB outbreak samples from Thailand, including the MTBC strain of the Beijing ST10 genotype, compared to the H37Rv strain under untreated conditions which did not show such downregulation (199). However, the impact of these regulation on *Rv0678* mutants necessitates further investigations.

Gene expression profiling of *Rv0678* mutants without BDQ exposure revealed varying profiles with few commonly regulated genes. Overall, genes encoding an oxidoreductase, an immunogenic protein and a gene with unknown function were upregulated. Other genes encoding an oxidoreductase, an immunogenic lipase, a stress response gene and also a gene with unknown function were downregulated. This indicates that clones with different *Rv0678* mutations exhibit an unexpected diverse set of DEGs at baseline. While RNAseq provides a snapshot of transcriptome changes, it may not directly reflect functional gene changes (182). Complementary proteomics and Western blot analyses would enhance these findings (164).

To further explore additional resistance mechanisms, the DEGs of the different *Rv0678* mutants vs. wild-type under BDQ exposure were assessed, then the transcriptional response of different *Rv0678* mutants to BDQ after 0.5 and 4 h was analysed as outlined in the following section (Chapter 3.1.3 Figure 3.10).

4.1.3 Chronological response of clones with different *Rv0678* mutations to bedaquiline exposure

To investigate the dynamic of the response to BDQ exposure, the DEGs of two timepoints (0.5 h and 4 h) were investigated by assessing DEGs of the different *Rv0678* mutants vs. wild-type H37Rv (Chapter 3.1.3 Figure 3.10). Based on the data, one hypothesis is that BDQ resistance in *Rv0678* mutants may result from a combination of metabolic changes and stress responses triggered by antibiotic exposure. Transcriptomic analysis of clones with *Rv0678* mutations, in the absence of BDQ exposure, identified seven commonly expressed DEGs, with genes involved in enhanced environmental stress responses, such as oxygen depletion (Chapter 3.1.2 Figure 3.9) (80). To further investigate the transcriptional response to BDQ, expression patterns in *Rv0678* mutants were compared to the ancestral wild-type H37Rv after 0.5 and 4 h of BDQ exposure (Chapter 3.1.3 Figure 3.10).

The DEGs observed in *Rv0678* mutants in response to BDQ exposure align with those seen in studies investigating dormancy conditions and infection models. In these models, harsh conditions such as high pH, oxygen depletion, reactive oxygen species (ROS), and nitric oxide (NO) are examined and trigger a metabolic shift from carbohydrate to lipid metabolism in MTBC bacteria (80,200,201). This switch is driven by changes in gene expression that activate the glyoxylate cycle, which is essential for bacterial persistence (152). A similar response was reported by Koul et al., where BDQ exposure in wild-type H37Rv led to the activation of a dormancy regulon, likely contributing to the delayed bactericidal effect of BDQ (168).

This work found that genes related to metabolic activity and persistence were upregulated in clones with *Rv0678* mutations upon BDQ exposure, suggesting a semi-dormant response of clones with different mutations in *Rv0678* to BDQ. The differential regulation of various functional categories in *Rv0678* mutants is highlighted in the following passage.

4.1.3.1 Expression of immunogenic loci in clones with different *Rv0678* mutations

One of the major transcriptional differences between the different *Rv0678* mutants and the ancestral wild-type H37Rv, as previously described, is the expression of *mpt70* gene with and without drug exposure in all *Rv0678* mutants (Appendix A Table 0.7, Table 0.8). Notably, at 0.5 h and 4 h post BDQ exposure the entire operon, consisting of *mpt70-dipZ-mpt83* is upregulated. The *mpt70* gene encodes

an secreted immunogenic protein, *mpt83* encodes a lipoprotein and *dipZ* encodes an integral membrane protein involved in C-type cytochrome biogenesis (192,202). Homologs of *mpt70* and *mpt83* in *M. bovis* have been extensively studied, and were implicated as virulence factors resulting in an tissue tropism in the ear (203). Other tissue tropism have also been suggested, such as in the lung tissue, given the presence of an specific adhesion domain (203). This operon is regulated by SigK, which plays a key role in sensing environmental redox state (204,205). The upregulation of these genes suggests a SigK-dependent response in these *Rv0678* mutants compared to wild-type H37Rv under BDQ exposure. Although the functional advantage conferred of the expression of these genes in *Rv0678* mutants requires further investigation, the observation of enhanced virulence in strains with increased expression of these immunogenic proteins is noteworthy and could offer insight into the mechanism driving their pathogenicity.

4.1.3.2 Virulence

Virulence factors, typically involved in the pathogen's ability to cause disease by evading host immune responses through growth regulation, sustaining infection, and modulating host-pathogen interactions, may also contribute to drug resistance by enabling prolonged survival under drug-induced stress (206).

Several virulence factors in the *Rv0678* mutants compared to the wild-type were upregulated under 0.5 h BDQ exposure, such as ESAT-6-like protein *esxT* (*Rv3444c*) and two virulence-associated genes (*Rv0662c*, *Rv3922c*) (Chapter 3.1.3 Figure 3.10) (207,208). Conversely, *esxJ* (*Rv1038c*), encoding an ESAT-6 like protein, was downregulated at this timepoint, which could result in growth inhibition and reduces energy consumption as response to antibiotics in the *Rv0678* mutants (209,210). However, further downregulation of the ESX-1 substrates *espF* (*Rv3865*), *espA* (*Rv3616c*), *espD* (*Rv3614c*), involved in immune escape and cell wall stability, after 0.5 h BDQ exposure, may not significantly affect the BDQ resistance of the *Rv0678* mutants (62,209,211–213). Furthermore, two ferredoxin reductases (*fdxA* (*Rv2007c*), *fdxD* (*Rv3503c*)) were downregulated after 0.5 h BDQ exposure in the *Rv0678* mutants compared to the wild-type (Chapter 3.1.3 Figure 3.10) Ferredoxins are cofactors which function as electron transfer partners for cytochrome P450 enzymes, which in turn belong to virulence factors involved in growth and host infection processes (214–216). Thus, downregulation of *fdxA* and *fdxD* cofactors could interrupt this energy pathway and restrain growth (216).

Notably, after 4 h BDQ exposure *esxT* (*Rv3444c*) was downregulated, while other ESAT-6 like transporter system genes, *esxK* (*Rv1197*) and *esxL* (*Rv1198*) of the ESX-5 system, were upregulated in the *Rv0678* mutants. *EsxK* forms an complex with *esxL* (*Rv1198*) and has immunogenic capacities (Chapter 3.1.3 Figure 3.10) (217). Consequently, the gene expression patterns under antibiotic stress observed

in clones with different *Rv0678* mutations are similar to the response of susceptible MTBC described in infection models (207).

The transition between upregulation of *esxT* after 0.5 h BDQ exposure, which is involved in apoptosis by activation of NF- κ B, and its downregulation after 4 h BDQ exposure underlines a dynamic response to antibiotic stress shared in the different *Rv0678* mutants compared to the wild-type (218). Furthermore, the downregulation of ESX-1 substrates and ferredoxin reductases is theorized to save energy under BDQ exposure in *Rv0678* mutants.

4.1.3.3 Cell wall associated transport and cell wall synthesis

The massive, wax-like cell envelope of MTBC strains plays a crucial role in its low permeability to external agents, especially when compared to other gram-positive bacteria (219). Transport across the cell wall is essential for the pathogen's virulence, as it facilitates the export of lipids and cell wall components that contribute to the thickening of the structure (219). Additionally, the active export of toxic by-products and antibiotics enhances resistance to antimicrobial agents (219). One mechanism associated with drug resistance is the increased expression of transporter genes, which can occur due to a malfunction in their negative regulators. For instance, in *Rv0678* mutants, the *mmpS5-mmpL5* efflux pump genes are upregulated under BDQ exposure. This is likely due to the loss of repression, leading to enhanced resistance (183). The downregulation of *iniB* (Rv0341) in *Rv0678* mutants compared to the wild-type 4 h after BDQ exposure is notable as it belongs to a resistance associated operon (Chapter 3.1.3 Figure 3.10, Appendix A Table 0.8). *IniB* is a member of the *iniBAC* operon that confers resistance to cell wall-active antibiotics like INH, as *iniB* is primarily induced by such substances, the observation might just reflect that BDQ targets a different mechanism not triggering the respective operon (220). Furthermore, *IniB* is regulated by the MtrAB two-component system, which responds to various environmental stimuli (221). However, this finding suggests that BDQ exposure does not activate the MtrAB system in *Rv0678* mutants.

Despite the downregulation of *iniB*, other resistance-related genes were upregulated in *Rv0678* mutants compared to the wild-type H37Rv after 0.5 h of BDQ exposure. These resistance-related genes are cell wall-associated transporter genes, such as the ABC transporter family genes *Rv2564*, *Rv2563*, and *irtA* (Chapter 3.1.3 Figure 3.10, Appendix A Table 0.8). The upregulation of *irtA* is particularly interesting, as it forms a complex with *irtB* to import iron, which is critical for survival under conditions such as hypoxia, ROS, NO, and nutrient limitation (208). Additionally, genes related to cell wall transport, such as *arsB2* and *mgtE*, encoding for a magnesium importer, were upregulated in *Rv0678* mutants under 0.5 h BDQ exposure. In *M. smegmatis*, *mgtE* plays a key role in cation homeostasis,

suggesting a similar function in stabilizing the cell wall and maintaining metal homeostasis in response to BDQ exposure (222,223).

Conversely, *Rv1353c*, encoding for a TetR family transcription regulator that represses the efflux pump gene *Rv0191*, was induced after 0.5 h of BDQ exposure, though its precise role in BDQ resistance remains unclear (Chapter 3.1.3 Figure 3.10, Appendix A Table 0.8) (224). Additionally, ABC transporter genes *Rv1686c* and *Rv1687c*, which are part of a multidrug efflux pump system, were downregulated at both 0.5 h and 4 h. Despite this, their downregulation does not appear to impact the MIC for BDQ, indicating that these transporters may not be critical for BDQ resistance of *Rv0678* mutants (208,225).

Lastly, the resuscitation-promoting factor gene *rpjA* (*Rv0867c*) was consistently upregulated under BDQ exposure in *Rv0678* mutants by about 2-fold compared to the wild-type. Resuscitation factors are involved in the reactivation of dormant cells, and the *rpj* family is expressed during different growth stages and stress conditions. The upregulation of *rpjA* reflects an important mechanism supporting resistance, as it enables the reactivation of dormant, non-replicating cells (80).

In conclusion, the cell envelope and its associated transport systems play a critical role in the ability of the *Rv0678* mutants to resist external stress, including antibiotics like BDQ. The upregulation of genes involved in iron import, metal homeostasis, and resuscitation of dormant cells, despite the downregulation of certain efflux pumps, underscores the diverse strategies the clones with diverse *Rv0678* mutations employ to survive certain stress. The understanding of this adaptive responses could facilitate treatment design.

4.1.3.4 Metabolic changes triggered by bedaquiline exposure in *Rv0678* mutants

The metabolic plasticity of *M. tuberculosis* has been widely discussed and is considered to be a key factor in its success, enabling the bacterium to switch between different respiratory cycles and utilize various nutrients in response to external stimuli (67,169,226,227). Two key genes of the methylcitrat cycle, *prpD* and *prpC* (*Rv1130* and *Rv1131*), were upregulated in response to BDQ exposure at 0.5 h and 4 h in *Rv0678* mutants compared to the wild-type (Chapter 3.1.3 Figure 3.10) (69,197,228,229). This upregulation likely facilitates the reduction of propionate toxicity, as suggested by Lee et al. (230). Kundu and Basu describe the induction of the methylcitrat cycle during dormancy, to metabolize acetyl-CoA and propionyl-CoA, which are produced during the metabolism of fatty acids or cholesterol as main carbon source (80). Therefore, it is postulated here that the ATP and electron deprivation caused by BDQ likely triggers this metabolic shift.

Furthermore, after 0.5 h of BDQ exposure, genes related to amino acid metabolism were enhanced in clones with *Rv0678* mutations compared to H37Rv wild-type (Chapter 3.1.3 Figure 3.11). The upregulation of *Rv2989*, an IclR transcription regulator, negatively regulates the *leuCD* operon (Chapter 3.1.3

Figure 3.10, Appendix A Table 0.8). However, after 4 h of BDQ exposure, *Rv2989* vanished and the *leuD* gene was upregulated in the *Rv0678* mutants. *LeuD* encodes alpha-IPM isomerase, an enzyme in the isopropylmalate (IPM) pathway, essential for leucine biosynthesis from acetyl-CoA (231,232). The expression of *leuCD* after 4 h of BDQ exposure indicates activation of the leucine biosynthetic pathway, crucial for survival under oxygen-deprived, nutrient-scarcity, and acidic conditions (231,232). Decreased *leuCD* expression generally leads to a dormancy-like phenotype, as seen in *M. smegmatis*. (232,233). The interaction between the *Rv2989* transcription regulator gene, expressed 0.5 h after BDQ exposure in *Rv0678* mutants, and *leuCD* expression after 4 h, highlights metabolic plasticity.

Metabolic plasticity plays a critical role in the ability of the *Rv0678* mutants to adapt to BDQ treatment. The upregulation of key genes in the methylcitrate cycle, like *prpD* and *prpC* suggest metabolic shift to reduce propionate toxicity and manage alternative carbon sources. Additionally, the dynamic regulation of *Rv2989* transcription regulator and the *leuCD* operon, highlights the ability to modulate leucin biosynthesis in response to BDQ induced stress.

4.1.3.5 Information pathways

Genes involved in information pathways and transcription regulators orchestrate multiple cell processes such as the upregulation of target genes, activation of efflux systems, and modification of drug targets. This extremely efficient response to external stimuli plays a crucial role in the bacterium's success against antibiotics (234). Transcription factors contribute to both drug resistance induced by genetic mutations and to phenotypic drug resistance, which can occur without genetic mutations, enabling MTBC strains to rapidly adapt and evade the effects of antibiotics (234).

After 0.5 h BDQ exposure, seven genes involved in information pathways were upregulated among all clones with *Rv0678* mutations compared to the wild-type, including *spoU* (*Rv3366*), two ribosomal proteins (*rpmB2* (*Rv2058c*) and *rpmF* (*Rv0979A*)), and an anti-anti-sigma factor (*Rv0516c*) (Chapter 3.1.3 Figure 3.10, Appendix A Table 0.8). *SpoU*, encoding a methyltransferase, which methylate RNA and impacts its stability and translation, can critically influence antibiotic response (235,236). Mutations in *spoU* are associated with increased MIC for EMB and RIF (236,237).

The upregulation of several ribosomal protein genes was observed when comparing *Rv0678* mutants to the H37Rv wild-type after 0.5 h of BDQ exposure, suggesting these genes play a role beyond protein synthesis, particularly in stress regulation (238). Typically, under environmental stress, ribosome activity is downregulated due to the high energy demands of protein synthesis (201,238). However, a study on growth rate control in *M. bovis* BCG reported a general downregulation of ribosomal proteins during slow growth, linking ribosomal production to growth rate, as seen in persistence models. In line with this, the study also noted the upregulation of four ribosomal genes: *rpsR2*, *rpsN2*, *rpmG*, and

rpmB2, which are organized in an operon (239). Notably, these same genes, particularly *rpmB2*, were upregulated, in this thesis, in *Rv0678* mutants after 0.5 h of BDQ exposure compared to the wild type. The separation of these genes from other ribosomal genes suggests a distinct role in growth modulation and stress adaptation under antibiotic pressure (239).

Sigma factors are proteins that bind to RNA polymerase and direct it to specific promoters, thereby initiating transcriptional changes in response to the environment. Anti-sigma factors bind to sigma factors and inhibit their activity, while anti-anti sigma factors can release sigma factors from this inhibition. The gene *Rv0516c*, encoding an anti-anti-sigma factor homolog may indirectly enhance the availability of SigF by modulating its inhibition, which is crucial for *M. tuberculosis* survival under stress environment (240–244). As it was shown that *sigF* is also induced by EMB, RIF, streptomycin and cycloserine, the upregulation of its regulatory factor may indicate a protective response against environmental stress, potentially leading to increased resistance in clones with different *Rv0678* mutations.

Conversely, the regulator *trcR* (*Rv1033c*), part of a two-component system with *trcS*, was downregulated 0.5 h after BDQ exposure (245). These systems are crucial for *M. tuberculosis* to respond to environmental changes, though the exact function of *trcR* remains unclear. The impact of *trcR* downregulation on clones with different *Rv0678* mutations requires further study.

Finally, *cysK2* was downregulated after 4 h of BDQ exposure in *Rv0678* mutants compared to the wild-type H37Rv (Chapter 3.1.3 Figure 3.10). This gene is crucial for cysteine synthesis, essential for survival and pathogenesis, particularly through its role in producing mycothiol, which protects *M. tuberculosis* from oxidative stress and harmful agents (246–248). The bacterium has two cysteine biosynthesis pathways, with only one involving *cysK2*, and the activation of these pathways depends on the infection state. The downregulation of *cysK2* in response to BDQ exposure may suggest a shift toward a latent-like state, reflecting the *Rv0678* mutants adaptive strategy to survive under antibiotic pressure (248).

In conclusion, genes involved in information pathways and transcriptional regulation play a critical role in the initial response to BDQ of clones harbouring various *Rv0678* mutations. After just 0.5 h of BDQ exposure, several genes related to growth modulation, stress adaptation, and antibiotic resistance are upregulated, reflecting the ability of *Rv0678* mutants to adapt almost immediately. Changes in ribosomal protein expression and sigma factor regulation indicate a protective response against antibiotic pressure. However, after 4 h BDQ exposure the response diminishes, with a notable downregulation in genes related to cysteine biosynthesis. This suggests a shift towards latent-like state, demonstrating the dynamic response of *Rv0678* mutants to environmental stress.

4.1.3.6 Downregulation of metal and iron storage

MTBC bacteria shut down crucial pathways to evade antimicrobial pressure by conserving energy entering a dormancy-like state (80). In *M. tuberculosis*, as in most organisms, the metals iron, copper, cobalt and nickel are essential nutrients (249–252). Iron, with its ability to transition between oxidation states, is crucial for various cellular processes, including energy metabolism, oxygen transport, gene regulation, DNA biosynthesis, and stress response (74). The bacterium regulates iron availability by producing iron-scavenging siderophores or storage proteins (73,116,188). Copper, serving as co-factor for certain enzymes, also has antibacterial properties and accumulates in infection sites like phagosomes along with zinc (251). Maintaining metal homeostasis is critical to prevent toxicity (253). Nickel and cobalt, sensed by proteins like KmtR, are vital for processes such as vitamin B12 synthesis (252). After 0.5 h BDQ exposure, *Rv0678* mutants compared to the H37Rv wild-type downregulate the iron storage gene bacterioferritin (*bfrB* (*Rv3841*)) and the cooper-homeostasis system genes *csaR* (*Rv0967*), *ctpV* (*Rv0969*) and *mymT* (*Rv0186A*) (Chapter 3.1.3 Figure 3.10, Appendix A Table 0.8). Additionally, the ArsR-SmtB family transcriptional repressor gene *kmtR* (*Rv0827c*) was downregulated 0.5 h after BDQ exposure. Its absence can increase metal exporter gene expression (250). Typically, these factors are important for balancing metal homeostasis to either prevent toxification upon high levels or deprivation when metals like iron are scarce. Downregulation might have an energy-saving purpose for the *Rv0678* mutants or indicate a metal-scarce environment.

4.1.3.7 Regulation of toxin-antitoxin systems and stress response genes

M. tuberculosis exhibits 88 toxin-antitoxin (TA) systems which are categorised in 6 families. These are hypothesised to play a role in pathogenicity and virulence as e.g. the VapBC family members; others were reported to be upregulated in persistent cells (254,255). Generally, it is thought that TA systems have a protective effect during cell stress. In this thesis, members of the Type II TA System families were downregulated 0.5 h after BDQ exposure in *Rv0678* mutants compared to the wild-type, namely the VapBC family (*vapB22* (*Rv2830c*), *vapB28* (*Rv0608*), *vapB36* (*Rv1982A*)), one MazEF family gene (*mazE6* (*Rv1991A*)); additionally, several stress response genes were downregulated including *hspX* (*Rv2031c*), encoding for a heat shock protein) and *Rv2624c*, (encoding for a universal stress family protein) (Chapter Figure 3.10, Appendix A Table 0.8). After 4 h BDQ exposure in clones with different *Rv0678* mutations, further virulence associated TA system genes of Type II family *higB* (*Rv1955*) and *higA* (*Rv1956*) were downregulated. Since *higB* is involved in growth inhibition, downregulation of these TA systems, could also sustain growth under stress conditions in *Rv0678* mutants (256). This suggests that the general stress response system in these *Rv0678* mutants might be damped, allowing sustained growth during stress. However, this requires further investigation.

In conclusion, this analysis outlined in Chapter 3.1.3 on the shared genes up- and downregulated in clones with different *Rv0678* mutations exhibit a multifaceted response to BDQ exposure, which includes both rapid transcriptional plasticity and metabolic flexibility. While these *Rv0678* mutants show enhanced MICs for BDQ, they still demonstrate a significant transcriptional response to the drug action, particularly within 0.5 h of exposure, indicating an immediate adaptation to environmental challenges such as oxidative and nitrosative stress, iron deprivation, copper and zinc toxicity, and acidic pH conditions (67,75,194,201,252). This suggests that *Rv0678* mutants can sustain growth under these harsh conditions, distinguishing them from the wild-type strain H37Rv, which shows a more conventional stress response.

The *mmpS5-mmpL5* efflux pump genes are upregulated in both clones with *Rv0678* mutations and the ancestral wild-type H37Rv under BDQ exposure, hinting at a potential additional resistance mechanism beyond efflux pump upregulation. This hypothesis is further supported by the observations of genes which enhanced expression under BDQ exposure and the diminished transcription reaction after 4 h of exposure. This mechanism may be linked to their enhanced metabolic plasticity, which allows these mutants to shift to a dormant-like state under stress while maintaining some cellular functions. Despite these insights, the exact pathways contributing to BDQ resistance in *Rv0678* mutants must be further explored, as no single mechanism has been definitively identified. The observed transcriptional plasticity, which likely alters drug susceptibility, highlights the potential for adaptive regulatory responses in drug resistance. Nonetheless, the precise molecular and metabolic processes underlying this plasticity, and how they interact with other resistance mechanisms, require further exploration to fully understand the drug resistance phenotype of these mutants.

4.2 The induction of cellular hysteresis effect enhanced sequential treatment on *Mycobacterium tuberculosis*

To address the challenges of antibiotic resistance evolution and the slow development of new treatments, innovative strategies must be explored to improve treatment efficacy and potentially slow down resistance evolution. The current strategy utilizes drug combinations with different modes of action, achieving good outcomes, but also leads to resistance selection even when patients adhere to treatment (87,257,258). Evolution informed treatment strategies, such as different drug cycling approaches have shown promising results for *P. aeruginosa* and *E. coli* (103,110). One such strategy exploits cellular hysteresis, where the short exposure to one antibiotic is enhancing the efficacy of second, potentially circumventing resistance (103).

This work explored for the first time *in vitro* sequential exposure of *M. tuberculosis* using two first-line drugs. This was based on the hypothesis that EMB pre-exposure may facilitate better RIF cell entry, resulting in a negative cellular hysteresis effect. This was confirmed by the experiments performed, when comparing EMB->RIF to no pre-exposure RIF observing a drop in CFU of about 1 log₂FC (Chapter 3.2.2 Figure 3.17). Whereas the reverse sequence of RIF->EMB showed almost identical growth dynamics and no CFU reduction similar to the no pre-exposure EMB sample.

In order to further investigate this cellular hysteresis effect, RNAseq was performed. To understand transcriptomic changes upon sequential exposure, a two-day assay was conducted. DEGs of pre-exposure mono drug (TP1) to 0.5 h after drug switch (TP2) and 48 h after drug switch (TP3) were compared. With this analysis it could be determined if EMB pre-exposure facilitates RIF entry by inducing differential regulation of cell-wall related genes, while RIF pre-exposure might induce solely a stress response, without demising cell wall structure. Additionally, the dynamics of DEGs during this sequential exposure could be determined. By focusing on an supposed metabolic switch as response to this strategy and investigating resistance associated genes induced or not, this study showed the potential to slow down or circumvent resistance evolution (103). The results are discussed in detail in the following chapter.

4.2.1 Ethambutol exposure enhances rifampicin-killing utilizing negative cellular hysteresis in *Mycobacterium tuberculosis*

In this work, the hypothesis that a cell-wall active drug used for pre-exposure could enhance the effect of the second drug by facilitating faster entry into the cell, was tested as detailed in Chapter 3.2.2. To prove this concept, EMB and RIF were evaluated, as they are well-studied antibiotics with distinct mode of action. EMB is active on the cell wall, while RIF inhibits transcription (31,33,34,229). Therefore, an enhanced effect of the RIF main-exposure noticeable in CFU / mL reduction, induced by pre-

exposure with EMB was hypothesized. As described in previous studies performed for other pathogens such effects are potentially drug order dependent.

In a first step, the optimal experimental setup for the hysteresis experiments was defined. Extensive testing identified a suitable concentration of 25 % of the MIC for both drugs, and an 8 h pre-exposure time was selected, as this is below the generation time of the bacteria, to better measure the physiological effect over the genetic effect (Chapter 3.2.1 Figure 3.15, Appendix B Table 0.1) (103).

Sampling was conducted on days 0, 1, 2, ,5, 10 and 14 to capture detailed growth suppression post-drug switch (Chapter 3.2.2 Figure 3.16). From day two onwards, a significant reduction in CFU / mL was observed for the sequence of EMB pre-exposure followed by RIF main-exposure, with sustained reduction of about one log-fold in bacterial growth compared to drug alone and one and half log-fold difference to the reverse sequence (Chapter 3.2.2 Figure 3.17). In concurrence with our hypothesis, this effect was drug-order dependent, as RIF pre-exposure did not enhance the efficacy of subsequent EMB main-exposure. This drug order dependent effect offers valuable insight into drug interactions. While simultaneous administration results in synergy, altering the order can enhance treatment efficacy in one direction but not the other (Chapter 3.2.2 Figure 3.17).

In this work, it was hypothesised that EMB reduces the cell-wall barrier, allowing better RIF entry (104,109,111,259). Previous studies support this hypothesis, showing promising results when cell-wall active substances are used as adjuvant for antibiotics (103,106,260).

The experiments performed here demonstrate for the first time the potential of using sequential drug application to reduce CFU *in vitro* in *M. tuberculosis*. The concept of careful combining drugs for TB treatment based on their interaction profile was investigated by Cokol et al. in their 2017 paper, which focused on identifying the most effective synergistic combinations (100). Their *in vitro* observations highlighted that the most potent synergies were observed between two drugs (BDQ + clofazimine, pretomanid + RIF) rather than four-way combinations (100). Thus, a more effective combination of two drugs might exhibit enhanced potency over a four-drug combination (114,261). However, the risk of resistance evolution with a two-drug combination might be increased, therefore cycling of two efficient drug pairs could be an alternative, and should be further exploited for efficacy and resistance selection (111,261–264). Furthermore, due to the high toxicity risk of long-term treatment of drug resistant TB patients, reducing the number of simultaneously applied drugs is highly valuable (104,158,265).

However, it's important to note that combination therapy could increase the resistant population if complete sterility is not achieved during the early treatment phase (257). Previous studies have shown that due to pharmacokinetic and pharmacodynamics, RIF can negatively impact the uptake of other

antibiotics (45,266). Additionally, observations of constrained resistance evolution upon alternating drug treatments in *S. aureus* support the potential of sequence dependent drug administration (259). Sequential treatment has also shown sustained efficacy at sub-inhibitory concentrations in *E. coli*, where the equivalent two-drug combination therapy was no longer efficient (104).

Highlighting the importance of investigating and understanding the underlying phenomenon of sequential treatments, the first randomized controlled trial on patients with severe tuberculosis meningitis was conducted in India (267). This parallel group randomized controlled trial, was conducted over two years in a single centre. The untreated patients with tuberculosis meningitis were randomized to the standard arm (RIF, INH, EMB and PZA together) or the sequential arm where the same antibiotics were administered but one after another in 1:1 ratio and not all together (267). They reported less drug induced hepatitis (20 % vs. 42.5 %; $p = 0.03$), lower in hospital mortality (2.5 % vs. 17.5 %; $p = 0.025$) and a better 6-month outcome (25 % vs. 50 %; $p = 0.02$), related to the primary endpoint of disability at 3- and 6-month and hepatitis, in the sequential treatment arm compared to standard combination arm (267).

Although the cellular hysteresis effect is still in its early stages of exploration, it should be expanded to other drug combinations, resistant strains, and a genetically diverse set of MTBC strains to determine its robustness (104,106). This study highlights the physiological effect and the significance of drug order in TB treatment design. The careful combination of antibiotics and consideration of pharmacokinetic and pharmacodynamics are crucial for designing new treatment strategies to combat *M. tuberculosis* (100,268,269). An understanding of such phenomena is of great importance to decide if such effects have the potential to sustainably improve treatment strategies in their efficacy but also reducing toxicity and contributing to slow down resistance evolution. Therefore, we investigated a potential induced mechanism with the applied drug order of EMB and RIF using RNAseq.

4.2.2 Transcriptomics landscape of cellular hysteresis in *Mycobacterium tuberculosis*

In the previous section, it was concluded that pre-exposure to EMB enhances the efficacy of RIF through a negative hysteresis effect (Chapter 3.2.2 Figure 3.17). This section delves into the underlying transcriptional dynamics of sequential drug exposure and the hypothesized cellular hysteresis effect by employing RNAseq, a highly precise technology that offers detailed observations of the transcriptome at various timepoints (Chapter 3.2.3).

To investigate this, DEGs were identified at each timepoint in the sequential treatment regimen, comparing them to a no-antibiotic e.g., medium-only control at the corresponding timepoint. Gene func-

tion and categorization were analysed using the DAVID online platform (david.ncifcrf.gov) and mycobrowser (mycobrowser.epfl.ch). Additionally, GO and KEGG pathway analyses were conducted to identify enriched pathways and functional categories.

The analysis focused on several key comparisons (Chapter 3.2.3 Figure 3.18):

- **Pre-exposure effects (TP1):** The gene expression profiles were compared between an 8 h pre-exposure to either EMB or RIF and the medium-only control at the same timepoint. This comparison aimed to capture the transcriptional changes induced by pre-exposure before any antibiotic switch, providing insight into the initial transcriptional landscape.
- **Antibiotic switch effects (TP2):** The gene expression following the switch from EMB to RIF (or vice versa) was compared to the medium-only control, with the medium control undergoing the same centrifugation, medium change, and a 0.5 h incubation at 37° C steps as the switch samples before RNA extraction. This timepoint was designed to reflect the transcriptional response to the antibiotic switch itself. To minimize potential stress signals from centrifugation and medium changes, samples were allowed to rest for 0.5 h prior to RNA extraction.
- **Post-switch effects (TP3):** The impact of the antibiotic switch was further examined 48 h after the switch, a timepoint at which previous *in vitro* assays demonstrated a significant decline in CFU. This comparison aimed to determine whether the transcriptional signature of an EMB-to-RIF switch influences the effectiveness of RIF exposure after 48 h, as the medium remained unchanged post-switch.

The objectives of this analysis were to address the following questions:

- What are the transcriptomic dynamics underlying sequential drug treatments?
- Does the transcriptome support the hypothesis that EMB exposure facilitates subsequent RIF treatment by altering the cell wall?
- Why is the reverse sequence (RIF to EMB) less effective? Due to a stress response induced by RIF?
- How are resistance-associated genes regulated in response to sequential drug exposure?

By answering these questions, this section provides a comprehensive analysis of the transcriptional dynamics across three timepoints in sequential antibiotic exposure, generating a detailed transcriptional landscape of this treatment approach and highlighting the response of *M. tuberculosis* H37Rv to first-line antibiotics (Appendix B Table 0.3, Table 0.4).

4.2.2.1 Negative hysteresis in MTBC bacteria might be driven by cell surface stress response

In MTBC strains, cell wall destabilisation would hypothetically trigger a cell surface stress response. *M. tuberculosis* can sense cell surface stress for example via two-component systems (TCSs) as well as sigma factors and alternative sigma factors (270). One of the major TCSs response networks is regulated by the MprAB signal transduction system in interplay with *sigE* (271). Additionally, the functionally similar and well documented CpxAR two-component system, which senses cell envelop stress in *E. coli* and other gram-negative pathogens, was previously reported to support hysteresis in *P. aeruginosa* (103). Overexpression of the system protects *P. aeruginosa* against negative hysteresis, as a reconstructed *cpxS* mutant did not show negative hysteresis with the same drug sequence which usually induced negative hysteresis the wild-type (103).

Thus, the involvement of MprAB system was investigated during EMB pre-exposure in the experiments outlined in Chapter 4.2.2. MprAB was reported to control a complex regulon of a various genes, including ESX system genes, *sigB*, *SigE* and *hsp* (*Rv0251c*) encoding an hat shock protein (271). Indeed, some of these genes were also significantly ($p_{adj} < 0.05$), differentially regulated in this study (Appendix B Table 0.3, Table 0.4). For example, *sigB* was upregulated 2-fold compared to no-antibiotic after 0.5 h of both drug switches (TP2) (EMB->RIF and RIF->EMB) (Appendix B Table 0.3, Table 0.4). However, *sigE* was not differentially expressed. Furthermore, the heat shock gene *hsp* was upregulated during EMB pre-exposure ($\log_2FC = 1.23$) and 0.5 h after the EMB->RIF switch compared to the respective medium only control ($\log_2FC = 1.44$) (Appendix B Table 0.3). The upregulation of *hsp* indicates an initial response to stress, also observed 0.5 h after the RIF->EMB switch ($\log_2FC = 1.65$) and vanished 48 h post-RIF->EMB switch ($\log_2FC = -1.13$) (Appendix B Table 0.4). Although genes regulated by MprAB were expressed, the MprAB genes themselves were not among the significantly DEGs in response to the pre-exposure or drug switches in this study. One explanation might be the upregulation of *dnaK* (*Rv0350*) 0.5 h after both switches compared to the unexposed H37Rv ($\log_2FC = 1.53$ for EMB->RIF, $\log_2FC = 1.20$ for RIF->EMB), as its overexpression might inhibit MprAB signalling (Appendix B Table 0.7, Table 0.9) (271).

In conclusion, while the study provides evidence that EMB pre-exposure followed by RIF exposure induces hysteresis through cell wall destabilization and triggers a stress response mediated by regulatory systems such as MprAB, the specific involvement of MprAB itself remains unclear. The observed differential regulation of stress response genes, such as *sigB* and *hsp*, supports the hypothesis of a stress response, but the lack of significant changes in MprAB expression suggests that other factors, such as the upregulation of *dnaK* chaperon stress response systems, may modulate the observed effects (272). Further investigation into these regulatory networks will be essential to comprehensively elucidate the

mechanisms underlying negative hysteresis in *M. tuberculosis*. To further investigate the differences in these stress responses between EMB->RIF induced hysteresis and the RIF->EMB sequence, the dynamics of transcriptional response to sequential treatment was analysed and compared between the two sequences at three timepoints (Chapter 3.2.3).

Ethambutol-only exposure induced cell wall stress

The transcriptomic response to EMB 8 h exposure revealed upregulation of genes involved in cell wall repair and stress responses as summarised in the following paragraphs (Chapter 3.2.3 Figure 3.22, Appendix B Table 0.3). GO enrichment analysis of EMB pre-exposure DEGs, identified a cluster of upregulated genes involved in cell wall organization, biogenesis, and degradation, including *murE*, *murG*, and *Rv2719c* (Appendix B Table 0.5). Conversely, downregulated genes were primarily associated with DNA transposition and molecular functions like transposase activity, exemplified by *IS1081* (*Rv2512c*) (Appendix B Table 0.6). The Mur enzymes (*murE* and *murG*), crucial for peptidoglycan biosynthesis, showed increased expression ($> 1 \log_2\text{FC}$) (Appendix B Table 0.3) (273,274). Upregulation of the Mur enzyme genes involved in peptidoglycan biosynthesis might be a response to cell wall destabilizing activity of EMB. EMB is thought to inhibit arabinogalactan and lipoarabinomannan (LAM), by inhibition of arabinosyltransferases which are required for arabinogalactan biosynthesis, it was further postulated that EMB also inhibits *muri* (*Rv1338*) which is involved in peptidoglycan biosynthesis (274–276).

Next, cell wall related pathways under EMB exposure compared to no antibiotic exposure were examined through KEGG enrichment analysis (Appendix B Table 0.7). The upregulation of genes such as *cobT* (*Rv2207*), *cobS* (*Rv2208*), and *hemY* (*Rv2677c*) indicated enrichment in porphyrin metabolism (Appendix B Table 0.7). Porphyrins, or tetrapyrroles, are organic compounds that function as pigments and are widespread in nature. In *M. tuberculosis*, notable porphyrins include heme, which also imparts color to blood, and cobalamin (vitamin B12) (277). These compounds play crucial roles in cellular processes, respiratory functions and have been linked to cell wall structure in dormant cells (278,279). Shleeva et al. reported that the transition of *M. tuberculosis* to a dormant state was associated with the activation of tetrapyrrole metabolism. Their laser scanning microscopy studies revealed the accumulation of free fluorescent tetrapyrrole compounds involved in early heme synthesis, such as *hemY* (*Rv2677c*), and in cobalamin biosynthesis, such as *cobT* (*Rv2207*) and *cobS* (*Rv2208*). These findings suggest a shift towards a dormancy-like state under EMB exposure (80). To further substantiate this observation, experiment which including live / dead staining or acridine orange staining to differentiate dead and viable but non-growing cells under EMB exposure are necessary.

Further supporting the response to EMB-induced cell wall damage is the enhanced *iniB* (*Rv0341*) expression over no-antibiotic stimulation ($\log_2FC$ of = 1.56), a member of the *iniBAC* operon, which is known to be activated by cell wall-targeting antibiotics like INH and EMB and involved in resistance against them (Chapter 3.2.3 Figure 3.22) (220,280). However, *iniA* involved in efflux pump regulation, was not significantly expressed under EMB-exposure. Additionally, the *iniBAC* operon was suggested to be associated with enhanced cell-wall permeability and stabilization, thus *iniB* expression alone could indicate a defensive response to the cell wall destabilization when treated with EMB (280).

In addition to cell wall related pathways and structural biosynthesis, the observed downregulation under EMB exposure of *pitB* (*Rv2281*), an inorganic phosphate transport permease ($\log_2FC$ of -1.55), likely contributes to the effectiveness of EMB. Phosphate is essential for cellular functions, including energy metabolism and cell wall biosynthesis (138). Even with compensatory transporters, such as genes of the phosphate-specific transporter (*pst*) group, impaired phosphate uptake could influence the cell wall repair mechanism (282).

Moreover, significant upregulation ($\log_2FC > 1$, $padj < 0.01$) of cell wall-related virulence factors was observed, including *esxG* (*Rv0287*) from the ESX-3 system, ABC transporter *Rv1463*, and PE_PGRS family proteins in the EMB exposed sample compared to no-exposure (16,80,283–285). The co-expression of PE_PGRS and secretion system subunits aligns with previous studies showing their involvement in exporting PE_PGRS components (286). These genes are typically induced during infection conditions, for immune escape (287). The observations in this study in combination with previous reports suggests that this gene set might be part of a broader stress response system, triggered by conditions observed in infection models, and antibiotic stress, here EMB (286).

The overall gene expression pattern of genes like *murE* and *murG* involved in peptidoglycan biosynthesis suppose the activation of a cell wall repair mechanism and the observed upregulation of genes involved in biosynthesis of porphyrin-like heme and cobalamin (vitamin B12) suggest the induction of a dormancy-like state under EMB exposure compared to no antibiotic exposure. Furthermore, the expression of *iniB* a gene of the *iniBAC* operon, which induces resistance to cell surface active antibiotics and the upregulation of cell wall-related virulence factors indicates a response of *M. tuberculosis* to the destabilisation of its cell wall by EMB. The regulation of *M. tuberculosis* H37Rv transcriptome under EMB exposure is detailed in Chapter 3.2.3 and Appendix B Table 0.3.

Metabolic remodelling and *dank* mediated stress response upon drug switch between ethambutol and rifampicin in *M. tuberculosis*

To understand the direct response of *M. tuberculosis* H37Rv to the drug switch, gene regulation 0.5 h after the drug switch EMB->RIF (TP2) was investigated in more detail in Chapter 3.2.3 Table 3.3 and

Appendix B Table 0.3 and is discussed in this paragraph. In contrast to the upregulation observed following EMB exposure, genes of the *iniBAC* operon were downregulated 0.5 h after exposure to the second antibiotic, RIF compared to the unexposed *M. tuberculosis* H37Rv at the same timepoint (in the EMB->RIF sequence). This downregulation may contribute to the effectiveness of the EMB->RIF sequence, as the *iniBAC* operon genes are associated with resistance to EMB and their reduced expression suggests a decreased defensive response against cell wall-active antibiotics.

Furthermore, several metabolism and respiration-related genes were significantly upregulated ($\log_2FC > 1$ and $p_{adj} < 0.05$) (Chapter 3.2.3 Figure 3.22, Appendix B Table 0.3). Notably, among these were *prpD* (Rv1130), *prpC* (Rv1131), *prpR* (Rv1129c), *trxB1* (Rv1471) and genes of the *mymA* operon (Rv3083-Rv3089) such as *Rv3085*, *chp* (Rv3088) and *fadD13* (Rv3089). The upregulation of *prpD* and *prpC* in *M. tuberculosis* after the EMB->RIF switch, two genes of the methylcitrate cycle, along with *prpR* which encodes their transcription regulator, suggests a metabolic response to detoxify propionyl-CoA, which is generated during β -oxidation of odd-chain fatty acids. This, potentially reduces propionate toxicity during antibiotic exposure rather than being a response to starvation or dormancy as it has been reported under these conditions as well (71,230,288,289). The expression of *mymA* operon, essential for mycolic acids synthesis and cell wall repair, probably reflects an effort to restore cell wall integrity further highlighting cell wall integrity changes in *M. tuberculosis* 0.5 h post drug switch (290).

The significant upregulation of *trxB1* (Rv1471), an antioxidant enzyme, and *Rv1816* ($\log_2FC = 3.24$), a member of the TetR regulator family involved in lipid transport, in the sample after the EMB-to-RIF switch compared to the unexposed sample, supports the presence of an ongoing oxidative stress response and cell wall destabilization 0.5 h post-antibiotic switch (Appendix B Table 0.3) (199). This observation aligns with previously noted metabolic responses following the EMB-to-RIF drug switch

Additionally conducted GO analysis on upregulated DEGs after the EMB->RIF switch revealed clusters related to heat response, stress response, protein refolding, as well as chaperone-dependent protein refolding. The observed clusters include genes such as *dnaJ2*, *dnaJ1*, *dnaK*, *sigB*, *hsp*, and *hspX*, all of which play critical role in preventing accumulation of damaged proteins and folding nascent proteins under stress (Appendix Table 0.5) (270,293). This observation is in concordance with the hypothesis about an additional stress response system to MprAB, induced during antibiotic stress in *M. tuberculosis* (270). The upregulation of these genes, which are typically elevated in response to the accumulation of toxic protein aggregates caused by stress, suggests a robust protein quality control mechanism in relation to antibiotic stress (272). Such mechanisms in turn would need longer treatment duration until the stress mechanism can't counter the antibiotic exposure, but this prolonged exposure might favour resistance evolution which is also observed in persistent or dormant cells. Furthermore, about 4-fold increase of a *dosR* regulon member, *Rv2028c* ($\log_2FC = 2.18$) in *M. tuberculosis* after EMB->RIF

switch compared to no antibiotic was observed. The association of *Rv2028c* with dormancy and hypoxia survival, is notable (Appendix B Table 0.3) (294,295). Although hypoxia is unlikely in this experimental setup, the induction of dormancy-related genes under antibiotic exposure warrants further investigation, as it might support resistance acquisition and enhance pathogen's survival (80).

In conclusion, the downregulation of *iniBAC* operon genes following 0.5 h post EMB->RIF switch (TP2), combined with the significant upregulation of metabolism, respiration-related genes, and stress response clusters, suggests a multifaceted adaptive response by *M. tuberculosis*. This includes efforts to counteract oxidative stress, maintain cell wall integrity, and manage protein damage. The observed gene expression patterns reflect an adaptive strategy that facilitates the effectiveness of the EMB->RIF sequence but also highlights a potential shift towards dormancy shortly after the drug switch. To understand the extended effect of the sequential exposure of EMB on RIF, the sample was incubated after the switch to RIF 48 h and further investigated.

Induction of *iniBAC* operon and *whiB6* genes 48 h post drug switch ethambutol->rifampicin

The previous section examined gene expression shortly after the drug switch EMB->RIF compared to no exposure, highlighting upregulation of stress response genes mediated by *dnaK*. To further understand the impact of the sequential treatment, the 48 h timepoint following the switch to RIF (EMB->RIF) was investigated, coinciding with a reduction in CFU observed in the in vitro experiments (Chapter 3.2.2 Table 3.3, Appendix B Table 0.3). A key difference at this timepoint was the notable upregulation in *M. tuberculosis* after 48 h post EMB->RIF of the transcription regulator *whiB6* (*Rv3862c*) ($\text{Log}_2\text{FC} = 1.07$) which was not observed in the earlier timepoints and not in any timepoint of the other sequence RIF->EMB (Appendix B Table 0.3). *WhiB6* responds to stress and regulates aerobic and anaerobic metabolism as well as cell division during resuscitation (80,288,296). Its upregulation could trigger metabolic adaptation potentially enhancing the efficacy of RIF, as *whiB6* has the potential to reverse dormancy-like states (83,297).

Contrary to observations 0.5 h after the EMB->RIF switch, 48 h post EMB->RIF, the *iniBAC* operon was significantly upregulated compared to the unexposed sample ($\text{Log}_2\text{FC} > 2$) (Appendix B Table 0.3). This time *iniA*, known to confer tolerance to EMB by regulating efflux pumps, was upregulated. This suggested that resistance to EMB could be induced (220,298,299). However, since EMB is no longer present in the sequential treatment, such observations might indicate resistance selection towards EMB rather than RIF (259). This aspect should be further exploited to see if such treatments could be used to refurbish antibiotics and to slow resistance.

In general, the dynamics of sequential treatment support the theory that EMB->RIF induces negative hysteresis due to cell wall destabilisation from EMB pre-exposure (Appendix B Table 0.3). The expression of stress response genes, like members of the PE_PGRS family, *dnaJ2*, *dnaJ1*, *dnaK*, *sigB*, *hsp*, and *hspX*, validates these findings (Appendix B Table 0.8, Table 0.9). The observation of dormancy-related genes such as *dosR*, *prpD* / *C*, suggests that a dormancy-like state could be induced under antibiotic stress 0.5 h after the antibiotic switch (Appendix B Table 0.3) (83). However, 48 h post EMB->RIF switch, the expression of *whiB6* suggests the reversal of such dormancy-like state, potentially marking this a difference which could enhance effectiveness of EMB->RIF sequence over RIF->EMB (Appendix B Table 3.3). Furthermore, the upregulation of the EMB resistance-conferring *iniBAC* operon 48 h after EMB->RIF, indicates resistance acquisition towards EMB, but not RIF. This sequence might induce resistance to the first antibiotic, thereby protecting the second antibiotic from rapid resistance acquisition.

These observations indicate that pre-exposure to EMB significantly affects the gene expression profile between the two exposure groups, primarily inducing cell wall function-related genes (Appendix B Table 0.5). To fully understand the relationship between this treatment strategy and resistance further genetically investigation of resistance population and their selection need to be conducted.

4.2.2.2 Stress response induced by rifampicin and ethambutol in *M. tuberculosis*

Examining gene expression patterns under EMB pre-exposure compared to the reverse sequence (RIF->EMB) is essential for understanding the differential effectiveness of these treatment sequences.

The molecular mechanisms that make one regimen more effective than the other, could be unravelled by analysing how *M. tuberculosis* responds to each sequence. This approach allows like in the previous section to explore how pre-exposure to RIF might prime the bacterial cells, potentially altering their response to subsequent EMB treatment. The previous section provided insights into the adaptive responses of the bacteria to EMB->RIF sequence, including changes in stress response pathways, cell wall integrity, and metabolic functions that could contribute to enhanced susceptibility or resistance. These observations are compared to the observations under in the RIF->EMB sequence. The data on RIF->EMB sequential exposure analysis compared to the unexposed *M. tuberculosis* at is detailed in Chapter 3.2.3 Table 3.4, Appendix B Table 0.4.

Rifampicin exposure activates primarily stress response

Exposure to RIF-only induced a gene expression similar to stress response pattern observed under EMB exposure, despite their different mode of action. Notably, genes such as *trxB1* (*Rv1471*), *cobT* (*Rv2207*), *mycP3* (*Rv0291*) (encoding a serine protease) and *bioF1* (*Rv1569*) involved in biosynthesis of cofactors, as well as PE-PGRS family genes were significantly upregulated ($\log_2FC > 1$) (Chapter 3.2.3 Table 3.4,

Appendix B Table 0.11) these genes are known to be induced in stressful environments, this observation suggests the induction of a stress response mechanism under RIF exposure (286).

Post rifampicin to ethambutol switch 0.5 h

Half an hour after switching from RIF to EMB, the transcriptome showed a similar response as observed after the EMB->RIF switch. The methylcitrat cycle genes (*prpD*, *prpC*) ($\log_2FC > 2$), their transcription regulator (*Rv1129c*) was significantly upregulated ($\log_2FC$ 3.07) (Appendix B Table 0.4). Furthermore, the resuscitation factor *pfkB* (*Rv2029c*) ($\log_2FC$ 1.75) and genes of the *mymA* operon (*Rv3083*-*Rv3089*), *Rv3085* ($\log_2FC$ 1.08) and *Rv3083* ($\log_2FC$ 1.11) were upregulated.

Both sequences (EMB->RIF and RIF->EMB) induced upregulation of *mymA* operon (Appendix B Table 0.3, Table 0.4). The induction might explain the delay in CFU reduction for both sequences, highlighting again the induction of a dormancy-like state under antibiotic pressure. Stress and protein folding genes (*grpE*, *narX*, *dnaK*, *hsp*, *hspX*, *SigB*), were also upregulated, further validating this observation of a general stress response mechanism of *M. tuberculosis* against antibiotic stress, at least to both EMB and RIF (Appendix B Table 0.12).

Post rifampicin to ethambutol switch 48 h

Most notably only 48 h after RIF->EMB, the *iniA* (*Rv0342*) ($\log_2FC$ 1.64) and *iniC* (*Rv0343*) ($\log_2FC$ 1.33) resistance conferring genes were upregulated, though to a lesser extent than in the reverse sequence (Chapter 3.2.3 Table 3.4, Appendix B Table 0.3). This upregulation could potentially select for resistance to EMB. Furthermore, this observation suggests that *iniBAC* operon might be regulated in response to various stress conditions and also to RIF. In contrast to EMB->RIF, the methylcitrat cycle genes *prpD* and *prpC*, remain significantly upregulated 48 h post-exposure. This either indicates antibiotic stress response or less efficacy of this sequence as it triggers a stress response.

Furthermore, a major contrast between the two sequences was the induction of the phthiocerol dimycocerosate biosynthetic (PDIM) pathway, as evidenced by the upregulation of *ppsC*, *ppsB*, *ppsD* and *ppsE* as well as *drrA*, which encodes the ABC transporter, solely after 48 h RIF->EMB (Appendix B Table 0.13) (300). This observation and the expression of an additional membrane transporter gene *mmpL8*, involved in lipid export, implies the activation of a cell wall stabilisation mechanism. Generally, the PDIM pathway is activated in response to RIF exposure, this suggests in this work the RIF pre-exposure also triggered PDIM pathway activation (300). This observation might help to explain why the RIF->EMB sequence does not exhibit a hysteresis effect compared to EMB->RIF sequence.

These investigations provide valuable insights into transcriptional dynamics under sequential treatment exposure and offer an initial glimpse into the mechanisms underlying cellular hysteresis. However, to fully understand this mechanism and assess its potential in circumventing resistance, further

experiments are essential. This work underscores the role of dynamic metabolism and adaptive strategies in enhancing the pathogen's success, suggesting that targeting these processes could improve treatment efficacy. By highlighting potential targets for intervention, this work contributes to the ongoing effort to refine therapeutic strategies against resistant pathogens.

Conclusion

This thesis aimed to provide a better understanding of the effect *Rv0678* mutations have on BDQ resistance, by analysing the plasticity of the transcriptional responses of isogenic clones with different *Rv0678* mutations in response to their target drug using RNAseq. Detailed insights into the response of *M. tuberculosis* to antibiotics revealed that the traditional “one size fits all” approach in medicine may no longer be applicable (37). Also, the thesis provides for the first-time evidence that negative hysteresis may be a suitable strategy to enhance treatment efficacy for *M. tuberculosis* strains. This shows the necessity to explore novel treatment strategies such as cellular hysteresis, to support slow progress in antibiotic research and to refurbish existing antibiotics.

By examining the landscape of BDQ resistant clones harbouring different *Rv0678* mutations with and without their target drug, valuable insights in resistance profiles and potential weaknesses of drug resistant MTBC strains were provided. This study highlighted the diversity of gene expression profiles among bacterial clones harbouring different mutations within the same gene, showing that a single mutation can significantly alter the transcriptome of these bacterial clones regardless of antibiotic exposure. Investigation of the resistance mechanism of *Rv0678* mutants, characterized by enhanced *mmpS5-mmpL5* efflux pump expression, confirmed the upregulation of these efflux pump genes in both BDQ exposed and unexposed conditions. Interestingly, the ancestral wild-type H37Rv exhibited *mmpS5-mmpL5* efflux pump gene expression at levels comparable to those of the *Rv0678* mutants, suggesting that *Rv0678* mutants may exhibit an additional resistance mechanism beyond efflux pump overexpression.

By applying evolutionary medicine principles, and exploring sequential treatment, it was shown that the phenomenon of negative cellular hysteresis can be successfully applied to *M. tuberculosis*. This provides new opportunities to use old and new drugs in innovative ways which can enhanced treatment efficacy as shown with EMB and RIF. Initial explanations of the underlying mechanism point to a surface stress response mediated by *dnaK* chaperon, but further validation is needed. This study provides a strong foundation for further research in the fight against antibiotic resistance by better understand resistance mechanism of widely spread resistant variants and utilizing alternative treatment approaches with the potential to enhance regimen efficacy. It aims to ultimately inspires further researchers to explore new avenues.

However, these investigations raised additional questions that were not fully addressed. Further investigations should focus on:

- Understanding the regulation of MmpS5-MmpL5 efflux pump system and additional mechanisms involved in resistance of *Rv0678* mutants.

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- Conducting proteomics and Western blot analysis to investigate presence of Rv0678, MmpL5, MmpS5, and other proteins of interest.
 - Exploring the potential of sequential treatment in a competition experiment with mix populations (drug cycling).
 - Using high throughput approaches to investigate cellular hysteresis effect with other antibiotics or adjuvants against *M. tuberculosis*.
 - Explore the inducibility of cellular hysteresis in other MTBC strains.
 - Investigate applicability of sequential treatment in more complex systems (macrophage infection systems for example).

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Abbreviations

BCG	Bacillus Calmette-Guerin
BDQ	Bedaquiline
BSL	Bio Safety Level
CFU	Colony Forming Unit
CFZ	Clofazimine
conc.	Concentration
CTAB	Hexadecyltrimethylammonium bromide
del	Deletion
DNA	Deoxyribonucleic acid
DOTS	Directly Observed Treatment, Short-course
DST	Drug Susceptibility Testing
EMB	Ethambutol
et al	"and others"
FICI	Fractional Inhibitory Concentration
fs	Frameshift
gWT	Genetically wild-type
i.e.	id est "in other word"
indel	Insertion or deletion
INH	Isoniazid
ins	Insertion
LIN	Linezolid
Log ₂ FC	Log ₂ fold change
<i>M. tuberculosis</i>	<i>Mycobacterium tuberculosis</i>
MDR	Multi-drug resistant – against Rifampicin and Isoniazid
MXF	Moxifloxacin
MGIT	Mycobacterium Growth Indicator Tube
MIC	Minimum inhibitory concentration
MSC	Minimum selection concentration
MSW	Mutant selection window
MTBC	<i>Mycobacterium tuberculosis</i> complex
NA	Not applicable
NGS	Next generation sequencing
ns	Not significant
NT	Nucleotide

OD	Optical density
<i>p</i> _{adj}	Adjusted p-value
PAS	Para-aminosalicylic acid
PCR	Polymerase-chain reaction
PRE	Pretomanid
PZA	Pyrazinamide
rcf	Relative centrifugal force
ref.	Reference
REMA	Resazurin Emission Microtiter Assay
RIF	Rifampicin
RNA	Ribonucleic acid
RR	Rifampicin-resistant
rRNA	Ribosomal RNA
RT	Room temperature
SDS	Sodium dodecyl sulphate
SM	Streptomycin
SNP	Single nucleotide polymorphisms
TB	Tuberculosis
WGS	Whole Genome Sequencing
WHO	World Health Organization
XDR	Extensively-drug resistant

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Appendix A: Supplementary Material and extended tables of Results 3.1

Table 0.1: Overview RNA extraction procedure test and quality controls, first experiment.

<i>Sample-ID</i>	<i>RNA extraction procedure</i>	<i>Qubit [ng / μL]</i>	<i>Fragment Analyzer [RQN]</i>
rTMW-37	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.4
rTMW-38	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.7
rTMW-39	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.7
rTMW-40	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.7
rTMW-41	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	7.6	9.8
rTMW-42	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	6.6	10
rTMW-43	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-44	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	5.6	9.9
rTMW-45	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	4	10
rTMW-46	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-47	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	5.18	9.5
rTMW-48	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.4
rTMW-49	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.6
rTMW-50	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.8
rTMW-51	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-52	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-53	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-54	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.6
rTMW-55	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	5.92	10
rTMW-56	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-57	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	4.6	9.8
rTMW-58	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-59	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-60	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-61	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-62	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-63	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-64	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	5.18	10
rTMW-65	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.9
rTMW-66	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-67	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.8
rTMW-68	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-69	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	9.8
rTMW-70	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-71	DNA / RNA Shield™ + Sterile filtration Spin-X tubes®	to low	10
rTMW-37	DNA / RNA Shield™ +TRIzol® 2mL	114	10
rTMW-38	DNA / RNA Shield™ +TRIzol® 2mL	82.8	10
rTMW-39	DNA / RNA Shield™ +TRIzol® 2mL	99.4	9.8
rTMW-40	DNA / RNA Shield™ +TRIzol® 2mL	98.2	9.8
rTMW-41	DNA / RNA shield + Millipore Ultrafree-MC Filter	NA	9.9

<i>Sample-ID</i>	<i>RNA extraction procedure</i>	<i>Qubit [ng / μL]</i>	<i>Fragment Analyzer [RQN]</i>
rTMW-42	DNA / RNA Shield™ +TRIzol® 2mL	116	10
rTMW-43	DNA / RNA Shield™ +TRIzol® 2mL	89.6	10
rTMW-44	DNA / RNA Shield™ +TRIzol® 2mL	86.4	10
rTMW-45	DNA / RNA shield + Millipore Ultrafree-MC Filter	NA	10
rTMW-46	DNA / RNA Shield™ +TRIzol® 2mL	95.4	9.9
rTMW-47	DNA / RNA Shield™ +TRIzol® 2mL	156	9.8
rTMW-48	DNA / RNA Shield™ +TRIzol® 2mL	156	10.9
rTMW-49	DNA / RNA Shield™ +TRIzol® 2mL	162	10
rTMW-50	DNA / RNA Shield™ +TRIzol® 2mL	172	10
rTMW-51	DNA / RNA Shield™ +TRIzol® 2mL	160	10.1
rTMW-52	DNA / RNA Shield™ +TRIzol® 2mL	190	9.9
rTMW-53	DNA / RNA Shield™ +TRIzol® 2mL	190	10
rTMW-54	DNA / RNA Shield™ +TRIzol® 2mL	200	9.8
rTMW-55	DNA / RNA Shield™ +TRIzol® 2mL	194	9.9
rTMW-56	DNA / RNA Shield™ +TRIzol® 2mL	188	9.9
rTMW-57	DNA / RNA Shield™ +TRIzol® 2mL	214	9.9
rTMW-58	DNA / RNA Shield™ +TRIzol® 2mL	208	9.7
rTMW-59	DNA / RNA Shield™ +TRIzol® 2mL	202	9.6
rTMW-60	DNA / RNA Shield™ +TRIzol® 2mL	224	9.6
rTMW-61	DNA / RNA Shield™ +TRIzol® 2mL	232	9.8
rTMW-62	DNA / RNA Shield™ +TRIzol® 2mL	152	9.5
rTMW-63	DNA / RNA Shield™ +TRIzol® 2mL	128	9.6
rTMW-64	DNA / RNA Shield™ +TRIzol® 2mL	122	9.7
rTMW-65	DNA / RNA Shield™ +TRIzol® 2mL	151	9.8
rTMW-66	DNA / RNA Shield™ +TRIzol® 2mL	131	9.4
rTMW-67	DNA / RNA Shield™ +TRIzol® 2mL	131	8
rTMW-68	DNA / RNA Shield™ +TRIzol® 2mL	120	9.6
rTMW-69	DNA / RNA Shield™ +TRIzol® 2mL	125	9.7
rTMW-70	DNA / RNA Shield™ +TRIzol® 2mL	134	9.7
rTMW-71	DNA / RNA Shield™ +TRIzol® 2mL	131	9.2
rTMW-72	DNA / RNA Shield™ +TRIzol® 2mL	148	9.3
rTMW-73	DNA / RNA Shield™ +TRIzol® 2mL	165	10
rTMW-74	DNA / RNA Shield™ +TRIzol® 2mL	161	9.9
rTMW-75	DNA / RNA Shield™ +TRIzol® 2mL	139	9.9
rTMW-76	DNA / RNA Shield™ +TRIzol® 2mL	152	10
rTMW-77	DNA / RNA shield + Millipore Ultrafree-MC Filter	too low / 5.44	10
rTMW-78	DNA / RNA Shield™ +TRIzol® 2mL	181	9.5
rTMW-79	DNA / RNA Shield™ +TRIzol® 2mL	197	9.4
rTMW-80	DNA / RNA Shield™ +TRIzol® 2mL	171	9.3
rTMW-81	DNA / RNA shield + Millipore Ultrafree-MC Filter	too low	10
rTMW-82	DNA / RNA Shield™ +TRIzol® 2mL	191	9.3
rTMW-83	DNA / RNA Shield™ +TRIzol® 2mL	195	9.6
rTMW-84	DNA / RNA Shield™ +TRIzol® 2mL	204	10
rTMW-85	DNA / RNA Shield™ +TRIzol® 2mL	228	9.8

<i>Sample-ID</i>	<i>RNA extraction procedure</i>	<i>Qubit [ng / μL]</i>	<i>Fragment Analyzer [RQN]</i>
rTMW-86	DNA / RNA Shield™ +TRIzol® 2mL	208	9.9
rTMW-87	DNA / RNA shield + Millipore Ultrafree-MC Filter	too low	9.9
rTMW-88	DNA / RNA Shield™ +TRIzol® 2mL	137	9.7
rTMW-89	DNA / RNA Shield™ +TRIzol® 2mL	186	10
rTMW-90	DNA / RNA Shield™ +TRIzol® 2mL	161	9.7
rTMW-91	DNA / RNA Shield™ +TRIzol® 2mL	136	10
rTMW-92	DNA / RNA Shield™ +TRIzol® 2mL	117	10
rTMW-93	DNA / RNA Shield™ +TRIzol® 2mL	121	10
rTMW-94	DNA / RNA Shield™ +TRIzol® 2mL	107	10
rTMW-95	DNA / RNA Shield™ +TRIzol® 2mL	118	10
rTMW-96	DNA / RNA Shield™ +TRIzol® 2mL	90.2	10
rTMW-97	DNA / RNA Shield™ +TRIzol® 2mL	144	9.5
rTMW-98	DNA / RNA Shield™ +TRIzol® 2mL	196	9.5
rTMW-99	DNA / RNA Shield™ +TRIzol® 2mL	185	9.4
rTMW-100	DNA / RNA Shield™ +TRIzol® 2mL	146	10
rTMW-101	DNA / RNA Shield™ +TRIzol® 2mL	178	9.9
rTMW-102	DNA / RNA Shield™ +TRIzol® 2mL	152	10
rTMW-103	DNA / RNA Shield™ +TRIzol® 2mL	148	10
rTMW-104	DNA / RNA Shield™ +TRIzol® 2mL	137	10
rTMW-105	DNA / RNA Shield™ +TRIzol® 2mL	144	10
rTMW-106	DNA / RNA Shield™ +TRIzol® 2mL	105	10
rTMW-107	DNA / RNA Shield™ +TRIzol® 2mL	152	10
rTMW-108	DNA / RNA Shield™ +TRIzol® 2mL	160	9.6
rTMW-109	DNA / RNA Shield™ +TRIzol® 2mL	144	9.6
rTMW-110	DNA / RNA Shield™ +TRIzol® 2mL	182	9.5
rTMW-111	DNA / RNA Shield™ +TRIzol® 2mL	158	9.4
rTMW-112	DNA / RNA Shield™ +TRIzol® 2mL	131	10
rTMW-113	DNA / RNA Shield™ +TRIzol® 2mL	142	9.8
rTMW-114	DNA / RNA Shield™ +TRIzol® 2mL	168	10
rTMW-115	DNA / RNA Shield™ +TRIzol® 2mL	74.8	9.8
rTMW-116	DNA / RNA Shield™ +TRIzol® 2mL	82.8	9.7
rTMW-117	DNA / RNA Shield™ +TRIzol® 2mL	54.6	9.3
rTMW-118	DNA / RNA Shield™ +TRIzol® 2mL	69.2	9.5
rTMW-119	DNA / RNA Shield™ +TRIzol® 2mL	59	9.2
rTMW-120	DNA / RNA Shield™ +TRIzol® 2mL	44	6.8
rTMW-121	DNA / RNA Shield™ +TRIzol® 2mL	51.2	9.1
rTMW-122	DNA / RNA Shield™ +TRIzol® 2mL	68.8	9.6
rTMW-123	DNA / RNA Shield™ +TRIzol® 2mL	64.8	9.6
rTMW-124	DNA / RNA Shield™ +TRIzol® 2mL	50.6	8.5
rTMW-125	DNA / RNA Shield™ +TRIzol® 2mL	53.6	9.6
rTMW-126	DNA / RNA Shield™ +TRIzol® 2mL	40	9.5
rTMW-127	DNA / RNA Shield™ +TRIzol® 2mL	78.4	9.4
rTMW-128	DNA / RNA Shield™ +TRIzol® 2mL	62.8	9.5
rTMW-129	DNA / RNA Shield™ +TRIzol® 2mL	61.2	9.5

<i>Sample-ID</i>	<i>RNA extraction procedure</i>	<i>Qubit [ng / μL]</i>	<i>Fragment Analyzer [RQN]</i>
rTMW-130	DNA / RNA Shield™ +TRIzol® 2mL	102	9.7
rTMW-131	DNA / RNA Shield™ +TRIzol® 2mL	99.2	9.5
rTMW-132	DNA / RNA Shield™ +TRIzol® 2mL	122	9.7
rTMW-133	DNA / RNA Shield™ +TRIzol® 2mL	71.6	9.1
rTMW-134	DNA / RNA Shield™ +TRIzol® 2mL	72.6	9
rTMW-135	DNA / RNA Shield™ +TRIzol® 2mL	108	9.4
rTMW-136	DNA / RNA Shield™ +TRIzol® 2mL	67.8	8.9
rTMW-137	DNA / RNA Shield™ +TRIzol® 2mL	40	9.8
rTMW-138	DNA / RNA Shield™ +TRIzol® 2mL	46	9.8
rTMW-139	DNA / RNA Shield™ +TRIzol® 2mL	44	8.6
rTMW-140	DNA / RNA Shield™ +TRIzol® 2mL	46	9.8
rTMW-141	DNA / RNA Shield™ +TRIzol® 2mL	53.6	10
rTMW-142	DNA / RNA Shield™ +TRIzol® 2mL	65.2	10
rTMW-143	DNA / RNA Shield™ +TRIzol® 2mL	48	10
rTMW-144	DNA / RNA Shield™ +TRIzol® 2mL	46	10

Table 0.2: Quality control of first experiment. Total non-rRNA content and RNA integrity assessed with Fragment Analyzer (RQN) of all four replicates. Grey fields = excluded.

<i>Sample ID</i>	<i>Sample name</i>	<i>Fragment Analyser [RQN]</i>	<i>Non-rRNA(total) [%]</i>
rTMW-5_lib47646_nextseq_nx0203_76bp.bam	WT ctrl 0.5h 1	8.2	93.97
rTMW-6_lib48759_nextseq_nx0209_76bp.bam	WT ctrl 0.5h 2	5.6	96.65
rTMW-7_lib47647_nextseq_nx0203_76bp.bam	WT ctrl 0.5h 3	8.4	93.67
rTMW-8_lib48760_nextseq_nx0209_76bp.bam	WT ctrl 0.5h 4	7.9	97.39
rTMW-1_lib47644_nextseq_nx0203_76bp.bam	WT +BDQ 0.5 h 1	6.6	88.94
rTMW-2_lib47645_nextseq_nx0203_76bp.bam	WT +BDQ 0.5 h 2	6.8	97.97
rTMW-3 --> RQN < 3 = excluded bevor sequencing	WT +BDQ 0.5 h 3	2.9	Not sequenced
rTMW-4_lib48758_nextseq_nx0209_76bp.bam	WT +BDQ 0.5 h 4	9.1	83.92
rTMW-13_lib47650_nextseq_nx0203_76bp.bam	atpE A63P ctrl 0.5h 1	6.2	94.19
rTMW-14_lib48763_nextseq_nx0209_76bp.bam	atpE A63P ctrl 0.5h 2	6.2	81.06
rTMW-15_lib47651_nextseq_nx0203_76bp.bam	atpE A63P ctrl 0.5h 3	6.1	95.21
rTMW-16_lib48764_nextseq_nx0209_76bp.bam	atpE A63P ctrl 0.5h 4	6.6	88.95
rTMW-9_lib47648_nextseq_nx0203_76bp.bam	atpE A63P +BDQ 0.5h 1	7.4	82.76
rTMW-10_lib48761_nextseq_nx0209_76bp.bam	atpE A63P +BDQ 0.5h 2	7.8	98.42
rTMW-11_lib47649_nextseq_nx0203_76bp.bam -->excluded non-rRNA <70 %	atpE A63P +BDQ 0.5h 3	5.6	38.37
rTMW-12_lib48762_nextseq_nx0209_76bp.bam	atpE A63P +BDQ 0.5h 4	6.3	90.30
rTMW-17_lib47652_nextseq_nx0203_76bp.bam	Insertion +BDQ 0.5h 1	6.6	89.63
rTMW-18_lib48765_nextseq_nx0209_76bp.bam	Insertion +BDQ 0.5h 2	9	81.96
rTMW-19_lib47653_nextseq_nx0203_76bp.bam	Insertion +BDQ 0.5h 3	4.1	79.18
rTMW-20_lib48766_nextseq_nx0209_76bp.bam	Insertion +BDQ 0.5h 4	6.7	98.45
rTMW-21_lib47654_nextseq_nx0203_76bp.bam	Insertion ctrl 0.5h 1	6.4	94.91
rTMW-22_lib48767_nextseq_nx0209_76bp.bam	Insertion ctrl 0.5h 2	8.1	96.66
rTMW-23_lib47655_nextseq_nx0203_76bp.bam -->excluded non-rRNA <70 %	Insertion ctrl 0.5h 3	6.6	67.92
rTMW-24_lib48768_nextseq_nx0209_76bp.bam	Insertion ctrl 0.5h 4	7.1	97.88

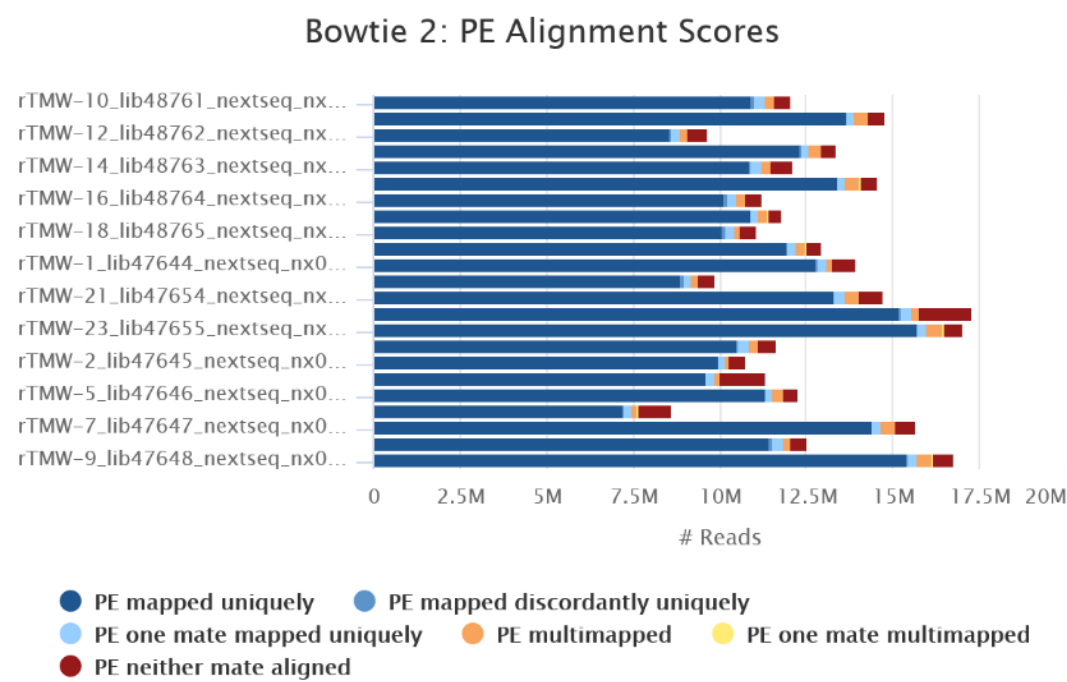


Figure 0.1: Bowtie2 PE alignment scores as quality control of first experiment. Bowtie2 Paired-end (PE) plot of number of reads aligning to reference in different ways as read counts.

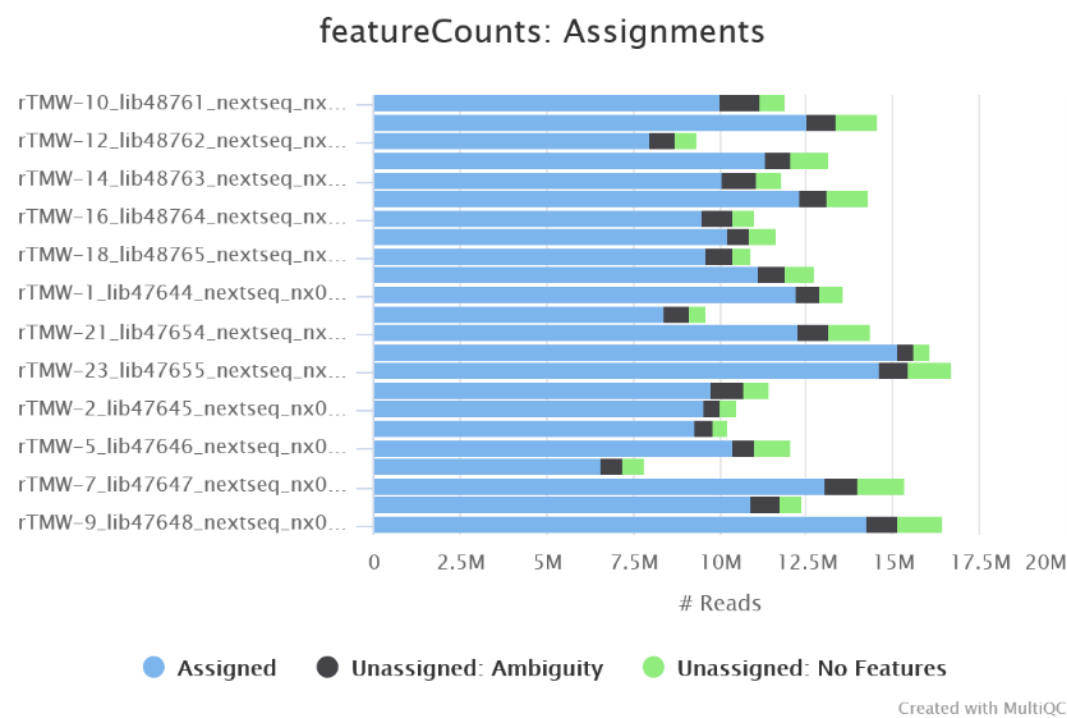


Figure 0.2: Quality control of assignment scores of main experiments. FeatureCounts was used to assign mapped reads with features.

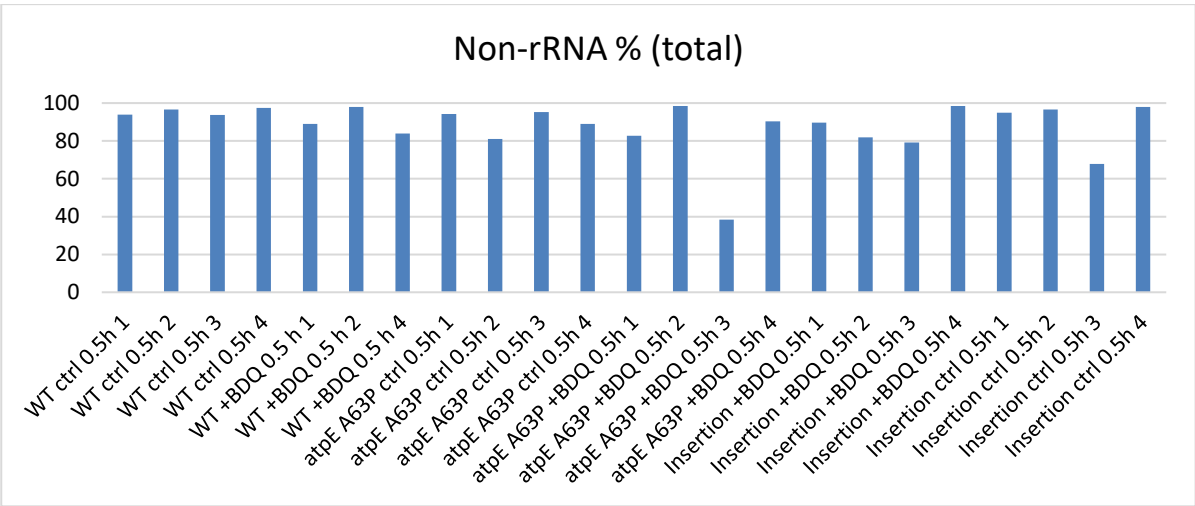


Figure 0.3: Quality control of non-rRNA in the first experiment. Non-RNA percentage was assessed from the featureCount count-table. Threshold of less than 20 % rRNA content selected for downstream analysis.

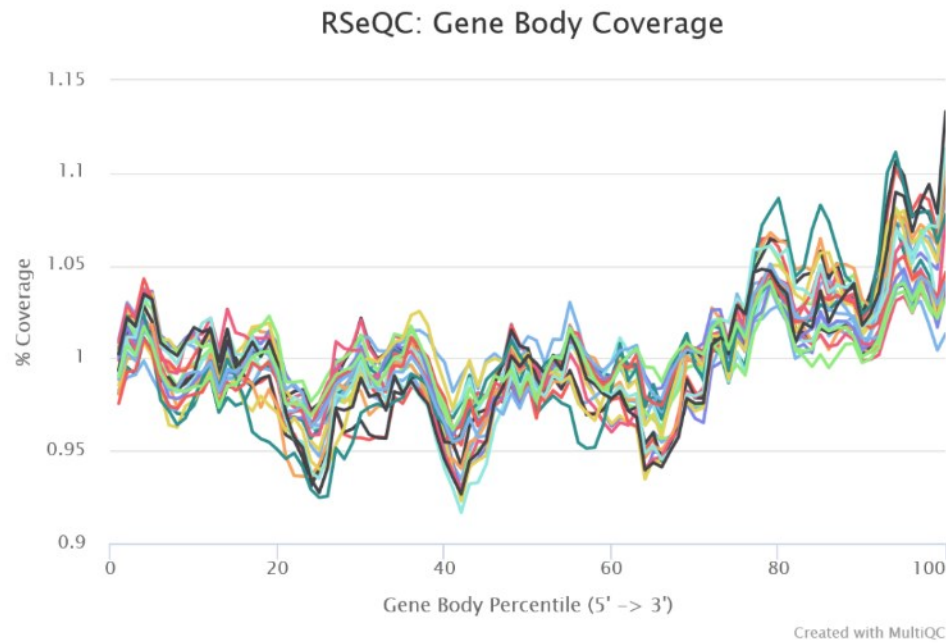


Figure 0.4: Gene body coverage as quality control of first experiment. Gene Body Coverage assessed with RSeQC, uses read coverage over gene bodies to control uniformity of read coverage and check for 5' or 3' bias.

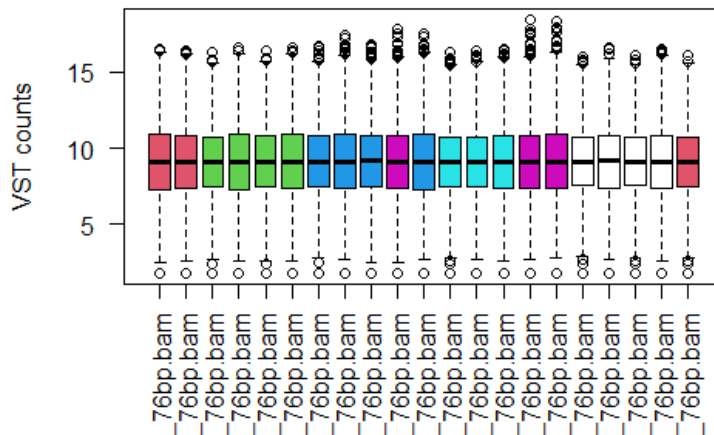


Figure 0.5: Biostatistics quality control using VST counts comparing means of samples of first BDQ experiment.

The variance stabilizing transformation (vst) was used to normalize counts. The means of these VST counts were compared using DESeq. This method was used to verify outliers after DESeq analysis before downstream differentially gene expression (DEG) analysis. Red = *atpE* A63P BDQ+, green = *atpE* A63P BDQ-, blue = *Rv0678* insertion BDQ+, purple = *Rv0678* insertion BDQ-, turquoise = wild-type BDQ+, white = wild-type BDQ-. BDQ+= Bedaquiline exposed, BDQ- = unexposed.

Table 0.3: Quality control main experiment. Main RNA sequencing experiment to assess transcriptomic landscape of BDQ resistant mutants Total non-rRNA percentage and RNA integrity by Fragment Analyzer (RQN). Grey highlighting = excluded, n= 1, four replicates.

<i>Sample-ID</i>	<i>Sample name</i>	<i>Fragment Analyzer [RQN]</i>	<i>Non-rRNA (total) [%]</i>
rTMW-37_lib57742_nextseq_ny0098_101bp.bam	WT ctrl 0.5h 1	10	92.50
rTMW-38_lib57743_nextseq_ny0098_101bp.bam	WT ctrl 0.5h 2	10	90.51
rTMW-39_lib57744_nextseq_ny0098_101bp.bam	WT ctrl 0.5h 3	9.8	88.55
rTMW-40_lib57745_nextseq_ny0098_101bp.bam	WT ctrl 0.5h 4	9.8	91.52
rTMW-42_lib57746_nextseq_ny0098_101bp.bam	WT +BDQ 0.5 h 1	10	87.23
rTMW-43_lib57747_nextseq_ny0098_101bp.bam	WT +BDQ 0.5 h 2	10	85.21
rTMW-44_lib57748_nextseq_ny0098_101bp.bam	WT +BDQ 0.5 h 3	10	81.94
rTMW-46_lib57749_nextseq_ny0098_101bp.bam	WT +BDQ 0.5 h 4	9.9	83.50
rTMW-47_lib57750_nextseq_ny0098_101bp.bam	Invers ctrl 0.5h 1	9.5	95.73
rTMW-48_lib57751_nextseq_ny0098_101bp.bam	Invers ctrl 0.5h 2	9.4	92.89
rTMW-49_lib57752_nextseq_ny0098_101bp.bam	Invers ctrl 0.5h 3	9.6	92.23
rTMW-50_lib57753_nextseq_ny0098_101bp.bam	Invers ctrl 0.5h 4	9.8	96.92
rTMW-52_lib57931_nextseq_ny0098_101bp.bam	Invers +BDQ 0.5h 1	9.9	86.32
rTMW-53_lib57932_nextseq_ny0098_101bp.bam	Invers +BDQ 0.5h 2	10	83.74
rTMW-54_lib57933_nextseq_ny0098_101bp.bam	Invers +BDQ 0.5h 3	9.6	93.63
rTMW-55_lib57934_nextseq_ny0098_101bp.bam	Invers +BDQ 0.5h 4	10	82.62
rTMW-57_lib57935_nextseq_ny0098_101bp.bam	274insA ctrl 0.5h 1	9.9	83.51
rTMW-58_lib57936_nextseq_ny0098_101bp.bam	274insA ctrl 0.5h 2	9.7	92.03
rTMW-59_lib57937_nextseq_ny0098_101bp.bam	274insA ctrl 0.5h 3	9.6	74.79
rTMW-60_lib57938_nextseq_ny0098_101bp.bam	274insA ctrl 0.5h 4	9.6	79.55
rTMW-62_lib57939_nextseq_ny0098_101bp.bam	274insA +BDQ 0.5h 1	9.5	83.37
rTMW-63_lib57940_nextseq_ny0098_101bp.bam	274insA +BDQ 0.5h 2	9.6	77.26
rTMW-64_lib57941_nextseq_ny0098_101bp.bam	274insA +BDQ 0.5h 3	9.7	83.63
rTMW-65_lib57942_nextseq_ny0098_101bp.bam	274insA +BDQ 0.5h 4	9.8	89.02
rTMW-67_lib57943_nextseq_ny0098_101bp.bam	A99V ctrl 0.5h 1	8	85.37
rTMW-68_lib57944_nextseq_ny0098_101bp.bam	A99V ctrl 0.5h 2	9.6	90.71
rTMW-69_lib57945_nextseq_ny0098_101bp.bam	A99V ctrl 0.5h 3	9.7	92.29
rTMW-70_lib57946_nextseq_ny0098_101bp.bam	A99V ctrl 0.5h 4	9.7	89.99
rTMW-72_lib57947_nextseq_ny0098_101bp.bam	A99V +BDQ 0.5h 1	9.3	82.98
rTMW-73_lib57948_nextseq_ny0098_101bp.bam	A99V +BDQ 0.5h 2	10	89.35
rTMW-74_lib57949_nextseq_ny0098_101bp.bam	A99V +BDQ 0.5h 3	9.9	76.96
rTMW-75_lib57950_nextseq_ny0098_101bp.bam	A99V +BDQ 0.5h 4	9.9	83.17
rTMW-78_lib57951_nextseq_ny0098_101bp.bam	atpE A63P ctrl 0.5h 2	9.5	88.06
rTMW-79_lib57952_nextseq_ny0098_101bp.bam	atpE A63P ctrl 0.5h 3	9.4	87.33
rTMW-80_lib57953_nextseq_ny0098_101bp.bam	atpE A63P ctrl 0.5h 4	9.3	83.60
rTMW-82_lib57954_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 0.5h 1	9.3	80.72
rTMW-83_lib57955_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 0.5h 2	9.6	89.04
rTMW-84_lib57956_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 0.5h 3	10	85.54
rTMW-85_lib57957_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 0.5h 4	9.8	82.87
rTMW-88_lib57958_nextseq_ny0098_101bp.bam	WT ctrl 4h 2	9.7	94.51
rTMW-89_lib57959_nextseq_ny0098_101bp.bam	WT ctrl 4h 3	10	90.36

<i>Sample-ID</i>	<i>Sample name</i>	<i>Fragment Analyzer [RQN]</i>	<i>Non-rRNA (total) [%]</i>
rTMW-90_lib57960_nextseq_ny0098_101bp.bam	WT ctrl 4h 4	9.7	95.02
rTMW-91_lib57961_nextseq_ny0098_101bp.bam	WT ctrl 4h 5	10	90.35
rTMW-92_lib57962_nextseq_ny0098_101bp.bam	WT +BDQ 4h 1	10	92.00
rTMW-93_lib57963_nextseq_ny0098_101bp.bam	WT +BDQ 4h 2	10	87.73
rTMW-94_lib57964_nextseq_ny0098_101bp.bam	WT +BDQ 4h 3	10	89.27
rTMW-95_lib57965_nextseq_ny0098_101bp.bam →excluded gene body coverage, sequence depth <8 M reads	WT +BDQ 4h 4	10	86.42
rTMW-97_lib57966_nextseq_ny0098_101bp.bam	Invers ctrl 4h 1	9.5	91.60
rTMW-98_lib57967_nextseq_ny0098_101bp.bam	Invers ctrl 4h 2	9.5	91.10
rTMW-99_lib57968_nextseq_ny0098_101bp.bam	Invers ctrl 4h 3	9.4	86.95
rTMW-100_lib57969_nextseq_ny0098_101bp.bam	Invers ctrl 4h 4	10	92.37
rTMW-102_lib57970_nextseq_ny0098_101bp.bam	Invers +BDQ 4h 1	10	83.15
rTMW-103_lib57971_nextseq_ny0098_101bp.bam →excluded non-rRNA <70 %	Invers +BDQ 4h 2	10	67.16
rTMW-104_lib57972_nextseq_ny0098_101bp.bam	Invers +BDQ 4h 3	10	87.74
rTMW-105_lib57973_nextseq_ny0098_101bp.bam	Invers +BDQ 4h 4	10	95.95
rTMW-107_lib57974_nextseq_ny0098_101bp.bam	274insA ctrl 4h 1	10	89.86
rTMW-108_lib57975_nextseq_ny0098_101bp.bam	274insA ctrl 4h 2	9.6	89.03
rTMW-109_lib57976_nextseq_ny0098_101bp.bam	274insA ctrl 4h 3	9.6	93.33
rTMW-110_lib57977_nextseq_ny0098_101bp.bam	274insA ctrl 4h 4	9.5	87.78
rTMW-112_lib57978_nextseq_ny0098_101bp.bam	274insA +BDQ 4h 1	10	94.50
rTMW-113_lib58286_nextseq_ny0098_101bp.bam	274insA +BDQ 4h 2	9.8	98.50
rTMW-114_lib58287_nextseq_ny0098_101bp.bam	274insA +BDQ 4h 3	10	98.16
rTMW-115_lib58288_nextseq_ny0098_101bp.bam	274insA +BDQ 4h 4	9.8	98.77
rTMW-117_lib58289_nextseq_ny0098_101bp.bam	A99V ctrl 4h 1	9.3	98.65
rTMW-118_lib58290_nextseq_ny0098_101bp.bam	A99V ctrl 4h 2	9.5	96.50
rTMW-119_lib58291_nextseq_ny0098_101bp.bam	A99V ctrl 4h 3	9.2	96.55
rTMW-120_lib58292_nextseq_ny0098_101bp.bam	A99V ctrl 4h 4	6.8	97.84
rTMW-122_lib58293_nextseq_ny0098_101bp.bam	A99V +BDQ 4h 1	9.6	95.95
rTMW-123_lib58294_nextseq_ny0098_101bp.bam	A99V +BDQ 4h 2	9.6	98.60
rTMW-124_lib58295_nextseq_ny0098_101bp.bam	A99V +BDQ 4h 3	8.5	92.88
rTMW-125_lib58296_nextseq_ny0098_101bp.bam	A99V +BDQ 4h 4	9.6	97.49
rTMW-127_lib58297_nextseq_ny0098_101bp.bam	atpE A63P ctrl 4h 1	9.4	96.71
rTMW-128_lib58298_nextseq_ny0098_101bp.bam	atpE A63P ctrl 4h 2	9.5	98.26
rTMW-129_lib58299_nextseq_ny0098_101bp.bam	atpE A63P ctrl 4h 3	9.5	98.24
rTMW-130_lib58300_nextseq_ny0098_101bp.bam	atpE A63P ctrl 4h 4	9.7	98.47
rTMW-132_lib58301_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 4h 1	9.7	98.22
rTMW-133_lib58302_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 4h 2	9.1	98.23
rTMW-134_lib58303_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 4h 3	9	91.48
rTMW-135_lib58304_nextseq_ny0098_101bp.bam	atpE A63P +BDQ 4h 4	9.4	96.19

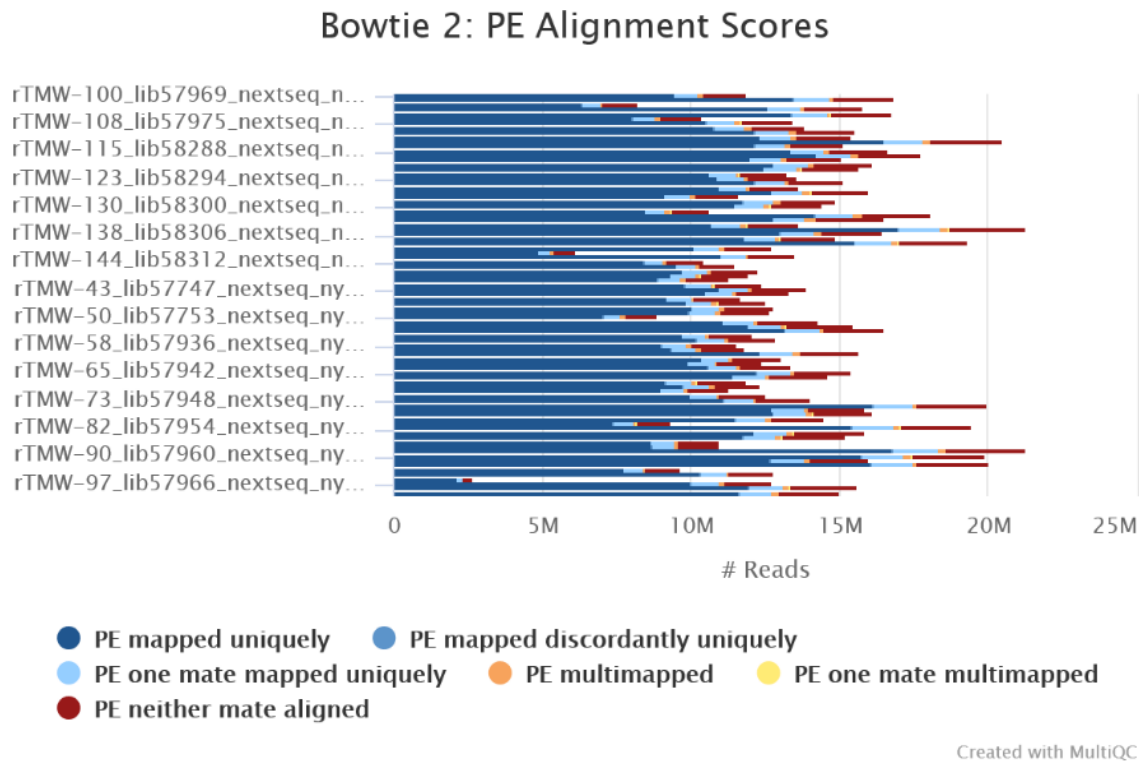


Figure 0.6: Bowtie2 PE alignment scores as quality control of main experiment. Bowtie2 Paired-end (PE) plot of number of reads aligning to reference in different ways as read counts.

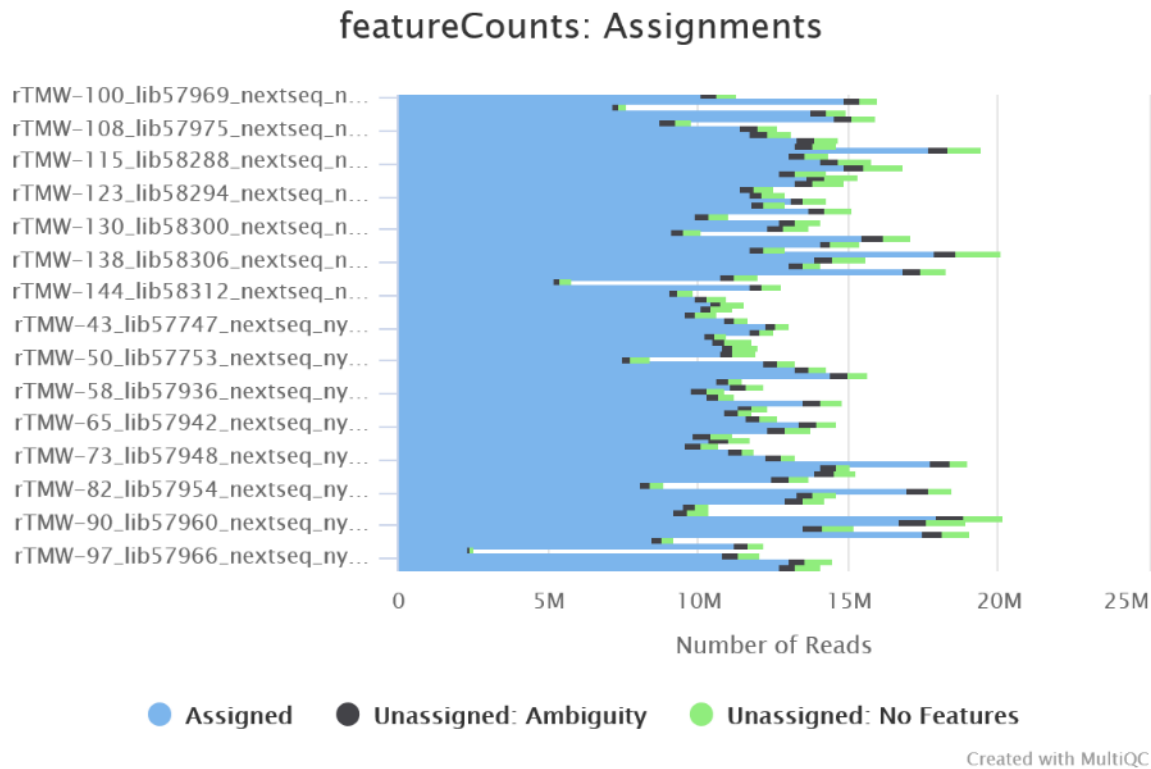


Figure 0.7: Quality control of assignment scores of main-experiment. FeatureCounts was used to assign mapped reads with features.

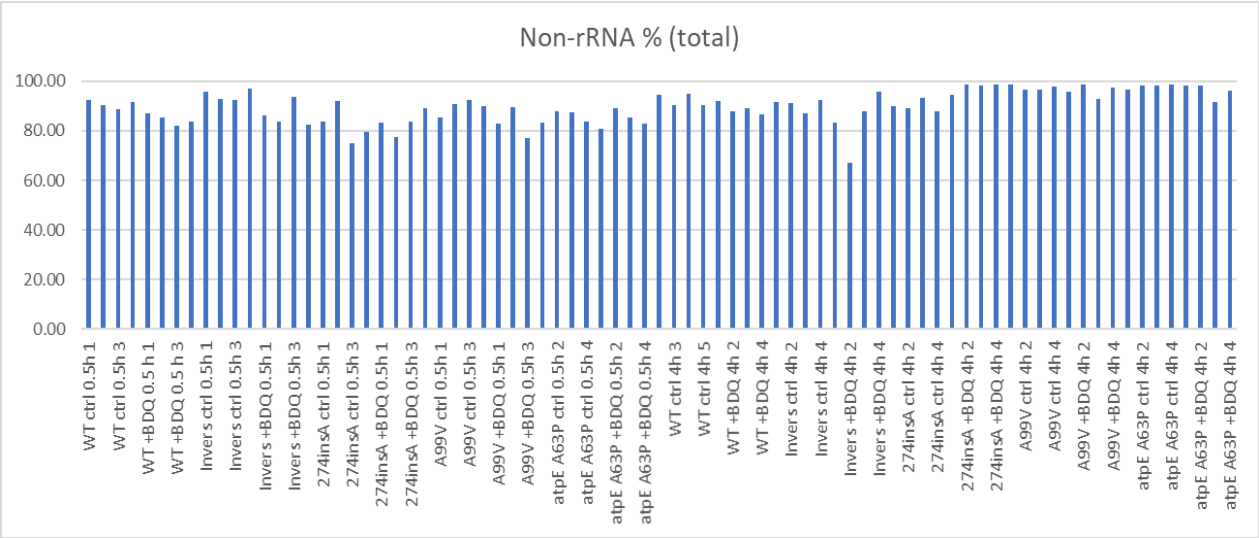


Figure 0.8: Quality control of non-rRNA in the main-experiment. Non-RNA percentage was assessed from the featureCount count-table. Threshold of less than 20 % rRNA content selected for downstream analysis.

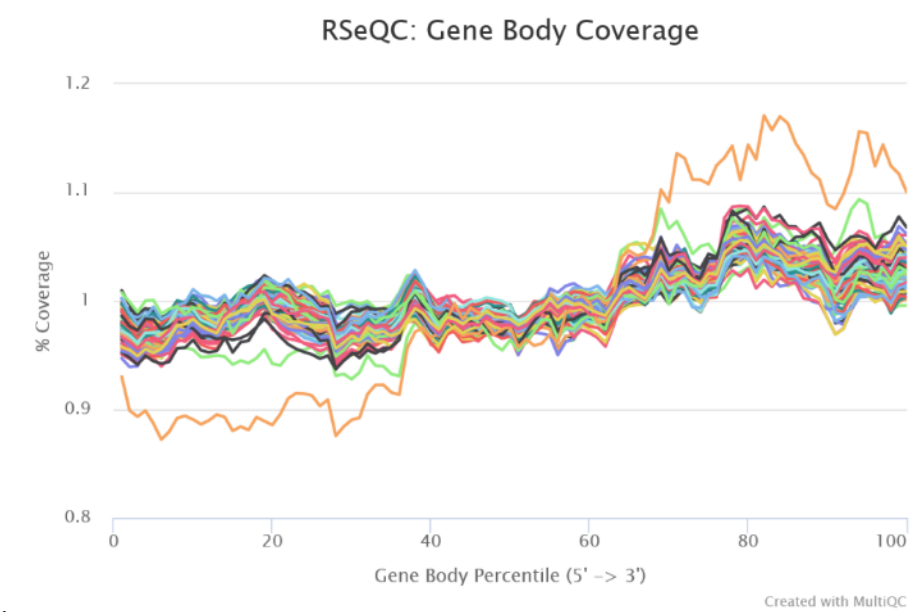


Figure 0.9: Gene body coverage as quality control of main-experiment. Gene Body Coverage assessed with RSeQC, uses read coverage over gene bodies to control uniformity of read coverage and check for 5' or 3' bias.

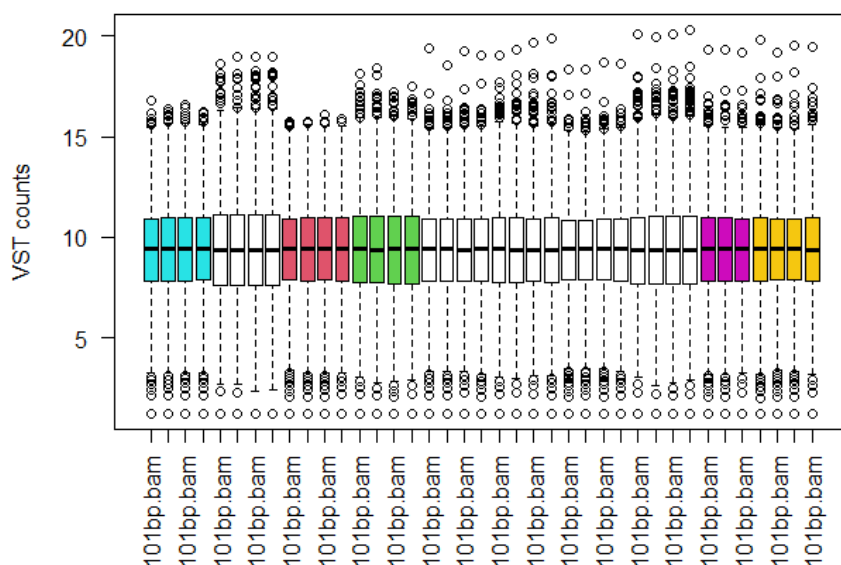


Figure 0.10: Biostatistics quality control using VST counts comparing means of samples of *Rv0678* mutants, *atpE* mutants and wild-type with and without 0.5 h BDQ exposure. The variance stabilizing transformation (vst) was used to normalize counts. The means of theses VST counts were compared using DESeq. This method was used to verify outliers after DESeq analysis before downstream differentially gene expression (DEG) analysis. Blue = *Rv0678* inversion BDQ-, white = *Rv0678* inversion BDQ+, red = wild-type BDQ+, green = wild-type BDQ-, purple = *atpE* A63P BDQ- and yellow = *atpE* A63P BDQ+. BDQ+ = Bedaquiline exposed, BDQ- = unexposed.

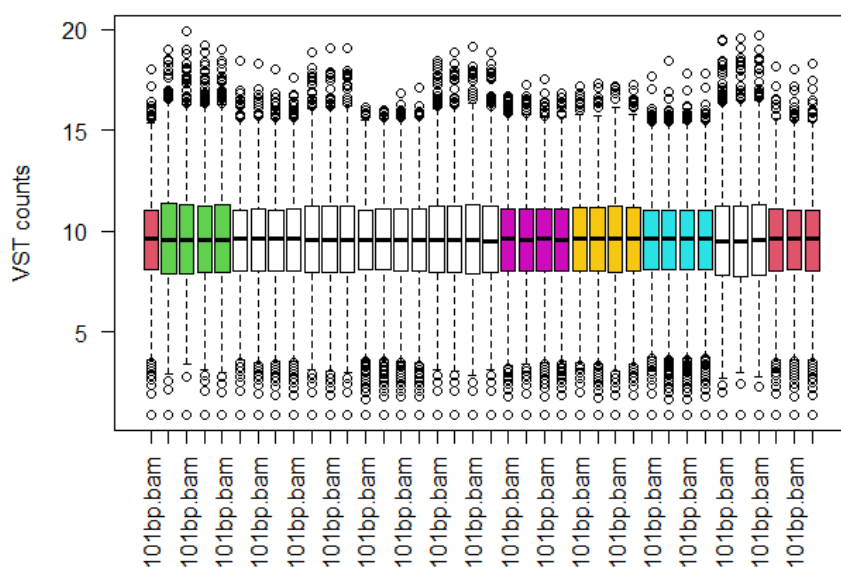


Figure 0.11: Biostatistics quality control using VST counts comparing means of samples of *Rv0678* mutants, *atpE* mutants and wild-type with and without 4 h BDQ exposure. The variance stabilizing transformation (vst) was used to normalize counts. The means of theses VST counts were compared using DESeq. This method was used to verify outliers after DESeq analysis before downstream differentially gene expression (DEG) analysis. Red = *Rv0678* inversion BDQ-, green = *Rv0678* inversion BDQ+, white = wild-type BDQ+, purple = wild-type BDQ-, yellow = *atpE* A63P BDQ- and blue = *atpE* A63P BDQ+. BDQ+ = Bedaquiline exposed, BDQ- = unexposed.

Table 0.4: Supplementary data to Chapter 3.1.1. Efflux pump system *mmpS5 mmpL5* and regulator *mmpR* expression with and without BDQ exposure after 4 h. First the *Rv0678* variants vs. wild-type (WT) without BDQ (BDQ-), then with BDQ exposure (BDQ+) and last each *Rv0678* variant and wild-type exposed vs. unexposed. BDQ 1.2µg / mL (=10xMIC). Log₂FC = Log₂ fold change, *p*_{adj} = adjusted p-value.

4 h <i>Rv0678</i> mutants BDQ- vs. WT BDQ-						
	<i>Rv0676c (mmpL5)</i>		<i>Rv0677c (mmpS5)</i>		<i>Rv0678 (mmpR)</i>	
	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	0.86	8.64E-26	1.04	2.12E-14	0.82	1.26E-08
<i>Rv0678</i> inversion	0.64	5.58E-15	0.61	1.06E-05	0.61	1.53E-05
<i>Rv0678</i> 274insA	0.62	2.85E-14	0.69	2.18E-07	0.77	2.56E-08
<i>AtpE</i> A63P	0.30	3.54E-04	0.44	1.65E-03	0.61	1.53E-05

<i>Rv0678</i> mutants BDQ+ vs. WT BDQ+						
	<i>Rv0676c (mmpL5)</i>		<i>Rv0677c (mmpS5)</i>		<i>Rv0678 (mmpR)</i>	
	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	-0.06	5.71E-01	0.19	2.54E-01	0.43	6.37E-03
<i>Rv0678</i> inversion	0.30	1.16E-03	-0.10	2.74E-01	-3.93	3.09E-167
<i>Rv0678</i> 274insA	-0.23	1.96E-02	-0.29	8.04E-02	-0.40	1.48E-02
<i>AtpE</i> A63P	-2.53	1.07E-198	-2.11	4.92E-53	-1.72	7.15E-34

Mutant BDQ+ vs Mutant BDQ-						
	<i>Rv0676c (mmpL5)</i>		<i>Rv0677c (mmpS5)</i>		<i>Rv0678 (mmpR)</i>	
	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}	Log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	1.98	3.57E-142	1.96	2.99E-53	2.46	6.91E-79
<i>Rv0678</i> inversion	2.56	3.36E-237	2.39	1.51E-78	1.78	5.60E-41
<i>Rv0678</i> 274insA	2.05	6.12E-131	1.83	4.17E-40	1.69	1.44E-32
<i>AtpE</i> A63P	0.07	5.93E-01	0.27	1.26E-01	0.53	1.29E-03
Wild-type (ancestral)	2.90	1.88E-260	2.81	3.37E-93	2.86	6.42E-91

Table 0.5: Supplementary data to Chapter 3.1.1. Efflux pump genes *mmpS5-mmpL5* expression of *Rv0678* mis-sense vs. frameshift mutation, without BDQ exposure after 4 h. The *Rv0678* A99V vs. *Rv0678* 274insA / inversion without BDQ (BDQ-). Log₂FC = Log₂ fold change, *p*_{adj} = adjusted p-value.

4 h <i>Rv0678</i> mutant vs. mutant BDQ-				
	<i>Rv0676c (mmpL5)</i>		<i>Rv0677c (mmpS5)</i>	
	log ₂ FC	<i>p</i> _{adj}	log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V vs. inversion	0.22	1.33E-02	0.43	2.10E-03
<i>Rv0678</i> A99V vs. 274insA	0.24	4.22E-03	0.35	1.24E-02

Table 0.6: *Rv0455c* expression levels in *Rv0678* mutants Supplementary Data to 4.1.1. *Rv0455c* was hypothesized by L. Zhang et al. to activate mmpS5-mmpL5 efflux pumps, thus its expression was investigated in search for additional mmpS5-mmpL5 regulation besides *Rv0678*. *Rv0678* mutants vs. wild-type H37Rv without BDQ (BDQ-), then with BDQ exposure (BDQ+) and last each *Rv0678* variant and wild-type exposed vs. unexposed. BDQ 1.2µg / mL (=10xMIC). Log₂FC = Log₂ fold change, *p*_{adj} = adjusted p-value

<i>Rv0455c</i>	0.5 h BDQ-		4 h BDQ-	
	<i>Rv0455c</i>		<i>Rv0455c</i>	
Mutant vs. wild-type	log ₂ FC	<i>p</i> _{adj}	log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	-0.68	9.81E-15	0.11	5.17E-01
<i>Rv0678</i> ins274A	-0.70	1.95E-15	-0.07	6.09E-01
<i>Rv0678</i> inversion	0.18	6.44E-02	-0.06	7.03E-01
<i>atpE</i> A63P	-0.74	1.48E-14	0.11	4.19E-01

	0.5 h BDQ-		4 h BDQ-	
	<i>Rv0455c</i>		<i>Rv0455c</i>	
Mutant vs. wild-type	log ₂ FC	<i>p</i> _{adj}	log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	-0.46	3.06E-07	0.19	2.37E-01
<i>Rv0678</i> ins274A	-0.46	2.28E-07	0.13	4.45E-01
<i>Rv0678</i> inversion	0.05	5.96E-01	-0.10	5.51E-01
<i>atpE</i> A63P	-0.61	5.96E-12	1.22	1.39E-20

4h	<i>Rv0455c</i>	
Mutant BDQ+ vs. BDQ-	log ₂ FC	<i>p</i> _{adj}
<i>Rv0678</i> A99V	-1.02	8.63E-18
<i>Rv0678</i> ins274A	-0.90	4.16E-12
<i>Rv0678</i> inversion	-1.15	4.75E-21
<i>atpE</i> A63P	0.01	9.70E-01
Wild-type	-1.10	4.82E-17

Table 0.7: Supplementary data to Figure 3.9. DEGs of *Rv0678* mutants against the ancestral wild-type without BDQ exposure, significant up- and downregulated after 0.5 h and 4 h. Significant $\log_2FC > 1$ highlighted in grey, Gene name assessed with david.ncifcrf.gov/. WT = ancestral wild-type, $\log_2FC$ = $\log_2$ fold change, *padj* = adjusted p-value.

BDQ-		0.5 h <i>Rv0678</i> A99V vs. WT		0.5 h <i>Rv0678</i> inver- sion vs. WT		0.5 h <i>Rv0678</i> 274insA vs. WT		4 h <i>Rv0678</i> A99V vs. WT		4 h <i>Rv0678</i> inver- sion vs. WT		4 h <i>Rv0678</i> 274insA vs. WT	
Locus	Gene name	$\log_2$ FC	<i>padj</i>	$\log_2$ FC	<i>padj</i>	$\log_2$ FC	<i>padj</i>	$\log_2$ FC	<i>padj</i>	$\log_2$ FC	<i>padj</i>	$\log_2$ FC	<i>padj</i>
RV0001	chromosomal replication initiator protein DnaA(dnaA)	-0.25	1.39E-02	-0.51	1.01E-06	-1.22	4.08E-35	-0.33	1.56E-02	-1.44	1.44E-33	-1.75	2.08E-49
RV0011C	cell division protein CrgA(Rv0011c)	-0.59	2.72E-10	-0.81	1.30E-17	-1.02	9.91E-28	0.34	3.96E-03	-0.58	1.77E-07	-0.75	3.72E-12
RV0030	hypothetical protein(Rv0030)	-0.42	4.13E-04	-0.29	2.75E-02	-1.18	1.58E-21	-0.02	9.16E-01	-0.45	1.09E-03	-0.83	9.25E-10
RV0033	acyl carrier protein AcpA(acpA)	0.92	1.42E-02	-0.53	2.98E-01	1.19	1.09E-03	0.52	1.30E-01	0.91	2.21E-03	0.55	7.38E-02
RV0040C	hypothetical protein(mtc28)	-0.40	1.83E-05	-0.07	5.70E-01	-1.06	5.79E-30	-0.64	7.16E-09	-0.44	7.58E-05	-0.95	3.14E-19
RV0047C	hypothetical protein(Rv0047c)	-0.32	1.27E-03	-1.19	9.28E-34	-1.06	2.48E-27	-0.20	1.49E-01	-0.86	2.79E-14	-1.04	1.04E-20
RV0076C	membrane protein(Rv0076c)	-0.82	1.07E-04	-0.40	8.87E-02	-1.59	4.00E-12	-0.22	3.08E-01	-0.95	1.88E-07	-1.28	5.08E-12
RV0078A	hypothetical protein(Rv0078A)	-0.73	4.83E-20	-0.17	5.39E-02	-1.13	5.65E-46	0.33	2.65E-02	-0.21	1.47E-01	-0.52	6.40E-05
RV0078B	hypothetical protein(Rv0078B)	-1.79	3.48E-14	0.06	8.60E-01	-2.05	2.19E-18	0.07	8.68E-01	0.27	3.56E-01	0.24	3.93E-01
RV0080	hypothetical protein(Rv0080)	-1.63	1.97E-26	-0.31	7.60E-02	-1.03	1.60E-11	0.17	5.88E-01	0.27	2.84E-01	-0.33	1.56E-01
RV0108C	hypothetical protein(Rv0108c)	-1.18	5.62E-25	-0.89	2.22E-14	-0.90	2.84E-15	-0.20	5.85E-01	-0.16	6.21E-01	-0.12	7.03E-01
RV0119	fatty-acid--CoA ligase FadD7(fadD7)	0.62	1.19E-07	-0.20	1.54E-01	1.06	5.78E-21	-0.36	4.04E-03	-0.03	8.71E-01	0.11	3.94E-01
RV0122	hypothetical protein(Rv0122)	-0.09	6.94E-01	-0.08	7.68E-01	0.21	2.99E-01	-0.33	7.55E-02	0.00	1.00E+00	0.14	4.19E-01
RV0159C	PE family protein PE3(PE3) transcriptional regulator	0.36	7.12E-02	0.07	8.07E-01	-0.36	8.73E-02	-0.26	3.24E-01	-0.42	6.26E-02	-1.06	9.06E-07
RV0165C	Mce1R(mce1R)	-0.34	6.25E-03	-0.71	1.42E-08	-0.94	3.51E-14	-0.69	6.93E-07	-1.13	1.75E-17	-1.52	4.81E-30
RV0179C	lipoprotein LprO(lprO)	-0.36	3.80E-06	-0.13	1.63E-01	-1.26	5.80E-60	0.09	4.17E-01	-0.47	3.30E-08	-0.77	2.94E-21
RV0186A	metallothionein(mymT)	-1.55	4.00E-22	-1.46	3.44E-19	-1.98	3.92E-35	-1.10	1.54E-10	-0.84	8.09E-07	-0.95	8.66E-09
RV0190	hypothetical protein(Rv0190)	-0.84	9.68E-16	-0.25	3.35E-02	-1.49	6.16E-46	0.13	4.04E-01	-0.19	1.55E-01	-0.33	7.94E-03
RV0198C	zinc metalloprotease(zmp1) tRNA (guanine-N(7)-)-methyltrans- ferase(Rv0208c)	0.03	6.69E-01	0.02	8.16E-01	0.19	1.75E-03	0.18	4.09E-02	0.36	2.29E-06	0.35	2.15E-06
RV0208C		-0.19	6.99E-02	-0.15	1.86E-01	-0.90	1.39E-19	-0.55	3.02E-05	-0.91	1.14E-13	-1.17	4.35E-22
RV0219	transmembrane protein(Rv0219)	0.52	8.63E-03	0.23	3.31E-01	1.17	1.63E-10	0.14	5.17E-01	0.33	6.59E-02	0.52	1.97E-03
RV0238	transcriptional regulator(Rv0238)	-0.87	1.08E-29	-0.18	4.01E-02	-1.13	3.91E-48	0.03	8.89E-01	-0.30	3.00E-02	-0.19	1.78E-01
RV0251C	heat shock protein(hsp)	-1.18	1.64E-08	-1.07	9.55E-07	-1.28	7.11E-10	-0.92	1.54E-07	-1.14	7.31E-12	-0.93	1.93E-08
RV0268C	hypothetical protein(Rv0268c)	-1.16	4.24E-35	-0.74	2.03E-14	-1.43	5.79E-52	-0.35	1.13E-01	-0.99	1.49E-07	-1.01	4.26E-08
RV0276	hypothetical protein(Rv0276)	-0.37	2.74E-17	-0.11	1.92E-02	-0.51	4.45E-31	-0.60	2.15E-46	-0.60	2.01E-46	-0.77	8.20E-75
RV0279C	PE-PGRS family protein PE_PGRS4(PE_PGRS4)	1.57	6.60E-08	0.12	7.80E-01	1.71	2.72E-09	-1.04	1.93E-04	0.47	8.80E-02	0.78	2.51E-03

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV0285	PE family protein PE5(PE5)	0.26	8.11E-03	0.27	1.19E-02	0.62	1.00E-10	0.23	1.81E-01	0.68	9.41E-07	1.05	5.31E-15
RV0287	ESAT-6 like protein EsxG(esxG)	0.19	4.23E-02	0.06	6.31E-01	0.99	2.11E-31	0.21	2.10E-01	0.83	1.24E-09	1.30	9.23E-23
RV0288	ESAT-6-like protein EsxH(esxH)	-0.08	3.20E-01	0.03	7.82E-01	0.77	1.83E-27	0.46	1.63E-04	0.82	2.26E-13	1.27	1.41E-31
RV0289	ESX-3 secretion-associated protein EspG3(espG3)	0.93	4.31E-35	0.04	7.19E-01	1.63	1.45E-106	0.09	6.08E-01	0.59	8.68E-07	1.03	1.81E-19
RV0290	ESX-3 secretion system protein EccD(eccD3)	0.98	5.48E-25	0.02	9.06E-01	1.78	3.84E-81	-0.14	3.53E-01	0.67	4.56E-09	0.98	6.05E-19
RV0291	membrane-anchored mycosin MycP(mycP3)	1.03	6.29E-29	-0.03	7.94E-01	1.75	3.90E-82	0.01	9.59E-01	0.46	4.17E-06	0.87	5.48E-20
RV0292	ESX-3 secretion system protein EccE(eccE3)	0.72	9.41E-26	-0.09	3.17E-01	1.11	2.70E-60	0.17	1.84E-01	0.38	4.43E-04	0.39	2.91E-04
RV0312	hypothetical protein(Rv0312)	0.29	1.39E-02	0.02	9.20E-01	-0.64	2.57E-08	-0.35	9.31E-03	-0.63	2.43E-07	-1.17	1.89E-23
RV0313	hypothetical protein(Rv0313)	-0.79	6.96E-07	-0.12	5.63E-01	-1.16	1.19E-13	-0.05	8.87E-01	-0.25	3.35E-01	-0.33	1.82E-01
RV0316	muconolactone isomerase(Rv0316)	1.02	2.46E-31	0.16	1.38E-01	0.87	4.72E-23	-0.10	4.98E-01	0.41	2.07E-04	0.35	9.84E-04
RV0335C	PE family protein PE6(PE6)	-0.29	6.17E-01	-0.30	6.55E-01	-0.26	6.43E-01	-1.45	7.94E-04	-1.40	1.27E-03	-0.57	1.65E-01
RV0340	hypothetical protein(Rv0340)	-1.17	1.37E-22	-0.74	1.75E-09	-1.39	5.98E-31	-0.25	2.08E-01	-0.40	1.86E-02	-0.40	1.53E-02
RV0341	isoniazid inducible protein IniB(iniB)	-0.44	1.44E-06	-1.85	1.94E-96	-1.11	1.15E-35	-0.50	1.26E-06	-1.01	4.20E-26	-0.42	1.41E-05
RV0342	isoniazid inductible protein IniA(iniA)	-0.40	1.68E-10	-0.94	2.88E-51	-1.13	1.20E-73	-0.15	1.35E-02	-0.81	3.04E-57	-0.58	9.75E-30
RV0350	chaperone protein DnaK(dnaK)	-1.45	2.76E-33	-0.15	3.21E-01	-1.06	1.04E-18	-1.18	2.03E-14	-0.02	9.10E-01	-0.32	4.30E-02
RV0351	stress response protein GrpE(grpE)	-1.29	2.80E-31	0.08	5.97E-01	-1.01	5.12E-20	-1.29	1.56E-20	-0.06	7.22E-01	-0.23	1.18E-01
RV0352	chaperone protein DnaJ(dnaJ1)	-1.04	6.68E-23	0.01	9.43E-01	-0.85	9.61E-16	-0.84	4.31E-10	0.03	8.48E-01	-0.14	3.14E-01
RV0354C	PPE family protein PPE7(PPE7)	-0.19	4.92E-01	-0.35	2.27E-01	-0.72	6.83E-03	-0.46	4.90E-02	-1.02	5.72E-06	-1.40	2.62E-09
RV0356C	hypothetical protein(Rv0356c)	0.41	1.61E-03	-0.45	2.32E-03	0.23	8.98E-02	-0.32	1.42E-02	-0.35	4.66E-03	-0.15	2.25E-01
RV0371C	hypothetical protein(Rv0371c)	0.51	1.48E-01	0.00	9.94E-01	1.07	8.01E-04	0.68	6.46E-02	1.04	1.39E-03	0.93	4.38E-03
RV0379	protein translocase subunit SecE(secE2)	-0.73	4.50E-03	0.18	5.68E-01	-0.54	3.71E-02	0.29	4.33E-01	1.15	3.95E-05	0.91	1.01E-03
RV0397	hypothetical protein(Rv0397)	-0.45	5.98E-01	-0.65	5.04E-01	-2.15	3.43E-02	-0.83	4.19E-01	-1.76	7.40E-02	-0.81	3.70E-01
RV0404	long-chain-fatty-acid--AMP ligase FadD30(fadD30)	-0.36	9.14E-07	-0.20	1.36E-02	-1.08	1.52E-49	0.25	1.95E-02	-0.27	6.61E-03	-0.49	1.53E-07
RV0440	molecular chaperone GroEL(groEL2)	-1.50	4.88E-27	-0.79	6.29E-08	-0.56	7.02E-05	-1.16	1.76E-31	0.13	2.79E-01	-0.15	1.80E-01
RV0448C	hypothetical protein(Rv0448c)	0.42	9.13E-02	2.11	4.45E-22	0.17	5.26E-01	0.42	2.53E-01	1.95	2.68E-12	0.24	4.83E-01
RV0450C	transmembrane transport protein MmpL4(mmpL4)	-0.52	8.69E-19	-0.10	1.58E-01	-0.93	4.08E-57	-0.04	6.85E-01	-0.62	6.23E-19	-0.79	2.42E-30

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
	membrane protein												
RV0451C	MmpS4(mmpS4)	-1.01	1.65E-35	-0.48	1.90E-08	-1.51	6.29E-79	0.04	7.35E-01	-0.68	1.48E-15	-0.85	1.36E-24
RV0456A	toxin MazF1(mazF1)	-0.21	2.39E-01	-0.46	1.22E-02	-1.17	1.57E-10	-0.11	6.45E-01	-0.45	1.93E-02	-1.08	3.13E-08
RV0465C	HTH-type transcriptional regulator(Rv0465c)	0.82	3.02E-14	-0.17	2.04E-01	0.18	1.20E-01	-0.40	3.97E-02	-0.73	2.29E-05	-1.08	8.31E-11
RV0469	mycolic acid synthase UmaA(umaA)	-0.69	2.06E-22	-0.43	2.84E-09	-1.03	6.47E-49	0.31	3.83E-05	-0.04	6.35E-01	-0.21	3.13E-03
RV0470C	cyclopropane mycolic acid synthase(pcaA)	-0.86	1.80E-33	-0.28	2.66E-04	-1.11	1.01E-54	0.03	8.67E-01	-0.27	1.79E-02	-0.52	1.71E-06
RV0506	membrane protein												
RV0506	MmpS2(mmpS2)	-0.71	8.89E-12	-0.56	2.04E-07	-1.06	4.93E-24	0.01	9.57E-01	-0.44	3.98E-04	-0.65	5.27E-08
RV0516C	anti-anti-sigma factor(Rv0516c)	0.52	8.70E-04	-0.11	5.74E-01	1.02	9.49E-12	-0.02	9.23E-01	0.14	4.11E-01	0.23	1.41E-01
RV0530A	hypothetical protein(Rv0530A)	-1.24	5.52E-08	-0.32	2.45E-01	-1.15	3.42E-07	-0.15	7.55E-01	0.24	5.39E-01	0.33	3.52E-01
RV0544C	transmembrane protein(Rv0544c)	1.17	3.91E-17	0.08	6.97E-01	1.14	3.70E-16	-0.07	8.28E-01	-0.06	8.45E-01	0.05	8.63E-01
RV0548C	1,4-dihydroxy-2-naphthoyl-CoA synthase(menB)	0.02	7.21E-01	-0.06	3.85E-01	-0.11	4.22E-02	0.14	3.19E-02	0.14	1.81E-02	-0.03	6.23E-01
RV0554	non-heme bromoperoxidase												
RV0554	BpoC(bpoC)	0.85	1.14E-13	0.11	4.58E-01	1.06	6.65E-21	0.11	6.07E-01	0.20	2.77E-01	0.28	9.12E-02
RV0559C	hypothetical protein(Rv0559c)	-0.82	5.66E-26	-0.58	4.82E-13	-1.53	2.21E-82	-0.06	6.49E-01	-0.45	6.67E-07	-0.59	1.11E-11
RV0569	hypothetical protein(Rv0569)	-1.11	2.98E-23	0.16	2.18E-01	-0.13	2.59E-01	-0.30	8.83E-02	0.47	1.93E-03	-0.04	8.17E-01
RV0575C	oxidoreductase(Rv0575c)	-0.86	7.32E-05	-0.99	9.06E-06	0.69	1.06E-03	1.23	1.89E-07	2.18	1.84E-23	4.08	5.10E-85
RV0608	antitoxin VapB28(vapB28)	-0.66	6.27E-14	-0.46	3.87E-07	0.01	8.95E-01	0.18	4.33E-01	0.85	7.87E-07	1.32	1.84E-15
RV0618	pseudo(galT)	1.04	2.32E-11	0.33	7.14E-02	0.94	1.58E-09	-0.23	1.67E-01	-0.01	9.53E-01	-0.24	1.11E-01
RV0649	malonyl CoA-acyl carrier protein												
RV0649	transacylase(fabD2)	1.56	2.00E-02	-0.09	9.34E-01	1.31	5.58E-02	-0.50	6.04E-01	0.57	4.61E-01	1.00	1.42E-01
RV0652	50S ribosomal protein L7/L12(rpL)	1.15	6.98E-27	-0.18	1.61E-01	1.62	1.08E-52	0.12	4.85E-01	0.24	7.30E-02	0.99	1.02E-16
RV0674	hypothetical protein(Rv0674)	0.64	1.86E-08	0.37	3.57E-03	1.17	1.39E-26	-0.48	1.80E-04	0.26	3.81E-02	0.31	8.55E-03
RV0677C	membrane protein												
RV0677C	MmpS5(mmpS5)	0.42	3.48E-14	0.33	1.58E-08	0.27	8.96E-07	1.04	2.12E-14	0.61	1.06E-05	0.69	2.18E-07
RV0678	hypothetical protein(Rv0678)	-0.07	7.21E-01	-3.49	9.39E-103	-0.32	6.03E-02	0.82	1.26E-08	-2.86	7.75E-102	0.77	2.56E-08
RV0742	hypothetical protein(Rv0742)	0.68	8.11E-10	0.08	5.75E-01	1.15	5.90E-27	-0.13	3.90E-01	0.11	4.29E-01	0.57	5.73E-07
RV0744C	transcriptional regulator(Rv0744c)	-1.08	1.36E-20	0.11	4.22E-01	-0.90	6.50E-15	0.25	1.85E-01	0.39	1.42E-02	0.70	3.46E-06
RV0746	PE-PGRS family protein												
RV0746	PE_PGRS9(PE_PGRS9)	0.51	6.56E-04	-0.01	9.66E-01	1.10	8.24E-15	-0.68	3.19E-02	0.95	8.12E-04	1.23	5.99E-06
RV0748	antitoxin VapB31(vapB31)	1.25	8.77E-11	0.14	6.17E-01	1.32	7.02E-12	-0.20	3.85E-01	0.57	1.46E-03	0.58	8.90E-04
RV0749	ribonuclease VapC31(vapC31)	1.08	2.63E-05	0.50	9.67E-02	0.86	1.14E-03	-0.25	4.07E-01	-0.01	9.75E-01	-0.39	1.23E-01
RV0749A	hypothetical protein(Rv0749A)	0.12	4.28E-01	-0.10	5.91E-01	-0.60	7.08E-05	-0.19	3.28E-01	-1.23	3.85E-14	-1.16	8.61E-13

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV0774C	hypothetical protein(Rv0774c)	0.94	8.96E-15	0.11	4.81E-01	1.02	1.97E-17	-0.41	1.46E-02	0.24	1.50E-01	0.20	2.16E-01
RV0787A	hypothetical protein(Rv0787A)	-1.22	4.34E-10	-0.10	7.00E-01	-1.25	9.81E-11	0.19	5.68E-01	0.29	3.00E-01	0.19	4.84E-01
RV0814C	hypothetical protein(Rv0814c)	1.00	2.63E-17	-0.47	3.48E-04	1.34	2.04E-30	-0.32	1.08E-02	0.28	1.59E-02	0.60	2.61E-08
RV0823C	tRNA-dihydrouridine synthase(Rv0823c)	-0.29	2.14E-07	-0.06	3.63E-01	-0.26	3.87E-06	-0.39	4.60E-04	-0.89	2.05E-18	-0.59	5.70E-09
RV0827C	HTH-type transcriptional regulator KmtR(kmtR)	-1.94	1.64E-40	-0.39	1.38E-02	-0.84	5.22E-09	-0.09	8.26E-01	1.36	1.87E-07	1.58	6.10E-10
RV0828C	deaminase(Rv0828c)	0.91	3.29E-02	0.70	1.48E-01	1.06	1.13E-02	0.25	5.42E-01	-1.19	2.06E-03	-0.40	2.83E-01
RV0833	PE-PGRS family protein PE_PGRS13(PE_PGRS13)	0.91	2.69E-03	0.39	2.72E-01	1.35	4.91E-06	-1.24	4.03E-03	0.80	5.65E-02	1.00	1.15E-02
RV0841	transmembrane protein(Rv0841)	1.71	4.54E-02	0.29	8.23E-01	2.58	1.28E-03	1.45	1.31E-01	1.41	1.19E-01	2.27	4.14E-03
RV0846C	oxidase(Rv0846c)	-0.57	4.83E-06	-0.77	1.91E-09	-1.44	9.44E-30	-0.85	1.99E-09	-1.08	5.81E-15	-1.42	1.17E-24
RV0847	lipoprotein LpqS(lpqS)	-0.54	8.78E-06	-1.10	6.86E-20	-1.08	7.59E-20	-1.11	5.33E-17	-0.81	8.39E-10	-1.03	1.84E-15
RV0848	cysteine synthase CysK(cysK2)	-0.72	1.79E-10	-0.55	3.63E-06	-1.25	5.56E-29	-0.80	3.67E-07	-0.53	6.61E-04	-0.94	2.20E-10
RV0849	MFS-type transporter(Rv0849)	-0.57	1.58E-05	-0.57	4.43E-05	-1.15	1.26E-18	-0.90	1.18E-06	-0.74	3.87E-05	-1.36	3.44E-15
RV0856	hypothetical protein(Rv0856)	-1.13	1.94E-26	-0.28	1.66E-02	-1.48	8.39E-44	-0.03	8.70E-01	-0.63	1.92E-06	-0.62	2.46E-06
RV0857	hypothetical protein(Rv0857)	-0.82	2.71E-19	0.11	3.12E-01	-1.15	6.50E-36	-0.13	3.20E-01	-0.58	7.95E-09	-0.63	1.32E-10
RV0885	hypothetical protein(Rv0885)	-1.02	3.28E-33	-0.10	3.16E-01	-0.65	5.92E-15	-0.35	1.87E-05	-0.26	1.20E-03	-0.44	1.78E-08
RV0888	hypothetical protein(Rv0888)	-0.26	1.12E-01	-0.43	1.16E-02	-1.09	3.87E-12	-0.53	2.68E-05	-0.87	2.24E-13	-1.24	7.47E-26
RV0893C	S-adenosylmethionine-dependent methyltransferase(Rv0893c)	-0.35	2.34E-03	-0.08	6.08E-01	-1.00	2.22E-17	0.22	8.51E-02	-0.25	3.29E-02	-0.56	9.97E-07
RV0898C	hypothetical protein(Rv0898c)	-0.95	9.70E-13	0.28	5.96E-02	-0.25	7.05E-02	0.24	2.37E-01	0.97	1.20E-09	1.79	1.41E-31
RV0909	antitoxin(Rv0909)	-1.32	4.68E-22	-0.54	1.62E-04	-1.77	1.48E-37	0.01	9.67E-01	-0.21	3.14E-01	-0.37	5.56E-02
RV0912	transmembrane protein(Rv0912)	-0.49	1.15E-05	-0.33	6.05E-03	-1.41	5.35E-32	-0.10	7.46E-01	-0.55	1.06E-02	-0.99	1.54E-06
RV0913C	dioxygenase(Rv0913c)	-0.74	1.31E-11	-0.33	5.03E-03	-1.07	2.93E-22	-0.21	7.54E-03	-0.57	6.96E-15	-0.82	1.53E-29
RV0919	GCN5-like N-acetyltransferase(Rv0919)	-0.71	7.07E-04	0.18	4.57E-01	-1.23	1.48E-08	-0.05	8.48E-01	-0.52	7.32E-03	-0.88	5.94E-06
RV0942	hypothetical protein(Rv0942)	1.06	5.55E-02	0.49	4.81E-01	1.36	1.03E-02	-0.16	7.86E-01	0.71	8.67E-02	0.88	2.36E-02
RV0967	copper-sensing transcriptional repressor CsoR(csoR)	-1.06	2.02E-24	-0.27	1.79E-02	-1.06	9.22E-25	-0.09	5.24E-01	0.13	2.91E-01	0.20	6.84E-02
RV0968	hypothetical protein(Rv0968)	-1.43	2.38E-22	-0.41	1.22E-02	-1.28	2.55E-18	-0.13	6.07E-01	0.26	1.91E-01	0.18	3.41E-01
RV0974C	acetyl-/propionyl-CoA carboxylase subunit beta(accD2)	0.64	2.82E-06	0.15	3.94E-01	1.00	5.20E-14	0.12	5.41E-01	-0.43	7.74E-03	0.36	1.91E-02
RV0978C	PE-PGRS family protein PE_PGRS17(PE_PGRS17)	0.75	5.90E-03	0.29	4.01E-01	0.69	1.24E-02	0.34	2.83E-01	0.56	3.54E-02	1.05	1.39E-05

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV0980C	PE-PGRS family protein PE_PGRS18(PE_PGRS18)	0.32	2.82E-01	0.07	8.48E-01	0.81	2.85E-03	-0.09	8.14E-01	1.14	3.16E-06	1.06	1.25E-05
RV0991C	hypothetical protein(Rv0991c) transcriptional regulator	-0.88	1.13E-10	-0.42	4.47E-03	-0.87	1.60E-10	-0.82	1.37E-08	-0.49	7.34E-04	-1.14	5.86E-17
RV1027C	KdpE(kdpE)	-0.17	1.97E-01	-0.26	6.02E-02	-0.45	4.11E-04	-0.03	8.64E-01	-0.75	7.23E-12	-1.13	1.48E-22
RV1033C	two component transcriptional regulator TrcR(trcR)	-1.22	5.58E-19	-1.07	1.74E-14	-1.28	1.52E-20	-0.06	9.05E-01	-1.11	2.20E-04	0.09	7.96E-01
RV1042C	IS2-like transposase(Rv1042c)	-1.16	1.03E-19	-0.68	1.95E-07	-1.39	9.61E-28	0.05	8.31E-01	-0.62	1.39E-04	-0.44	5.79E-03
RV1052	hypothetical protein(Rv1052)	-0.88	1.37E-28	0.11	2.52E-01	-1.42	1.16E-65	0.14	3.92E-01	-0.14	3.38E-01	-0.31	1.46E-02
RV1057	hypothetical protein(Rv1057)	0.34	4.82E-04	0.36	3.61E-04	0.41	1.45E-05	0.83	2.47E-16	1.12	5.12E-30	0.89	2.44E-19
RV1067C	PE-PGRS family protein PE_PGRS19(PE_PGRS19)	1.07	1.11E-06	0.22	4.41E-01	1.47	6.86E-12	-0.33	1.39E-01	0.65	4.04E-04	0.83	1.83E-06
RV1073	hypothetical protein(Rv1073)	-0.84	1.23E-08	-0.58	1.70E-04	-1.09	5.07E-14	-0.59	6.43E-06	-0.50	6.27E-05	-0.74	9.06E-10
RV1075C	hypothetical protein(Rv1075c)	-0.77	4.78E-13	-0.34	2.58E-03	-1.32	1.44E-31	0.03	8.72E-01	-0.55	1.99E-05	-0.67	9.59E-08
RV1081C	membrane protein(Rv1081c)	0.20	1.11E-01	0.16	2.63E-01	-0.42	1.49E-03	-0.18	2.98E-01	-0.70	7.79E-07	-1.05	2.02E-13
RV1087	PE-PGRS family protein PE_PGRS21(PE_PGRS21)	0.70	9.07E-05	0.39	5.36E-02	1.05	1.98E-09	-0.94	1.89E-04	0.41	1.06E-01	0.58	1.46E-02
RV1091	PE-PGRS family protein PE_PGRS22(PE_PGRS22)	0.59	2.73E-03	0.23	3.34E-01	1.07	1.70E-08	-0.98	4.19E-04	0.60	2.65E-02	0.87	6.87E-04
RV1120C	hypothetical protein(Rv1120c)	-0.37	3.97E-01	0.57	1.83E-01	-1.15	9.03E-03	0.54	1.41E-01	-0.03	9.55E-01	-0.34	3.91E-01
RV1130	2-methylcitrate dehydratase(prpD)	0.22	2.80E-01	4.05	2.53E-144	1.52	5.03E-19	0.65	3.07E-03	4.41	6.99E-144	1.62	9.79E-18
RV1131	methylcitrate synthase PrpC(prpC)	0.66	6.14E-03	4.22	1.97E-92	1.51	1.09E-11	0.52	3.64E-02	3.86	9.32E-92	1.00	3.77E-06
RV1149	transposase(Rv1149)	-0.72	9.62E-18	-0.01	9.19E-01	-1.03	1.42E-34	0.15	3.57E-01	0.00	9.86E-01	-0.34	8.69E-03
RV1150	pseudo(Rv1150)	0.20	3.34E-01	0.05	8.52E-01	-0.50	1.56E-02	-0.32	1.32E-01	-0.98	4.73E-07	-1.06	4.26E-08
RV1155A	hypothetical protein(Rv1155a)	-1.12	5.58E-12	-0.26	1.83E-01	-0.64	8.97E-05	0.33	3.45E-01	1.02	1.69E-04	1.23	3.00E-06
RV1159A	4a-hydroxytetrahydrobiopterin dehydratase(Rv1159A)	-1.36	2.71E-27	-0.41	2.55E-03	-0.49	1.25E-04	-0.02	9.54E-01	0.26	1.62E-01	0.66	5.30E-05
RV1165	GTP-binding translation elongation factor(typA)	0.16	2.29E-03	0.10	8.87E-02	0.18	2.62E-04	-0.12	7.04E-02	-0.23	9.18E-05	-0.10	1.05E-01
RV1191	hypothetical protein(Rv1191)	1.00	9.83E-17	-0.15	3.52E-01	1.03	9.77E-18	-0.17	2.08E-01	0.41	2.98E-04	0.18	1.30E-01
RV1196	PPE family protein PPE18(PPE18)	0.24	2.16E-02	0.22	5.29E-02	1.08	1.73E-27	-0.05	8.35E-01	0.83	1.24E-08	1.10	1.34E-14
RV1197	ESAT-6 like protein EsxK(esxK)	-0.20	1.26E-01	0.45	5.28E-04	0.85	5.33E-13	-0.10	5.30E-01	0.80	1.76E-11	1.24	7.01E-27
RV1198	ESAT-6 like protein EsxL(esxL)	0.29	6.94E-03	0.29	1.09E-02	1.35	1.68E-39	-0.03	8.30E-01	0.78	1.54E-18	1.24	1.34E-46
RV1214C	PE family protein PE14(PE14)	1.05	3.18E-13	0.21	2.41E-01	1.19	5.40E-17	0.48	1.97E-03	0.41	6.33E-03	0.73	2.54E-07
RV1216C	integral membrane protein(Rv1216c)	-1.04	6.21E-34	0.29	1.15E-03	-0.76	3.88E-19	-0.44	1.19E-03	-0.31	1.91E-02	-0.51	4.66E-05

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV1218C	tetronasin ABC transporter ATP-binding protein(Rv1218c)	-1.03	1.17E-38	0.54	5.14E-12	-0.62	2.74E-15	-0.53	1.15E-04	-0.43	1.12E-03	-0.38	3.78E-03
RV1219C	transcriptional regulator(Rv1219c)	-1.21	3.03E-41	0.23	1.53E-02	-1.24	1.24E-42	-0.46	1.22E-03	-0.70	1.49E-07	-0.61	3.61E-06
RV1227C	transmembrane protein(Rv1227c)	0.77	8.31E-11	0.35	8.36E-03	1.50	8.52E-39	0.27	7.52E-02	0.77	3.55E-09	1.57	4.11E-37
RV1284	beta-carbonic anhydrase(canA)	-1.10	2.30E-13	-0.92	1.36E-09	-1.11	2.69E-13	-1.13	1.28E-17	-1.09	3.88E-16	-0.94	1.26E-12
RV1285	sulfate adenylyltransferase subunit 2(cysD)	-1.58	6.03E-61	-0.72	1.83E-13	-1.40	2.27E-48	-0.94	4.64E-16	-0.92	1.19E-15	-0.26	3.32E-02
RV1286	adenylyl-sulfate kinase(Rv1286)	-1.76	5.76E-94	-0.77	6.88E-19	-1.82	1.33E-97	-1.14	1.07E-17	-1.28	8.83E-23	-1.36	6.50E-26
RV1325C	PE-PGRS family protein PE_PGRS24(PE_PGRS24)	1.30	5.47E-13	0.35	1.03E-01	1.54	5.65E-18	-0.57	1.15E-02	0.74	2.76E-04	0.96	8.60E-07
RV1348	iron ABC transporter ATP-binding protein/permease IrtA(irtA)	0.49	5.03E-09	0.56	2.70E-11	1.14	7.86E-45	0.45	2.05E-03	0.29	4.07E-02	0.53	6.36E-05
RV1349	iron ABC transporter ATP-binding protein/permease IrtB(irtB)	0.70	1.72E-17	0.24	9.43E-03	1.31	4.16E-61	0.48	4.65E-04	0.43	1.09E-03	0.70	1.54E-08
RV1352	hypothetical protein(Rv1352)	-1.11	8.88E-21	0.09	5.74E-01	-0.87	2.18E-13	-0.12	6.17E-01	0.28	1.39E-01	0.50	3.19E-03
RV1365C	anti-sigma-F factor antagonist RsfA(rsfA)	-0.27	4.49E-02	-0.18	2.39E-01	-0.38	4.17E-03	-0.55	1.24E-04	-0.98	1.33E-12	-1.04	5.30E-14
RV1371	membrane protein(Rv1371)	-0.35	3.06E-01	0.05	9.03E-01	-1.34	1.76E-04	0.76	3.12E-02	0.37	3.21E-01	-0.16	6.87E-01
RV1372	alpha-pyrone synthesis polyketide synthase-like protein(Rv1372)	-0.48	1.41E-04	0.07	6.49E-01	-1.08	4.52E-17	0.22	1.78E-01	-0.44	2.42E-03	-0.73	2.80E-07
RV1397C	ribonuclease VapC10(vapC10)	-0.09	2.27E-01	-0.52	3.54E-13	-0.78	3.14E-29	-0.35	4.11E-03	-1.12	5.32E-25	-1.17	1.08E-27
RV1405C	methyltransferase(Rv1405c)	-0.45	6.90E-02	-0.85	8.22E-04	-0.03	9.17E-01	0.22	3.68E-01	0.95	5.89E-07	1.31	9.04E-13
RV1412	riboflavin synthase(ribC)	-0.16	3.79E-02	0.12	1.66E-01	0.01	9.35E-01	0.06	6.90E-01	0.43	4.47E-05	0.39	1.19E-04
RV1413	hypothetical protein(Rv1413)	0.94	1.09E-14	-0.16	3.09E-01	1.16	2.91E-22	0.11	5.96E-01	0.26	1.17E-01	0.13	4.57E-01
RV1436	glyceraldehyde 3-phosphate dehydrogenase(gap)	-0.20	6.67E-04	-0.07	3.19E-01	0.29	3.04E-07	0.19	3.82E-04	0.38	1.02E-14	0.69	5.93E-48
RV1440	protein-export membrane protein SecG(secG)	-1.18	6.39E-29	-0.58	1.28E-07	-1.49	7.51E-46	0.28	2.17E-01	0.26	2.02E-01	-0.20	3.32E-01
RV1450C	PE-PGRS family protein PE_PGRS27(PE_PGRS27)	0.60	2.99E-03	0.36	1.11E-01	1.12	5.90E-09	-0.76	1.70E-03	0.49	3.19E-02	0.78	3.43E-04
RV1463	ABC transporter ATP-binding protein(Rv1463)	0.59	4.73E-12	0.08	4.54E-01	1.23	4.92E-51	-0.22	2.25E-01	0.62	2.91E-05	0.76	1.51E-07
RV1497	esterase LipL(lipL)	-0.76	3.35E-27	-0.30	5.43E-05	-1.10	2.19E-56	-0.29	6.62E-02	-0.31	2.99E-02	-0.45	9.05E-04
RV1499	hypothetical protein(Rv1499)	-0.14	6.31E-01	-0.61	4.55E-02	-1.14	1.52E-04	0.02	9.68E-01	-0.74	1.19E-02	-0.91	1.63E-03
RV1501	hypothetical protein(Rv1501)	-0.70	6.19E-16	-0.48	7.46E-08	-1.54	1.33E-69	0.11	2.98E-01	-0.70	4.86E-16	-0.87	4.42E-24
RV1502	hypothetical protein(Rv1502)	-0.95	3.22E-43	-0.43	1.85E-09	-1.36	7.64E-88	0.22	1.87E-02	-0.49	5.50E-09	-0.56	1.35E-11

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV1507A	hypothetical protein(Rv1507A)	0.33	3.15E-01	-0.14	7.36E-01	-0.57	1.00E-01	-0.25	4.46E-01	-0.92	1.44E-03	-1.27	2.03E-05
RV1508A	hypothetical protein(Rv1508A)	-0.66	7.78E-09	-0.53	5.72E-06	-1.09	1.19E-19	0.14	3.17E-01	-0.51	3.91E-05	-0.59	1.42E-06
RV1509	hypothetical protein(Rv1509)	-0.88	9.72E-23	-0.23	1.69E-02	-1.21	1.49E-40	0.00	9.93E-01	-0.72	2.30E-16	-0.70	7.07E-16
RV1528C	polyketide synthase associated protein PapA(papA4)	-1.23	1.37E-06	-0.19	5.27E-01	-1.26	9.52E-07	-0.24	6.10E-01	-0.23	5.72E-01	-0.43	2.43E-01
RV1552	fumarate reductase flavoprotein subunit(frdA)	-1.38	5.64E-12	-0.63	1.25E-03	-0.90	2.76E-06	-0.45	6.79E-02	-1.56	8.32E-11	-0.61	6.65E-03
RV1553	fumarate reductase iron-sulfur subunit(frdB)	-0.17	6.93E-01	0.38	3.81E-01	-0.63	1.22E-01	-0.86	1.69E-02	-1.15	1.25E-03	-0.90	9.05E-03
RV1563C	maltooligosyl trehalose synthase(treY)	0.20	2.95E-02	0.34	2.54E-04	0.11	2.64E-01	0.19	6.77E-02	0.00	1.00E+00	-0.09	3.70E-01
RV1578C	phage protein(Rv1578c)	0.62	2.57E-07	0.15	3.37E-01	1.32	2.24E-31	-0.08	6.21E-01	0.19	1.45E-01	0.93	3.71E-17
RV1581C	phage protein(Rv1581c)	0.32	4.78E-01	0.08	8.92E-01	0.21	6.42E-01	-1.00	5.89E-03	-1.03	4.27E-03	-0.99	5.02E-03
RV1582C	phage protein(Rv1582c)	-0.48	4.07E-04	0.37	7.26E-03	-0.59	1.39E-05	-0.72	1.98E-08	-1.04	8.63E-16	-0.97	5.87E-14
RV1583C	phage protein(Rv1583c)	1.03	2.39E-04	0.29	4.35E-01	0.92	1.26E-03	-0.49	7.20E-02	-0.79	3.43E-03	-0.07	7.92E-01
RV1584C	phage protein(Rv1584c)	0.23	5.49E-01	-0.71	8.04E-02	0.21	5.74E-01	-0.80	5.72E-03	-1.19	3.27E-05	-0.16	5.90E-01
RV1587C	hypothetical protein(Rv1587c)	1.20	4.32E-02	0.97	1.47E-01	-0.04	9.55E-01	-0.66	2.70E-01	-0.52	3.62E-01	-0.74	1.79E-01
RV1620C	cytochrome biosynthesis ABC transporter ATP-binding protein/permease CydC(cydC)	-0.06	5.93E-01	-0.89	1.93E-19	0.06	5.47E-01	-0.65	5.11E-10	-1.39	3.60E-43	-0.40	7.98E-05
RV1621C	cytochrome biosynthesis ABC transporter ATP-binding protein/permease CydD(cydD)	0.27	2.52E-03	-0.32	7.69E-04	0.30	8.85E-04	-0.66	1.57E-13	-1.12	3.25E-38	-0.55	2.99E-10
RV1622C	cytochrome D ubiquinol oxidase subunit II CydB(cydB)	-0.41	7.08E-11	-0.47	1.42E-13	-0.33	1.25E-07	-0.54	2.79E-06	-1.33	2.62E-35	-0.70	1.28E-10
RV1644	23S rRNA methyltransferase TsnR(tsnR)	0.68	1.66E-15	-0.08	4.80E-01	1.03	3.12E-34	-0.24	2.02E-02	0.24	1.20E-02	0.47	1.17E-07
RV1672C	integral membrane transport protein(Rv1672c)	0.92	1.64E-12	0.22	1.77E-01	1.23	5.84E-22	-0.39	3.56E-02	0.27	1.14E-01	0.51	1.32E-03
RV1682	hypothetical protein(Rv1682)	-0.65	4.53E-10	-0.18	1.33E-01	-1.35	3.15E-34	0.21	1.99E-01	-0.60	2.24E-05	-0.89	1.48E-10
RV1724C	hypothetical protein(Rv1724c)	0.22	1.43E-01	-0.13	4.76E-01	-0.40	6.26E-03	0.10	5.75E-01	-0.94	1.62E-11	-1.01	2.87E-13
RV1733C	transmembrane protein(Rv1733c)	-1.92	2.32E-39	-0.66	2.33E-06	0.07	6.28E-01	-0.94	2.61E-06	-0.26	2.14E-01	-0.80	2.24E-05
RV1734C	hypothetical protein(Rv1734c)	-1.62	2.14E-02	-0.55	4.72E-01	-1.47	3.54E-02	-0.23	5.93E-01	-0.88	1.18E-02	-1.74	4.39E-06
RV1735A	hypothetical protein(Rv1735a)	-1.71	4.91E-07	-0.36	3.18E-01	-2.19	4.44E-09	-0.65	1.86E-02	-0.96	1.55E-04	-1.35	6.53E-08
RV1735C	membrane protein(Rv1735c)	-1.68	9.57E-05	-0.38	3.95E-01	-1.23	2.97E-03	-0.38	2.43E-01	-0.86	2.21E-03	-1.44	2.68E-07

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV1736C	nitrate reductase-like protein NarX(narX)	-1.67	6.55E-22	-0.27	1.97E-01	-0.88	4.09E-07	-0.63	2.40E-03	-0.55	5.33E-03	-1.18	1.99E-10
RV1737C	nitrate/nitrite transporter(narK2)	-1.61	4.76E-34	0.05	7.83E-01	-0.59	8.34E-06	-0.36	1.15E-01	-0.17	4.67E-01	-0.72	1.65E-04
RV1738	hypothetical protein(Rv1738)	-3.10	2.90E-47	-0.98	1.65E-05	-0.03	8.95E-01	-0.49	8.17E-02	0.57	2.20E-02	0.15	5.79E-01
RV1740	antitoxin VapB34(vapB34)	-0.70	6.40E-08	-0.25	7.90E-02	-1.04	4.22E-15	0.08	5.99E-01	-0.28	2.56E-02	-0.40	8.00E-04
RV1754C	hypothetical protein(Rv1754c)	-0.60	2.17E-05	-0.17	3.11E-01	-1.38	8.32E-22	-0.54	2.53E-05	-0.95	3.60E-15	-1.45	1.16E-33
RV1759C	PE-PGRS family protein Wag22(wag22)	0.49	2.89E-03	0.13	5.16E-01	1.04	3.35E-11	-0.64	6.72E-03	0.59	6.85E-03	0.97	2.46E-06
RV1798	ESX-5 type VII secretion system protein EccA(eccA5)	-0.45	9.57E-16	0.25	1.77E-05	-0.56	3.74E-24	-0.08	3.55E-01	-0.39	3.28E-09	-0.34	1.54E-07
RV1813C	hypothetical protein(Rv1813c)	-2.07	8.71E-92	-0.52	1.18E-06	-1.30	2.31E-37	-0.25	2.64E-01	-0.36	6.18E-02	-1.21	3.22E-12
RV1815	hypothetical protein(Rv1815)	-0.74	1.37E-23	-0.11	2.26E-01	-1.12	2.08E-51	-0.22	9.07E-02	0.28	1.75E-02	-0.44	9.79E-05
RV1818C	PE-PGRS family protein PE_PGRS33(PE_PGRS33)	0.75	1.11E-04	0.08	7.56E-01	1.07	1.38E-08	-0.99	7.28E-04	0.43	1.50E-01	0.64	2.11E-02
RV1882C	short-chain type dehydrogenase/reductase(Rv1882c)	-0.81	2.71E-27	-0.06	5.53E-01	-1.17	2.98E-53	-0.13	2.98E-01	-0.18	1.01E-01	-0.57	6.39E-09
RV1930C	hypothetical protein(Rv1930c)	1.34	1.05E-03	0.46	3.88E-01	1.40	5.67E-04	0.32	4.81E-01	0.50	1.82E-01	0.53	1.47E-01
RV1933C	acyl-CoA dehydrogenase FadE18(fadE18)	0.21	7.42E-01	-0.13	8.67E-01	0.74	1.83E-01	-1.05	4.69E-02	-0.12	8.36E-01	-0.54	2.76E-01
RV1974	membrane protein(Rv1974)	-0.36	4.40E-01	-1.04	4.25E-02	-1.18	1.73E-02	-0.64	1.07E-01	0.08	8.51E-01	-0.40	2.87E-01
RV1982A	antitoxin VapB36(vapB36)	-1.16	4.88E-22	-0.30	2.46E-02	-0.40	1.04E-03	-0.40	6.54E-02	0.19	3.69E-01	1.25	5.00E-12
RV1988	23S rRNA (adenine(2058)-N(6))-methyltransferase Erm(37)(erm(37))	-0.71	4.14E-05	0.24	2.42E-01	-1.13	2.70E-11	-0.05	8.94E-01	-0.20	4.63E-01	-0.03	9.19E-01
RV1991A	antitoxin MazE6(mazE6)	-1.03	5.01E-19	-0.23	5.99E-02	-0.26	2.03E-02	-0.15	3.68E-01	0.23	1.01E-01	0.60	2.60E-06
RV1996	universal stress protein(Rv1996)	-1.11	8.53E-25	0.41	2.70E-04	1.16	7.29E-29	-0.33	1.08E-01	0.66	1.70E-04	0.74	1.49E-05
RV1997	cation transporter ATPase F(ctpF)	-1.33	2.62E-30	0.24	6.70E-02	-0.53	3.88E-06	-0.29	2.97E-01	-0.11	6.83E-01	-0.95	1.58E-05
RV1999C	transporter(Rv1999c)	0.67	1.46E-05	2.30	1.93E-60	0.79	2.49E-07	0.08	7.86E-01	1.67	1.31E-21	0.48	1.25E-02
RV2005C	universal stress protein(Rv2005c)	-1.30	2.69E-29	-0.31	1.47E-02	-0.75	8.76E-11	0.07	8.32E-01	-0.03	9.12E-01	-0.26	2.65E-01
RV2007C	ferredoxin(fdxA)	-3.17	2.04E-83	-2.05	2.58E-35	-1.54	3.71E-21	-0.24	2.63E-01	-0.33	7.48E-02	-0.90	1.48E-07
RV2022C	hypothetical protein(Rv2022c)	-0.83	5.66E-18	-0.35	5.21E-04	-1.03	5.32E-27	0.01	9.65E-01	-0.43	1.93E-02	-0.47	8.84E-03
RV2028C	universal stress protein(Rv2028c)	-2.38	5.86E-39	-0.73	1.25E-04	-1.98	3.89E-28	-0.38	8.96E-02	-0.57	4.27E-03	-1.20	1.73E-10
RV2029C	6-phosphofructokinase PfkB(pfkB)	-2.12	1.24E-42	-0.51	2.54E-03	-1.63	4.27E-26	-0.29	2.84E-01	-0.59	9.06E-03	-1.01	2.49E-06
RV2030C	hypothetical protein(Rv2030c)	-1.95	4.16E-45	-0.85	3.10E-09	-1.12	4.07E-16	-0.47	3.95E-02	-0.49	1.79E-02	-0.95	8.16E-07
RV2031C	alpha-crystallin(hspX)	-2.76	5.06E-63	-1.53	5.73E-20	-0.60	3.05E-04	-0.69	6.10E-03	-0.05	8.72E-01	-0.89	8.06E-05
RV2032	NAD(P)H nitroreductase(acg)	-1.69	9.31E-32	-0.19	2.51E-01	-0.05	7.51E-01	-0.92	1.21E-05	-0.32	1.44E-01	-1.18	1.19E-09

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV2063	antitoxin MazE7(mazE7)	0.59	1.51E-01	0.07	9.13E-01	1.09	3.99E-03	-0.88	7.79E-02	0.22	6.59E-01	0.35	4.28E-01
RV2085	hypothetical protein(Rv2085)	-0.98	4.17E-06	-0.03	9.29E-01	-1.02	1.89E-06	-0.44	1.74E-01	-0.15	6.59E-01	-0.31	2.93E-01
	Sec-independent protein translocase membrane-bound protein												
RV2094C	TatA(tatA)	-1.22	1.11E-30	-0.15	2.42E-01	-0.65	1.26E-09	0.24	2.33E-01	0.71	8.41E-06	0.84	7.18E-08
RV2101	helicase HelZ(helZ)	0.40	1.35E-09	0.04	6.32E-01	0.23	7.23E-04	-0.07	4.56E-01	0.22	2.32E-03	0.10	1.95E-01
RV2104A	hypothetical protein(Rv2104a)	0.07	6.70E-01	-0.04	8.37E-01	-0.64	3.54E-05	-0.02	9.14E-01	-0.90	7.31E-12	-1.13	5.25E-17
RV2111C	ubiquitin-like protein Pup(pup)	-0.94	3.98E-13	-0.46	9.04E-04	-1.07	6.39E-17	-0.11	6.86E-01	-0.02	9.47E-01	-0.02	9.46E-01
RV2132	hypothetical protein(Rv2132)	-0.74	3.50E-11	-0.01	9.52E-01	-1.42	4.23E-33	-0.10	6.00E-01	-0.43	2.81E-03	-0.80	7.78E-09
RV2142A	antitoxin ParD2(parD2)	-0.96	8.52E-13	-0.25	1.03E-01	-1.06	4.13E-15	-0.19	3.80E-01	-0.25	1.76E-01	-0.16	3.78E-01
RV2142C	toxin ParE2(parE2)	-0.39	2.12E-02	-0.27	1.58E-01	-1.20	1.37E-11	-0.17	4.04E-01	-1.17	2.24E-11	-1.49	8.52E-17
RV2153C	(murG)	1.27	6.73E-30	0.07	6.40E-01	1.16	6.14E-25	-0.38	2.74E-03	-0.07	6.37E-01	0.03	7.99E-01
RV2158C	(murE)	0.91	1.62E-14	-0.07	6.55E-01	1.16	4.40E-23	-0.79	9.54E-13	-0.09	5.17E-01	0.18	1.22E-01
RV2159C	hypothetical protein(Rv2159c)	0.61	8.09E-07	-0.07	6.82E-01	1.12	1.66E-20	-1.12	3.17E-16	-0.53	1.61E-04	0.12	4.51E-01
	transcriptional regulator												
RV2166C	MraZ(Rv2166c)	-0.73	9.72E-16	0.08	4.80E-01	-1.73	9.61E-79	-0.13	3.90E-01	-0.53	1.20E-05	-1.02	1.80E-18
RV2169C	transmembrane protein(Rv2169c)	-0.93	8.27E-29	-0.27	2.67E-03	-1.39	4.00E-62	0.03	7.82E-01	-0.31	1.83E-04	-0.69	1.61E-18
RV2204C	hypothetical protein(Rv2204c)	-1.12	4.31E-43	-0.38	8.50E-06	-1.00	3.53E-35	-0.06	6.45E-01	-0.06	5.55E-01	0.21	2.37E-02
RV2237A	hypothetical protein(Rv2237A)	-1.52	6.65E-20	-0.58	1.30E-03	-1.55	7.97E-21	0.09	8.26E-01	0.03	9.42E-01	0.03	9.30E-01
RV2253	hypothetical protein(Rv2253)	-0.05	9.09E-01	0.62	1.01E-01	-1.06	8.73E-03	-0.16	7.66E-01	0.04	9.31E-01	-0.89	2.90E-02
RV2269C	hypothetical protein(Rv2269c)	2.95	6.28E-02	2.19	2.41E-01	3.40	2.62E-02	-0.01	9.88E-01	-0.09	9.22E-01	-0.93	2.70E-01
RV2272	transmembrane protein(Rv2272)	1.34	4.63E-14	0.00	9.89E-01	1.52	7.04E-18	-0.03	9.22E-01	0.23	2.25E-01	0.45	9.04E-03
RV2274A	antitoxin MazE8(mazE8)	-0.64	2.19E-03	-0.30	2.05E-01	-1.41	4.47E-10	0.24	3.30E-01	-0.42	6.46E-02	-0.62	5.41E-03
RV2292C	hypothetical protein(Rv2292c)	0.62	9.25E-02	0.70	7.64E-02	0.24	5.50E-01	-0.72	3.00E-02	-0.87	6.35E-03	-1.40	2.03E-05
RV2293C	hypothetical protein(Rv2293c)	-0.66	4.26E-14	-0.20	3.30E-02	-1.19	2.75E-37	-0.47	8.40E-08	-1.23	4.69E-45	-1.31	5.30E-49
RV2294	cystathionine beta-lyase(Rv2294)	0.43	5.50E-07	0.23	1.51E-02	0.71	1.41E-17	0.23	2.17E-02	0.61	1.79E-12	0.54	2.63E-10
RV2301	cutinase(cut2)	0.82	3.62E-12	-0.11	4.82E-01	1.20	3.36E-25	-0.40	2.89E-02	0.23	1.86E-01	0.38	1.88E-02
RV2304C	hypothetical protein(Rv2304c)	-1.06	1.16E-09	-0.08	7.27E-01	-1.32	1.35E-13	-0.01	9.70E-01	-0.09	6.71E-01	-0.06	7.78E-01
RV2307B	hypothetical protein(Rv2307B)	-1.04	2.14E-11	-0.19	2.59E-01	-1.38	6.61E-17	0.35	1.01E-01	-0.31	1.48E-01	-0.12	5.88E-01
RV2313C	hypothetical protein(Rv2313c)	-0.81	5.78E-28	-0.13	1.36E-01	-1.12	9.36E-52	-0.21	3.63E-02	-0.55	3.14E-10	-0.72	2.03E-17
RV2331	hypothetical protein(Rv2331)	-0.52	2.89E-03	-0.07	7.77E-01	-1.17	1.27E-11	0.09	6.90E-01	-0.69	1.24E-05	-0.52	8.26E-04
RV2336	hypothetical protein(Rv2336)	-0.48	7.67E-07	-0.04	7.43E-01	-1.20	4.74E-34	-0.24	5.06E-02	-1.00	4.05E-21	-1.22	1.65E-30
	molybdopterin biosynthesis protein												
RV2338C	MoeW(moeW)	-1.22	8.88E-59	-0.55	8.05E-13	-1.77	1.07E-112	-0.31	1.44E-03	-0.76	1.71E-17	-1.20	3.57E-42

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV2353C	PPE family protein PPE39(PPE39)	-0.69	1.31E-01	0.17	7.59E-01	-1.14	1.77E-02	0.15	8.10E-01	-0.24	6.59E-01	0.05	9.21E-01
RV2365C	hypothetical protein(Rv2365c)	1.45	1.95E-19	0.38	4.53E-02	1.17	8.48E-13	-0.45	5.69E-04	-0.33	1.05E-02	-0.41	1.22E-03
RV2368C	phosphate starvation-inducible protein PhoH(phoH1)	-0.35	2.01E-07	-0.25	7.19E-04	-1.02	2.94E-51	-0.01	9.33E-01	-0.70	1.09E-22	-0.91	1.17E-37
RV2382C	polyketide synthetase(mbtC)	0.46	1.27E-06	0.20	6.24E-02	1.04	5.70E-30	-0.09	7.08E-01	-0.19	3.24E-01	0.43	1.04E-02
RV2383C	phenyloxazoline synthase(mbtB)	0.74	2.63E-18	0.89	9.29E-26	1.03	1.91E-35	0.10	7.10E-01	0.15	5.03E-01	0.14	4.98E-01
RV2386A	hypothetical protein(Rv2386a)	-0.38	5.95E-05	-0.66	1.02E-11	-0.80	7.36E-17	-0.44	4.89E-03	-1.18	7.67E-17	-0.98	3.43E-12
RV2390C	hypothetical protein(Rv2390c)	-0.49	1.09E-04	-0.46	5.99E-04	-0.06	6.64E-01	0.52	2.30E-04	0.90	6.42E-12	1.78	7.18E-47
RV2397C	sulfate ABC transporter ATP-binding protein CysA(cysA1)	0.85	6.00E-15	0.15	2.55E-01	1.07	3.23E-23	0.07	6.89E-01	0.45	2.58E-04	0.46	1.26E-04
RV2401	hypothetical protein(Rv2401)	-0.65	5.46E-02	-0.27	4.98E-01	-0.65	5.59E-02	0.03	9.50E-01	-0.27	4.93E-01	-1.20	1.49E-03
RV2406C	hypothetical protein(Rv2406c)	-1.22	2.00E-33	-0.36	7.33E-04	-1.26	1.90E-35	-0.37	1.08E-02	-0.36	8.78E-03	-0.60	2.54E-06
RV2428	alkyl hydroperoxide reductase sub-unit AhpC(ahpC)	0.06	6.73E-01	0.27	6.46E-02	0.22	1.06E-01	0.38	7.43E-03	1.04	3.26E-17	0.52	3.52E-05
RV2431C	PE family protein PE25(PE25)	-0.78	2.58E-14	-0.17	1.68E-01	-1.04	5.49E-25	0.18	2.62E-01	-0.06	7.12E-01	-0.04	8.14E-01
RV2432C	hypothetical protein(Rv2432c)	-1.12	4.01E-07	-0.61	1.09E-02	-2.40	4.90E-28	-0.19	6.11E-01	-0.99	3.16E-04	-1.53	6.85E-09
RV2439C	glutamate 5-kinase protein(proB)	0.67	2.01E-14	-0.07	5.26E-01	1.14	1.30E-40	0.17	3.96E-02	0.29	9.67E-05	0.95	4.33E-42
RV2450C	resuscitation-promoting factor RpfE(rpfE)	1.48	2.11E-41	0.40	1.34E-03	1.61	1.95E-49	0.45	4.37E-03	1.40	3.44E-24	1.35	6.28E-23
RV2451	hypothetical protein(Rv2451)	-0.04	9.29E-01	-0.30	5.80E-01	-1.34	8.72E-03	0.23	7.78E-01	-0.15	8.48E-01	-0.27	6.80E-01
RV2452C	hypothetical protein(Rv2452c)	-0.42	3.91E-03	-0.26	1.03E-01	-1.29	1.11E-18	-0.21	2.18E-01	-1.01	3.81E-12	-1.13	7.93E-15
RV2470	hemoglobin GlbO(glbO)	-1.09	5.66E-26	-0.36	1.27E-03	-1.46	7.77E-45	0.32	7.30E-02	0.00	9.89E-01	-0.25	1.20E-01
RV2485C	carboxylesterase LipQ(lipQ)	-0.50	4.25E-04	-0.61	3.84E-05	-0.09	5.54E-01	-1.33	1.47E-16	-1.19	1.55E-13	-1.13	1.99E-12
RV2490A	hypothetical protein(Rv2490a)	-1.12	5.30E-04	0.12	7.63E-01	-0.43	1.73E-01	-0.01	9.78E-01	0.07	8.44E-01	-0.09	7.57E-01
RV2490C	PE-PGRS family protein PE_PGRS43(PE_PGRS43)	0.46	9.45E-02	0.50	9.75E-02	1.08	3.28E-05	-1.05	2.75E-03	0.75	2.48E-02	0.96	2.57E-03
RV2491	hypothetical protein(Rv2491)	0.05	9.13E-01	-0.05	9.29E-01	-0.82	5.01E-02	0.44	1.72E-01	-0.24	4.88E-01	-1.30	1.44E-04
RV2494	ribonuclease VapC38(vapC38)	0.32	1.77E-02	-0.34	3.02E-02	0.00	9.91E-01	-0.56	2.83E-03	-0.75	1.99E-05	-1.04	2.62E-09
RV2512A	hypothetical protein(Rv2512a)	-0.26	9.50E-02	0.13	4.89E-01	-1.65	4.26E-24	0.53	7.14E-04	-0.57	2.76E-04	-0.99	2.46E-10
RV2516C	hypothetical protein(Rv2516c)	-0.87	3.71E-18	0.29	5.80E-03	-1.12	7.65E-29	-0.40	1.36E-05	-0.50	1.16E-08	-0.83	4.91E-22
RV2517C	hypothetical protein(Rv2517c)	-0.59	3.61E-04	0.29	1.10E-01	-1.22	7.72E-13	-0.17	3.13E-01	-0.57	5.93E-05	-0.78	2.56E-08
RV2522C	hypothetical protein(Rv2522c)	0.50	1.86E-08	0.38	4.37E-05	1.06	4.10E-35	0.23	6.80E-02	0.51	4.18E-06	0.55	2.71E-07
RV2525C	hypothetical protein(Rv2525c)	-0.32	5.47E-03	-0.03	8.74E-01	-1.38	2.43E-34	-0.25	5.88E-02	-0.39	1.12E-03	-0.85	1.12E-13
RV2541	hypothetical protein(Rv2541)	1.60	1.45E-09	-0.47	1.81E-01	1.38	2.35E-07	-1.27	2.26E-04	0.78	1.48E-02	0.65	3.99E-02
RV2550C	antitoxin VapB20(vapB20)	-0.30	1.60E-02	-0.16	2.69E-01	0.17	1.72E-01	0.57	8.40E-03	1.33	2.01E-12	1.67	1.35E-19

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV2566	hypothetical protein(Rv2566)	1.28	2.23E-37	0.30	9.44E-03	1.21	2.00E-33	-0.26	1.85E-01	0.08	6.73E-01	0.21	2.11E-01
RV2594C	crossover junction endodeoxyribo-nuclease RuvC(ruvC)	-0.80	1.62E-13	-0.09	5.10E-01	-1.12	2.67E-25	0.27	1.45E-01	0.33	3.81E-02	0.04	8.42E-01
RV2623	universal stress protein(TB31.7)	-1.24	4.41E-19	0.57	1.33E-04	-0.33	2.32E-02	-0.14	6.73E-01	-0.20	4.55E-01	-0.71	1.78E-03
RV2624C	universal stress protein(Rv2624c)	-2.17	3.41E-31	0.22	3.08E-01	-1.88	2.97E-24	-0.49	9.64E-02	-0.33	2.37E-01	-1.31	9.74E-08
RV2625C	zinc metalloprotease Rip3(Rv2625c)	-1.99	3.09E-26	0.35	1.12E-01	-1.41	4.08E-14	-0.46	1.76E-01	-0.22	4.98E-01	-1.10	5.87E-05
RV2626C	hypoxic response protein(hrp1)	-2.02	1.70E-42	-0.21	2.01E-01	0.41	3.62E-03	-0.43	1.38E-01	0.21	4.61E-01	-0.94	8.25E-05
RV2627C	hypothetical protein(Rv2627c)	-1.05	1.41E-16	1.07	2.04E-17	0.34	8.66E-03	-0.21	5.54E-01	0.14	6.65E-01	-0.80	1.97E-03
RV2628	hypothetical protein(Rv2628)	-1.44	1.41E-22	0.81	1.90E-08	0.62	9.81E-06	-0.49	1.98E-01	0.31	3.80E-01	-0.13	7.28E-01
RV2632C	hypothetical protein(Rv2632c)	-1.16	1.57E-35	-0.68	4.46E-13	-0.88	3.14E-21	-0.35	7.38E-02	-0.57	9.33E-04	-0.28	1.17E-01
RV2639C	integral membrane pro- tein(Rv2639c)	-1.00	6.69E-12	-0.37	2.14E-02	-1.11	1.29E-14	0.17	5.60E-01	0.38	9.64E-02	0.05	8.35E-01
RV2641	cadmium inducible protein CadI(cadI)	-1.44	3.75E-15	-1.09	1.06E-08	-1.93	4.12E-26	0.10	4.43E-01	-0.04	7.42E-01	-0.37	2.41E-04
RV2645	hypothetical protein(Rv2645)	1.52	1.55E-09	0.18	6.23E-01	1.49	3.35E-09	-0.44	1.25E-01	0.19	4.95E-01	0.17	5.31E-01
RV2646	integrase(Rv2646)	1.21	1.71E-12	0.26	2.29E-01	1.05	1.77E-09	-0.39	4.58E-02	0.15	4.52E-01	-0.08	6.80E-01
RV2647	hypothetical protein(Rv2647)	1.26	1.68E-06	0.29	4.15E-01	1.05	8.80E-05	-0.15	6.78E-01	-0.19	5.33E-01	-0.07	8.19E-01
RV2654C	antitoxin(Rv2654c)	3.13	6.84E-02	0.62	8.11E-01	3.69	2.45E-02	-2.03	1.49E-01	-0.17	8.97E-01	0.14	9.00E-01
RV2658C	prophage protein(Rv2658c)	-1.07	1.27E-08	-0.69	2.76E-04	-1.33	1.50E-11	-1.14	4.31E-10	-0.85	3.15E-06	-1.41	2.85E-14
RV2659C	prophage integrase(Rv2659c)	-0.81	3.21E-06	-0.32	8.68E-02	-0.39	2.52E-02	-1.53	3.58E-26	-0.66	4.66E-06	-0.98	1.04E-11
RV2663	hypothetical protein(Rv2663)	-1.47	2.82E-27	-0.28	5.39E-02	-1.78	4.44E-36	-0.54	1.93E-02	-0.58	7.95E-03	-0.72	5.22E-04
RV2669	GCN5-like N-acetyltransfer- ase(Rv2669)	-0.11	5.16E-01	-0.35	4.08E-02	-0.11	5.05E-01	-0.48	3.34E-04	-1.09	3.21E-16	-0.58	7.63E-06
RV2677C	protoporphyrinogen oxidase(hemY)	1.15	4.17E-26	0.20	1.19E-01	1.32	2.77E-34	-0.37	2.44E-03	0.21	6.82E-02	0.40	2.44E-04
RV2694C	hypothetical protein(Rv2694c)	-0.87	4.48E-15	-0.32	8.67E-03	-1.09	3.73E-23	-0.20	1.52E-01	-0.52	7.01E-06	-0.60	1.03E-07
RV2698	transmembrane protein(Rv2698)	-0.93	3.11E-24	-0.32	9.66E-04	-1.13	7.78E-34	-0.02	9.24E-01	-0.07	6.38E-01	-0.34	7.42E-03
RV2723	integral membrane pro- tein(Rv2723)	0.14	1.13E-01	-0.28	2.79E-03	-0.80	6.97E-18	-0.14	3.34E-01	-0.95	3.34E-17	-1.25	4.65E-28
RV2730	hypothetical protein(Rv2730)	-0.88	3.10E-03	-0.15	7.08E-01	-1.13	1.33E-04	-0.09	8.83E-01	0.29	5.32E-01	-0.32	4.57E-01
RV2767C	membrane protein(Rv2767c)	0.41	4.42E-02	0.51	1.71E-02	1.12	1.37E-09	-0.16	5.18E-01	0.23	2.61E-01	0.27	1.58E-01
RV2768C	PPE family protein PPE43(PPE43)	0.66	1.74E-08	0.17	2.36E-01	1.36	5.78E-34	-0.41	9.50E-04	0.62	1.29E-08	0.67	4.95E-10
RV2782C	zinc protease(pepR)	0.77	6.20E-20	0.01	9.55E-01	1.31	9.80E-57	-0.10	4.17E-01	0.30	2.40E-03	0.68	7.00E-14
RV2800	hydrolase(Rv2800)	0.91	2.23E-17	0.29	2.01E-02	1.10	1.90E-25	-0.42	5.13E-04	-0.14	2.63E-01	0.02	8.63E-01
RV2801C	mRNA interferase MazF9(mazF9)	1.00	1.27E-07	0.20	4.17E-01	1.18	3.19E-10	-0.12	6.60E-01	0.12	6.35E-01	-0.02	9.46E-01
RV2808	hypothetical protein(Rv2808)	-1.09	3.89E-25	-0.37	7.93E-04	-1.18	1.02E-28	0.39	7.07E-03	-0.25	8.27E-02	-0.01	9.33E-01

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV2809	hypothetical protein(Rv2809)	-1.07	5.11E-16	-0.04	8.49E-01	-1.03	5.65E-15	0.04	8.70E-01	-0.13	3.98E-01	0.11	4.76E-01
RV2812	transposase(Rv2812)	-0.23	4.61E-01	0.64	2.83E-02	0.48	8.73E-02	0.41	3.68E-01	1.23	5.05E-04	1.17	7.93E-04
RV2816C	CRISPR-associated endoribonuclease Cas2(Rv2816c)	-1.71	9.29E-14	-0.99	3.80E-05	-2.38	3.06E-24	0.44	2.70E-01	-0.30	4.31E-01	-0.54	1.10E-01
	enoyl-CoA hydratase												
RV2831	EchA16(echA16)	0.61	1.41E-05	-0.01	9.44E-01	1.18	2.15E-18	-0.37	1.17E-02	0.43	1.37E-03	0.49	1.31E-04
RV2866	toxin RelG(relG)	0.67	1.95E-01	-0.20	7.80E-01	0.00	9.96E-01	-0.55	1.90E-01	-1.09	7.14E-03	-0.50	1.99E-01
RV2873	cell surface lipoprotein(mpt83)	1.24	1.19E-21	2.37	2.33E-82	1.56	1.42E-34	0.39	4.41E-02	1.89	9.54E-32	0.70	3.66E-05
RV2874	DipZ(dipZ)	1.03	9.15E-12	2.08	3.95E-47	0.96	2.81E-10	0.52	1.52E-02	1.78	2.22E-22	0.12	5.83E-01
	major secreted immunogenic protein Mpt70(mpt70)												
RV2875		2.19	2.79E-44	4.11	1.80E-158	2.39	8.58E-53	1.67	2.71E-19	4.02	2.29E-116	1.97	8.68E-28
RV2876	transmembrane protein(Rv2876)	-0.25	9.05E-03	0.84	3.89E-20	-0.33	3.67E-04	0.22	3.46E-01	1.11	9.13E-10	-0.09	6.68E-01
RV2886C	resolvase(Rv2886c)	-0.67	1.15E-06	0.43	3.16E-03	-0.28	4.87E-02	0.17	4.21E-01	0.80	3.77E-07	0.88	1.15E-08
RV2891	hypothetical protein(Rv2891)	1.02	7.95E-02	1.07	8.87E-02	1.27	2.36E-02	-0.54	4.90E-01	-0.38	6.02E-01	-0.49	4.81E-01
RV2897C	hypothetical protein(Rv2897c)	0.15	1.62E-01	0.57	3.38E-08	1.04	6.67E-27	0.39	3.56E-02	0.88	5.28E-08	1.26	7.07E-16
RV2898C	hypothetical protein(Rv2898c)	-0.68	7.27E-11	0.06	6.94E-01	0.04	7.23E-01	0.40	1.11E-02	0.59	4.05E-05	1.12	2.36E-16
RV2909C	30S ribosomal protein S16(rpsP)	0.65	4.08E-17	0.11	2.51E-01	1.02	2.70E-41	-0.26	7.42E-02	0.18	1.97E-01	0.55	1.06E-05
	D-amino acid aminohydro-												
RV2913C	lase(Rv2913c)	-1.04	5.65E-18	-0.13	3.70E-01	-0.95	2.40E-15	-0.82	5.04E-19	-0.75	5.55E-16	-0.93	3.65E-24
	phthiocerol synthesis polyketide synthase type I PpsC(ppsC)												
RV2933		0.40	1.06E-12	0.39	1.42E-11	0.80	7.77E-47	0.64	5.90E-11	0.67	1.79E-12	1.10	7.05E-33
	phthiocerol synthesis polyketide synthase type I PpsE(ppsE)												
RV2935		0.79	7.91E-20	0.15	1.39E-01	1.15	1.42E-40	0.44	1.30E-04	0.69	1.22E-10	0.84	9.22E-16
RV2943A	transposase(Rv2943A)	-1.35	1.06E-04	-0.08	8.62E-01	-0.91	7.52E-03	-0.11	7.99E-01	-0.66	4.38E-02	-0.33	3.10E-01
RV2946C	polyketide synthase(pks1)	0.12	1.90E-01	0.56	5.93E-11	0.81	3.24E-23	0.14	5.15E-01	0.62	1.06E-04	1.18	4.53E-15
RV2947C	polyketide synthase(pks15)	-0.24	6.18E-03	0.53	1.20E-09	0.47	2.01E-08	0.08	7.22E-01	0.78	3.91E-07	1.21	3.59E-16
RV2961	transposase(Rv2961)	-0.72	4.26E-02	-0.68	7.63E-02	-1.19	1.26E-03	0.04	9.54E-01	-0.15	7.74E-01	-0.44	3.32E-01
	PGL/p-HBAD biosynthesis rhamnosyltransferase(Rv2962c)												
RV2962C		-0.63	2.37E-08	-0.38	1.65E-03	-1.08	7.52E-22	-0.40	4.08E-05	-0.26	6.92E-03	-0.65	1.17E-12
	integral membrane protein(Rv2963)												
RV2963		-1.08	3.75E-32	-0.91	1.04E-22	-1.81	9.59E-87	-0.68	1.54E-10	-0.99	1.77E-22	-1.38	1.53E-42
	formyltetrahydrofolate deformylase(purU)												
RV2964		-0.07	5.64E-01	-0.04	7.87E-01	-0.66	1.57E-08	-0.25	1.15E-01	-0.78	7.27E-09	-1.08	1.41E-15
RV2978C	transposase(Rv2978c)	0.68	8.46E-19	0.19	3.21E-02	1.08	1.73E-47	0.21	5.78E-02	0.36	2.57E-04	0.68	2.87E-13
RV2986C	DNA-binding protein HU(hupB)	-0.27	3.36E-03	0.52	1.06E-08	0.10	2.94E-01	0.44	1.25E-03	0.76	7.01E-10	1.20	2.20E-23
RV2994	MFS-type transporter(Rv2994)	0.84	5.77E-21	0.06	6.25E-01	1.02	1.19E-30	-0.24	5.91E-02	0.28	1.80E-02	0.20	8.08E-02

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV3008	hypothetical protein(Rv3008)	-0.91	1.85E-12	-0.18	2.29E-01	-1.37	1.02E-25	-0.07	6.55E-01	-0.44	2.66E-04	-0.44	2.27E-04
RV3011C	glutamyl-tRNA(GLN) amidotransferase subunit A(gatA)	0.74	2.05E-20	-0.01	9.28E-01	1.14	4.34E-47	0.00	9.83E-01	0.54	5.69E-12	0.72	3.78E-21
RV3012C	glutamyl-tRNA(GLN) amidotransferase subunit C(gatC)	0.91	2.82E-06	0.33	1.59E-01	1.19	3.19E-10	0.01	9.84E-01	0.05	8.87E-01	0.07	8.14E-01
RV3019C	ESAT-6 like protein EsxR(esxR)	0.54	2.81E-04	0.46	3.59E-03	1.19	1.31E-17	-0.09	6.72E-01	1.03	1.81E-12	1.38	1.61E-22
RV3020C	ESAT-6 like protein EsxS(esxS)	-0.13	3.79E-01	0.48	3.76E-04	0.75	3.16E-09	0.14	4.30E-01	1.12	4.41E-18	1.51	1.78E-33
RV3022C	pseudo(PPE48)	-0.41	2.62E-01	0.09	8.49E-01	0.29	3.90E-01	0.25	6.72E-01	1.13	5.92E-03	1.31	8.17E-04
RV3036C	hypothetical protein(TB22.2)	1.05	2.45E-17	0.19	2.21E-01	0.60	2.33E-06	-0.27	2.70E-01	-0.67	8.22E-04	-0.67	6.41E-04
RV3084	acetyl-hydrolase LipR(lipR)	0.64	7.05E-06	0.54	3.28E-04	1.03	4.65E-14	-0.58	4.07E-04	0.22	1.87E-01	0.11	5.32E-01
RV3093C	oxidoreductase(Rv3093c)	0.30	2.72E-05	-1.27	3.70E-70	0.61	2.55E-18	-0.22	3.43E-02	-1.09	1.75E-33	0.02	8.85E-01
RV3094C	hypothetical protein(Rv3094c)	0.27	4.53E-05	-1.18	5.01E-76	0.38	2.64E-09	-0.22	1.52E-02	-1.18	7.03E-51	-0.16	6.07E-02
RV3095	HTH-type transcriptional regulator(Rv3095)	-0.35	1.10E-04	-0.82	2.88E-20	-0.95	2.72E-27	-0.32	2.04E-03	-1.21	8.32E-38	-0.96	1.07E-24
RV3108	hypothetical protein(Rv3108)	-1.54	5.47E-02	-0.73	3.91E-01	-2.91	5.86E-03	-0.03	9.77E-01	-0.50	5.72E-01	-1.99	2.96E-02
RV3109	cyclic pyranopterin monophosphate synthase(moaA1)	-0.79	9.81E-05	-0.67	1.71E-03	-1.79	1.14E-15	0.10	6.41E-01	-0.76	2.06E-05	-0.68	1.09E-04
RV3110	pterin-4-alpha-carbinolamine dehydratase(moaB1)	-1.04	5.17E-03	-0.85	3.16E-02	-1.63	4.23E-05	0.26	5.24E-01	-0.20	6.18E-01	-0.46	2.14E-01
RV3114	hypothetical protein(Rv3114)	-1.01	3.78E-02	-0.52	3.41E-01	-1.44	4.36E-03	0.11	8.28E-01	-0.88	3.19E-02	-0.90	2.69E-02
RV3118	hypothetical protein(Rv3118)	0.86	1.41E-14	-0.40	1.78E-03	1.18	7.08E-27	-0.19	6.77E-02	0.29	1.71E-03	0.49	1.65E-08
RV3127	hypothetical protein(Rv3127)	-1.31	6.35E-32	-0.36	2.83E-03	0.48	1.50E-05	-0.46	6.86E-02	0.04	9.04E-01	-0.44	5.23E-02
RV3128C	pseudo(Rv3128c)	-1.48	2.28E-16	-0.07	7.56E-01	-1.31	3.26E-13	-0.44	4.62E-02	-0.76	1.26E-04	-0.79	4.70E-05
RV3129	pseudo(Rv3129)	-0.51	5.78E-02	0.37	2.04E-01	-1.28	2.87E-06	-0.20	3.66E-01	-1.15	5.37E-10	-1.19	1.68E-10
RV3130C	diacylglycerol O-acyltransferase(tgs1)	-2.28	1.71E-44	-0.59	9.23E-04	0.69	3.16E-05	-0.65	9.32E-04	0.09	6.90E-01	-0.20	3.03E-01
RV3131	NAD(P)H nitroreductase(Rv3131)	-1.25	1.12E-17	0.32	5.37E-02	1.17	2.96E-16	-0.61	6.92E-03	0.73	4.68E-04	0.09	6.93E-01
RV3132C	two component sensor histidine kinase DevS(devS)	-1.02	3.11E-21	-0.04	7.84E-01	-0.91	2.01E-17	-0.26	2.46E-01	-0.45	1.65E-02	-1.11	1.07E-10
RV3133C	two component transcriptional regulator DevR(devR)	-1.42	2.07E-41	-0.05	7.07E-01	-0.90	1.28E-17	-0.16	4.89E-01	-0.14	5.23E-01	-1.02	2.84E-09
RV3136	PPE family protein PPE51(PPE51)	0.58	1.65E-18	0.10	2.29E-01	0.89	1.24E-42	0.19	3.71E-02	0.72	4.29E-20	1.04	2.94E-41
RV3177	peroxidase(Rv3177)	-0.66	1.44E-01	-0.86	8.12E-02	-2.23	7.03E-05	0.64	2.00E-01	-0.23	6.84E-01	-0.47	3.68E-01
RV3182	hypothetical protein(Rv3182)	-0.48	7.34E-05	-0.24	6.72E-02	-1.38	5.90E-27	-0.07	5.60E-01	-0.58	6.63E-09	-0.84	1.14E-16
RV3209	hypothetical protein(Rv3209)	-0.55	6.18E-04	-0.10	6.39E-01	-1.75	5.52E-29	-0.35	1.50E-02	-0.69	6.85E-08	-1.02	2.01E-16

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV3221C	acetyl-CoA carboxylase biotin carboxyl carrier protein subunit(TB7.3)	-1.04	1.44E-18	-0.09	5.81E-01	-0.87	1.58E-13	0.21	2.27E-01	0.33	3.23E-02	0.40	5.74E-03
RV3270	manganese/zinc-exporting P-type ATPase(ctpC)	-0.05	6.38E-01	0.60	1.43E-09	-0.05	6.04E-01	-1.47	2.14E-26	-0.20	2.04E-01	-1.30	1.72E-21
RV3289C	transmembrane protein(Rv3289c)	-0.92	3.70E-06	-0.64	2.51E-03	-1.26	1.84E-10	-0.51	7.72E-02	-1.45	7.17E-09	-1.13	5.25E-06
RV3290C	L-lysine-epsilon aminotransferase(lat)	-0.56	9.31E-10	-1.12	4.38E-35	-1.33	5.39E-50	-0.25	7.57E-02	-1.61	3.97E-43	-0.90	2.44E-14
RV3291C	transcriptional regulator LrpA(lrpA)	-0.77	2.89E-05	-0.39	5.32E-02	-1.15	1.11E-09	0.06	8.72E-01	-0.36	1.43E-01	-0.48	4.03E-02
RV3307	purine nucleoside phosphorylase(deoD)	0.65	2.52E-11	-0.33	2.17E-03	1.10	1.16E-30	-0.14	2.88E-01	0.27	1.77E-02	0.95	3.39E-20
RV3320C	ribonuclease VapC44(vapC44)	-1.12	5.29E-13	-0.61	1.99E-04	-1.79	5.54E-30	0.51	4.81E-02	-0.31	2.18E-01	-0.50	3.32E-02
RV3324A	pseudo(Rv3324A)	0.45	6.38E-01	0.70	5.04E-01	0.29	7.64E-01	0.71	4.25E-01	1.61	2.10E-02	0.32	7.05E-01
RV3332	N-acetylglucosamine-6-phosphate deacetylase NagA(nagA)	0.89	1.96E-21	0.15	1.83E-01	1.21	3.66E-39	-0.32	2.72E-02	0.05	7.85E-01	0.22	1.06E-01
RV3344C	PE-PGRS family protein PE_PGRS49(PE_PGRS49)	1.45	1.15E-10	0.43	1.10E-01	1.95	8.99E-19	-0.95	1.98E-02	0.68	7.57E-02	0.90	1.33E-02
RV3345C	PE-PGRS family protein PE_PGRS50(PE_PGRS50)	0.76	9.40E-04	0.40	1.25E-01	1.38	4.66E-10	-0.79	9.92E-03	0.56	5.73E-02	0.93	6.26E-04
RV3353C	hypothetical protein(Rv3353c)	-0.34	5.98E-02	-0.32	1.05E-01	-1.04	9.75E-09	-0.10	6.59E-01	-0.35	3.44E-02	-0.66	3.98E-05
RV3354	hypothetical protein(Rv3354)	0.57	2.55E-02	-0.47	1.32E-01	-0.71	1.28E-02	-0.52	2.85E-02	-0.67	2.19E-03	-1.03	1.29E-06
RV3366	tRNA/rRNA methylase SpoU(spoU)	0.67	6.18E-04	0.17	5.07E-01	1.15	8.97E-10	0.14	5.31E-01	0.17	4.01E-01	0.84	2.94E-07
RV3368C	oxidoreductase(Rv3368c)	0.65	3.65E-08	0.11	4.54E-01	1.00	2.54E-18	0.11	2.73E-01	0.48	3.29E-10	0.74	2.94E-24
RV3369	hypothetical protein(Rv3369)	0.05	5.69E-01	0.05	6.01E-01	0.53	1.34E-12	0.20	1.34E-01	0.85	1.85E-14	1.24	4.22E-31
RV3371	diacylglycerol O-acyltransferase(Rv3371)	0.07	5.69E-01	-0.94	4.97E-17	-0.38	4.73E-04	-0.76	5.42E-09	-1.19	1.34E-21	-1.16	9.19E-21
RV3376	phosphatase(Rv3376)	-0.47	3.61E-08	-0.23	1.28E-02	-1.47	4.79E-67	0.06	5.07E-01	-0.53	4.09E-14	-1.00	6.11E-48
RV3395C	hypothetical protein(Rv3395c)	-0.08	4.22E-01	0.44	2.20E-06	0.34	1.97E-04	0.17	2.93E-01	0.90	9.95E-13	1.04	3.52E-17
RV3404C	hypothetical protein(Rv3404c)	-0.84	6.17E-22	-0.06	6.11E-01	-1.22	3.35E-44	0.02	9.37E-01	-0.36	7.27E-02	-0.59	1.36E-03
RV3407	antitoxin VapB47(vapB47)	-0.76	3.82E-21	-0.11	2.66E-01	-0.11	1.97E-01	0.50	7.04E-03	0.68	5.74E-05	1.53	1.21E-21
RV3414C	ECF RNA polymerase sigma factor SigD(sigD)	-0.86	3.26E-18	-0.23	3.36E-02	-1.49	4.65E-52	0.02	9.51E-01	-0.29	8.44E-02	-0.41	9.85E-03
RV3417C	chaperonin GroEL(groEL1)	-0.95	1.35E-15	0.08	6.17E-01	-0.24	5.06E-02	-1.11	1.00E-18	-0.05	7.68E-01	-0.46	2.90E-04
RV3418C	chaperonin GroES(groES)	-1.33	7.58E-18	-0.49	4.02E-03	-0.94	1.34E-09	-1.03	1.70E-19	0.06	6.95E-01	-0.21	7.77E-02
RV3424C	hypothetical protein(Rv3424c)	0.14	4.01E-01	-0.13	4.85E-01	-1.24	1.83E-16	-0.16	6.49E-01	-1.61	2.32E-10	-1.82	2.82E-13
RV3425	PPE family protein PPE57(PPE57)	0.46	8.41E-10	0.84	2.77E-30	0.87	1.47E-32	0.87	7.53E-17	0.84	3.55E-16	1.35	1.72E-41
RV3426	PPE family protein PPE58(PPE58)	0.33	5.94E-06	0.80	2.53E-29	1.03	2.41E-49	0.50	3.66E-04	0.82	1.93E-10	1.35	1.08E-27

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV3429	PPE family protein PPE59(PPE59) bifunctional ADP-dependent (S)-NAD(P)H-hydrate dehydratase/NAD(P)H-hydrate epimerase(Rv3433c)	-0.78	8.71E-20	0.13	1.86E-01	-1.00	2.66E-32	0.30	3.09E-03	0.07	5.39E-01	0.07	4.70E-01
RV3433C		1.10	5.70E-17	0.15	3.62E-01	1.31	5.19E-24	-0.43	2.41E-03	-0.03	8.81E-01	0.15	2.82E-01
RV3434C	transmembrane protein(Rv3434c)	-0.04	6.70E-01	-0.18	3.58E-02	-0.29	1.71E-04	0.16	7.75E-02	-0.21	1.13E-02	-0.36	6.74E-06
RV3438	hypothetical protein(Rv3438)	1.14	4.88E-25	0.18	1.87E-01	1.12	4.04E-24	-0.53	5.39E-04	0.11	5.33E-01	0.00	9.87E-01
RV3444C	ESAT-6 like protein EsxT(esxT) translation initiation factor IF-1(infA)	1.48	9.91E-06	-0.05	9.36E-01	1.26	2.10E-04	0.10	7.90E-01	0.53	5.67E-02	0.54	4.45E-02
RV3462C		-1.12	1.91E-23	-0.31	1.20E-02	-1.15	4.53E-25	0.21	3.37E-01	0.12	5.50E-01	0.20	2.68E-01
RV3463	hypothetical protein(Rv3463)	0.33	3.94E-02	-0.24	1.96E-01	-0.29	8.08E-02	-0.61	4.56E-05	-0.33	2.42E-02	-1.00	2.46E-12
RV3492C	Mce associated protein(Rv3492c)	-0.81	1.14E-23	-0.66	1.03E-15	-1.00	3.34E-35	-0.12	3.99E-01	-0.50	3.37E-06	-0.62	2.81E-09
RV3503C	ferredoxin FdxD(fdxD)	-1.34	2.12E-14	-0.35	7.90E-02	-0.72	4.22E-05	0.01	9.79E-01	0.68	2.66E-04	0.78	1.59E-05
RV3507	PE-PGRS family protein PE_PGRS53(PE_PGRS53)	1.01	6.50E-07	0.21	4.16E-01	1.12	2.51E-08	-0.75	1.64E-04	0.33	9.84E-02	0.61	9.57E-04
RV3508	PE-PGRS family protein PE_PGRS54(PE_PGRS54)	0.85	2.66E-05	-0.02	9.43E-01	1.10	2.70E-08	-0.99	1.43E-04	0.25	3.67E-01	0.39	1.24E-01
RV3512	PE-PGRS family protein PE_PGRS56(PE_PGRS56)	1.48	1.43E-05	0.45	2.77E-01	1.72	2.50E-07	-1.21	1.87E-04	0.76	1.24E-02	1.24	1.17E-05
RV3528C	hypothetical protein(Rv3528c)	-1.03	4.62E-23	-0.44	7.02E-05	-1.24	1.33E-32	0.12	3.43E-01	-0.03	8.48E-01	-0.43	1.79E-05
RV3548C	short-chain type dehydrogenase/reductase(Rv3548c)	-1.13	7.83E-33	0.16	1.46E-01	-1.04	1.12E-27	-0.37	1.22E-03	0.55	8.24E-08	-0.32	2.25E-03
RV3549C	short-chain type dehydrogenase/reductase(Rv3549c)	-1.18	5.86E-39	0.27	5.68E-03	-1.25	6.88E-43	-0.38	5.37E-03	0.34	8.86E-03	-0.47	1.25E-04
RV3553	oxidoreductase(Rv3553)	-0.48	3.66E-06	0.83	1.76E-17	-0.58	3.37E-08	-0.54	9.06E-06	0.96	5.03E-19	-0.48	4.38E-05
RV3556C	acetyl-CoA acetyltransferase FadA(fadA6)	-0.37	8.95E-10	0.32	2.62E-07	-0.13	3.68E-02	-0.18	5.78E-02	0.75	1.05E-20	0.19	2.40E-02
RV3557C	HTH-type transcriptional regulator KstR2(Rv3557c)	-1.03	1.53E-52	-0.15	4.22E-02	-1.38	6.21E-89	-0.21	1.55E-02	0.02	8.64E-01	-0.72	2.42E-20
RV3570C	flavin-dependent monooxygenase oxygenase subunit HsaA(hsaA)	-1.14	1.95E-44	-0.38	9.76E-06	-1.25	1.18E-53	-0.26	4.23E-02	-0.20	9.94E-02	-0.19	1.06E-01
RV3575C	LacI family transcriptional regulator(Rv3575c)	0.24	3.58E-03	-0.08	4.31E-01	0.24	4.31E-03	0.35	8.60E-04	0.60	4.10E-10	0.41	1.55E-05
RV3576	lipoprotein LppH(lppH)	-0.61	3.63E-18	0.03	7.87E-01	-1.06	1.95E-49	0.19	4.24E-02	-0.44	1.22E-07	-0.54	4.77E-11
RV3589	A/G-specific adenine glycosylase(mutY)	1.07	2.55E-22	-0.01	9.43E-01	1.01	5.40E-20	-0.22	2.57E-01	0.39	1.74E-02	0.21	2.10E-01

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV3597C	iron-regulated H-NS-like protein(Isr2)	-1.16	4.93E-21	-0.60	5.15E-06	-1.12	6.40E-20	0.26	4.23E-01	0.40	1.37E-01	0.45	7.72E-02
RV3613C	hypothetical protein(Rv3613c)	-0.46	2.95E-03	-0.60	2.10E-04	-0.46	2.71E-03	-0.91	9.35E-06	-1.08	2.40E-08	-1.00	1.79E-07
RV3614C	ESX-1 secretion-associated protein EspD(espD)	-1.21	4.56E-40	-0.69	1.85E-13	-1.00	4.86E-28	-1.01	8.46E-15	-0.76	5.33E-09	-0.77	1.63E-09
RV3616C	ESX-1 secretion-associated protein EspA(espA)	-1.27	4.86E-49	-0.89	2.91E-24	-0.89	5.57E-25	-0.78	5.67E-08	-0.64	6.16E-06	-0.70	3.61E-07
RV3634C	UDP-glucose 4-epimerase(galE1)	1.03	1.85E-24	-0.16	2.03E-01	0.83	3.57E-16	0.17	3.94E-01	0.10	5.93E-01	-0.07	6.76E-01
RV3638	transposase(Rv3638)	0.61	2.36E-01	0.83	1.28E-01	0.50	3.39E-01	0.82	2.44E-01	0.83	1.99E-01	1.23	3.62E-02
RV3639C	hypothetical protein(Rv3639c)	1.90	6.44E-03	0.61	5.21E-01	1.32	7.33E-02	0.35	6.02E-01	0.50	3.83E-01	0.02	9.78E-01
RV3642C	hypothetical protein(Rv3642c)	-0.76	6.02E-07	0.26	1.39E-01	-1.39	9.84E-20	0.02	9.33E-01	-0.51	3.40E-03	-0.59	4.56E-04
RV3647C	hypothetical protein(Rv3647c)	0.79	3.03E-20	0.31	9.66E-04	1.05	3.15E-35	-0.30	1.79E-02	0.33	4.88E-03	0.59	1.04E-07
RV3648C	cold shock protein A(cspA)	-1.08	2.38E-29	0.13	2.59E-01	-0.55	1.15E-08	0.22	2.25E-01	0.35	2.18E-02	0.88	8.55E-10
RV3653	PE-PGRS family-related protein PE_PGRS61(PE_PGRS61)	0.69	2.73E-03	0.69	4.91E-03	1.25	1.43E-08	-0.29	2.86E-01	0.87	3.30E-05	1.02	4.24E-07
RV3655C	hypothetical protein(Rv3655c)	0.57	1.67E-01	0.73	9.25E-02	1.19	1.59E-03	0.10	8.64E-01	0.54	1.87E-01	0.79	3.77E-02
RV3657C	membrane protein(Rv3657c)	0.72	1.39E-03	0.59	1.61E-02	1.01	4.13E-06	-0.59	1.89E-02	-0.04	8.95E-01	0.16	5.06E-01
RV3670	epoxide hydrolase EphE(ephE)	0.49	1.15E-04	0.01	9.76E-01	0.13	3.54E-01	-0.13	3.32E-01	-0.32	5.71E-03	-0.40	3.63E-04
RV3675	membrane protein(Rv3675)	-1.20	1.46E-39	-0.95	4.59E-25	-1.96	1.25E-95	-0.24	1.52E-01	-0.88	3.02E-10	-1.21	1.43E-18
RV3679	anion transporter ATPase(Rv3679)	-0.26	2.61E-07	-0.41	1.10E-15	-0.62	8.50E-36	-0.07	3.37E-01	-0.65	8.87E-33	-0.72	7.23E-41
RV3681C	transcriptional regulator WhiB4(whiB4)	-0.83	3.88E-22	-0.27	2.92E-03	-1.08	3.68E-36	0.14	3.23E-01	-0.35	1.99E-03	-0.61	1.35E-08
RV3703C	iron(II)-dependent oxidoreductase EgtB(etgB)	-0.07	3.00E-01	0.01	9.37E-01	0.03	5.84E-01	-0.09	2.29E-01	-0.25	8.80E-05	0.05	4.32E-01
RV3705A	hypothetical protein(Rv3705A)	2.62	6.40E-43	0.54	1.89E-02	2.19	6.14E-30	0.80	8.11E-06	-0.01	9.79E-01	0.79	2.93E-06
RV3706C	hypothetical protein(Rv3706c)	2.90	1.15E-28	0.67	2.55E-02	2.31	7.95E-19	1.31	4.89E-09	0.46	5.09E-02	1.46	5.57E-12
RV3727	oxidoreductase(Rv3727)	0.32	1.50E-03	-1.08	1.36E-22	-0.63	3.30E-09	0.21	1.40E-01	-1.02	2.07E-15	-0.60	1.89E-06
RV3741C	oxidoreductase(Rv3741c)	-0.35	2.09E-02	-0.55	5.47E-04	-1.06	1.11E-11	-0.78	2.83E-06	-0.81	5.28E-07	-1.29	1.36E-15
RV3742C	oxidoreductase(Rv3742c)	-0.32	3.64E-02	-0.80	3.98E-07	-1.50	1.63E-20	-1.02	7.30E-13	-1.19	4.83E-17	-2.01	8.00E-42
RV3744	HTH-type transcriptional regulator NmtR(nmtR)	-0.64	3.02E-08	-0.14	3.16E-01	-1.21	2.67E-25	0.07	8.24E-01	0.05	8.44E-01	-0.18	3.84E-01
RV3764C	two component sensor kinase TcrY(tcrY)	1.76	4.86E-101	0.49	9.10E-08	1.19	3.54E-45	1.33	1.34E-45	0.17	1.33E-01	0.65	4.48E-11
RV3765C	two component transcriptional regulator TcrX(tcrX)	0.86	6.14E-33	0.43	1.28E-08	0.08	2.91E-01	1.76	7.68E-63	0.47	3.43E-05	0.80	1.30E-13
RV3768	hypothetical protein(Rv3768)	-1.04	1.33E-14	-0.33	2.27E-02	-1.34	8.90E-22	0.18	3.90E-01	-0.03	8.95E-01	-0.19	2.92E-01

BDQ-		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
RV3787C	S-adenosyl-L-methionine-dependent methyltransferase(Rv3787c)	0.69	3.11E-05	0.15	4.86E-01	0.29	9.51E-02	-0.15	4.51E-01	0.00	9.98E-01	-0.16	3.51E-01
RV3794	arabinosyltransferase A(embA)	1.04	1.36E-43	0.04	6.70E-01	1.24	1.21E-62	0.17	2.32E-01	0.55	1.72E-06	0.59	1.75E-07
RV3819	hypothetical protein(Rv3819)	0.40	2.79E-03	0.07	7.15E-01	-0.49	6.58E-04	0.10	6.47E-01	-0.90	1.90E-08	-1.16	2.81E-13
RV3833	AraC family transcriptional regulator(Rv3833)	0.10	6.88E-01	0.13	6.41E-01	-0.30	1.82E-01	-0.83	1.07E-06	-0.74	9.83E-06	-1.09	5.63E-11
RV3841	bacterioferritin BfrB(bfrB)	-0.69	2.08E-05	-0.95	1.08E-08	0.28	8.70E-02	-0.25	2.10E-01	1.07	1.99E-11	0.92	6.27E-09
RV3851	membrane protein(Rv3851)	1.00	4.69E-06	-0.10	7.59E-01	0.79	3.92E-04	-0.15	5.93E-01	-0.54	1.33E-02	-0.39	7.01E-02
RV3862C	transcriptional regulator WhiB6(whiB6)	0.76	2.14E-03	2.21	3.12E-22	0.18	5.21E-01	0.81	5.11E-04	1.96	4.76E-22	0.16	5.37E-01
RV3865	ESX-1 secretion-associated protein EspF(espF)	-1.02	3.81E-27	-0.08	5.16E-01	-0.59	4.43E-10	0.01	9.64E-01	0.20	2.59E-01	0.41	9.04E-03
RV3872	PE family protein PE35(PE35)	-0.79	1.27E-11	0.31	1.70E-02	-1.42	1.26E-34	0.01	9.79E-01	-0.27	1.50E-01	-0.31	8.11E-02
RV3874	ESAT-6-like protein EsxB(esxB)	-1.03	1.37E-18	0.33	1.10E-02	-0.69	4.45E-09	0.02	9.39E-01	0.41	8.43E-03	0.71	1.20E-06
RV3877	ESX-1 secretion system protein EccD1(eccD1)	-0.70	4.14E-20	-0.42	6.39E-08	-1.02	1.49E-38	-0.67	2.10E-10	-0.73	1.79E-12	-0.78	2.64E-14
RV3881C	ESX-1 secretion-associated protein EspB(espB)	-0.43	3.32E-09	0.26	9.14E-04	-0.25	7.24E-04	0.10	3.58E-01	0.30	4.11E-04	0.53	3.55E-11
RV3902C	hypothetical protein(Rv3902c)	-0.64	6.26E-11	-0.39	1.58E-04	-1.25	2.66E-34	0.08	5.44E-01	-0.70	5.62E-11	-0.97	1.04E-19
RV3904C	ESAT-6 like protein EsxE(esxE)	-0.52	3.31E-06	0.03	8.33E-01	0.23	3.80E-02	0.34	1.48E-01	0.61	2.68E-03	1.08	1.50E-08
RV3905C	ESAT-6 like protein EsxF(esxF)	-0.61	4.83E-04	0.01	9.85E-01	0.02	9.22E-01	0.25	2.98E-01	0.47	2.04E-02	1.15	6.42E-10
RVNC0001	ncRNA(AS1890)	-1.45	2.13E-07	-1.06	1.53E-04	-1.76	3.51E-09	-1.07	5.73E-05	-1.51	6.47E-08	-1.75	1.67E-09
RVNC0004	ncRNA(B11)	1.61	3.54E-16	0.04	8.98E-01	1.67	2.37E-17	-0.57	5.16E-03	-0.41	4.04E-02	-0.34	8.84E-02
RVNC0006	ncRNA(C8)	1.65	8.25E-03	0.19	8.26E-01	1.37	2.82E-02	-0.72	3.97E-01	1.02	1.48E-01	-0.13	8.69E-01
RVNC0012	ncRNA(mcr10)	-0.01	9.65E-01	0.60	6.46E-06	0.66	2.42E-07	0.43	1.57E-02	0.66	3.13E-05	1.22	4.92E-16
RVNC0013	ncRNA(mcr11)	-0.15	8.05E-01	-0.35	6.10E-01	-1.00	6.07E-02	-1.09	2.34E-02	0.48	3.10E-01	-0.01	9.84E-01
RVNC0015	ncRNA(mcr15)	-0.92	1.86E-05	-0.21	4.19E-01	-1.34	6.30E-10	-0.37	9.81E-02	-0.43	3.14E-02	-0.39	4.94E-02
RVNC0018	ncRNA(mcr16)	2.63	6.09E-02	0.89	6.52E-01	3.41	1.05E-02	0.38	6.73E-01	1.03	1.33E-01	0.58	4.20E-01
RVNC0021	ncRNA(mcr3)	2.41	2.31E-19	-0.46	1.43E-01	1.54	9.30E-09	-0.36	2.67E-01	0.51	6.93E-02	0.83	1.33E-03
RVNC0024	ncRNA(mcr7)	1.18	4.77E-12	-0.37	5.79E-02	0.89	1.55E-07	-0.38	2.48E-01	-0.82	2.55E-03	-0.52	5.83E-02
RVNC0034	ncRNA(MTS0858)	0.84	1.36E-01	0.19	8.03E-01	0.67	2.40E-01	-0.35	5.96E-01	-1.15	3.81E-02	-1.28	2.00E-02
RVNC0036	ncRNA(MTS1338)	0.03	9.31E-01	-0.17	7.10E-01	-0.30	4.01E-01	-1.08	3.52E-07	-0.10	6.84E-01	-1.23	2.31E-09
RVNC0046	miscRNA(ssr)	2.05	3.18E-14	-1.05	2.70E-04	2.62	8.20E-23	-1.43	1.88E-04	0.16	7.24E-01	0.17	6.74E-01
RVNS01	miscRNA(rnpB)	0.54	1.32E-01	-1.02	5.86E-03	0.93	6.73E-03	-1.17	3.56E-02	0.63	2.51E-01	0.42	4.31E-01
RVNT02	tRNA-Ala(alaT)	-0.25	1.81E-01	-0.52	7.74E-03	-1.27	5.26E-11	-0.07	7.90E-01	-1.18	1.12E-11	-1.37	8.29E-15

BDQ-		0.5 h <i>Rv0678</i> A99V vs. WT		0.5 h <i>Rv0678</i> inver- sion vs. WT		0.5 h <i>Rv0678</i> 274insA vs. WT		4 h <i>Rv0678</i> A99V vs. WT		4 h <i>Rv0678</i> inver- sion vs. WT		4 h <i>Rv0678</i> 274insA vs. WT	
RVNT03	tRNA-Leu(leuT)	0.36	4.99E-01	-0.12	8.61E-01	-0.49	3.78E-01	-0.67	2.74E-01	-0.35	5.58E-01	-1.15	3.42E-02
RVNT07	tRNA-Met(metT)	-1.11	3.84E-05	-0.29	3.81E-01	-1.70	2.41E-10	-0.05	9.35E-01	-0.10	8.14E-01	-0.35	3.36E-01
RVNT08	tRNA-Trp(trpT)	-0.80	9.43E-17	-0.37	4.12E-04	-1.19	6.50E-36	0.09	6.39E-01	-0.37	6.00E-03	-0.48	2.32E-04
RVNT11	tRNA-Glu(gluT)	1.60	3.52E-14	-0.14	6.61E-01	1.16	8.15E-08	-0.11	7.72E-01	-0.53	5.62E-02	-0.67	1.25E-02
RVNT18	tRNA-Arg(argV)	-0.11	8.37E-01	0.20	7.55E-01	-0.75	1.41E-01	0.44	4.66E-01	-0.34	5.68E-01	-1.18	3.01E-02
RVNT20	tRNA-Leu(leuV)	1.91	1.37E-01	0.89	5.98E-01	0.85	5.52E-01	-1.76	4.58E-02	-0.95	2.45E-01	-1.76	4.16E-02
RVNT22	tRNA-Leu(leuU)	0.48	6.56E-01	-1.22	3.56E-01	-0.60	5.89E-01	0.33	7.62E-01	-2.64	1.54E-02	-0.58	5.05E-01
RVNT26	tRNA-Pro(proU)	-0.68	4.44E-07	-0.15	3.46E-01	-1.02	1.27E-13	-0.27	9.85E-02	-0.63	1.81E-05	-0.68	2.93E-06
RVNT27	tRNA-Gly(glyV)	0.83	4.06E-02	-0.18	7.48E-01	0.20	6.43E-01	-0.12	8.53E-01	-0.82	7.92E-02	-1.20	8.03E-03
RVNT28	tRNA-Arg(argW)	-0.79	6.42E-02	-0.40	4.12E-01	-1.76	2.10E-04	0.66	1.32E-01	-0.05	9.26E-01	-0.41	3.68E-01
RVNT30	tRNA-Lys(lysU)	-1.07	6.95E-15	-0.53	2.92E-04	-1.83	1.48E-39	-0.27	2.14E-01	-0.86	1.82E-06	-1.21	5.40E-12
RVNT33	tRNA-Cys(cysU)	-0.72	1.65E-01	-0.66	2.59E-01	-1.99	1.10E-04	-0.29	6.15E-01	-1.08	1.36E-02	-1.21	4.44E-03
RVNT34	tRNA-Val(valU)	1.03	3.22E-01	0.45	7.40E-01	0.05	9.63E-01	0.10	9.23E-01	-0.57	4.37E-01	-1.43	4.96E-02
RVNT35	tRNA-Glu(gluU)	0.21	7.80E-01	-2.21	1.05E-02	-0.19	7.93E-01	-0.59	4.32E-01	-1.40	3.97E-02	-1.75	1.19E-02
RVNT36	tRNA-Gln(glnU)	1.00	4.66E-03	-0.35	4.56E-01	0.21	5.98E-01	-0.29	4.52E-01	-0.39	2.55E-01	-0.94	3.31E-03
RVNT38	tRNA-Met(metU)	-0.10	5.21E-01	0.06	7.66E-01	0.45	1.47E-03	1.19	2.41E-06	1.57	2.83E-11	1.84	1.42E-15
RVNT40	tRNA-Pro(proY)	-0.31	5.57E-02	-0.38	3.10E-02	-0.94	7.74E-09	-0.09	7.08E-01	-0.66	9.23E-05	-1.09	4.28E-11
RVNT42	tRNA-Ser(serX)	0.07	7.93E-01	-0.49	8.47E-02	-0.21	4.32E-01	-0.40	2.18E-01	-1.33	7.53E-06	-1.33	5.98E-06

Table 0.8: Supplementary data to Figure 3.10. Significant DEGs of BDQ exposed *Rv0678* mutants against the ancestral wild-type, significant up- and down-regulated after 0.5 h and 4 h. Significant $\log_2FC > 1$ highlighted in grey, Gene name assessed with david.ncicrf.gov/. BDQ+ = bedaquiline exposed, WT = ancestral wild-type, $\log_2FC$ = $\log_2$ fold change, *padj* = adjusted p-value.

BDQ+	<i>Gene name</i>	0.5 h <i>Rv0678</i> A99V vs. WT		0.5 h <i>Rv0678</i> inversion vs. WT		0.5 h <i>Rv0678</i> 274insA vs. WT		4 h <i>Rv0678</i> A99V vs. WT		4 h <i>Rv0678</i> inversion vs. WT		4 h <i>Rv0678</i> 274insA vs. WT	
		$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>	$\log_2FC$	<i>padj</i>
Rv0001	chromosomal replication initiator protein DnaA(dnaA)	0.66	2.13E-08	0.46	1.31E-04	1.87	6.41E-63	-0.06	7.48E-01	-0.37	1.69E-02	-0.29	6.85E-02
Rv0009	iron-regulated peptidyl-prolyl cis-trans isomerase PpiA(ppiA)	0.63	1.27E-15	0.20	1.32E-02	1.38	6.36E-74	0.87	1.03E-09	0.80	3.70E-08	1.33	1.60E-19
Rv0013	anthranilate synthase component II(trpG)	0.88	1.35E-11	0.86	2.50E-11	1.61	5.63E-39	0.08	6.79E-01	0.34	6.04E-02	0.23	2.06E-01
Rv0025	hypothetical protein(Rv0025)	-0.56	1.23E-10	-0.23	8.57E-03	-1.00	2.23E-30	0.15	2.51E-01	-0.15	2.79E-01	-0.06	6.98E-01
Rv0046c	inositol-3-phosphate synthase(ino1)	0.29	5.69E-02	-0.58	7.23E-05	0.13	3.74E-01	0.07	6.99E-01	-1.08	4.06E-12	0.07	7.38E-01
Rv0047c	hypothetical protein(Rv0047c)	0.03	7.77E-01	-1.03	2.11E-24	0.09	3.69E-01	-0.18	1.92E-01	-1.12	5.35E-18	-0.98	1.16E-13
Rv0053	30S ribosomal protein S6(rpsF)	-0.15	2.57E-01	0.33	5.31E-03	0.92	4.12E-16	1.07	4.73E-09	0.90	1.59E-06	1.67	2.84E-19
Rv0056	50S ribosomal protein L9(rplI)	-0.14	9.49E-02	0.03	7.41E-01	-0.03	7.55E-01	1.20	8.87E-52	0.77	1.32E-20	1.52	8.52E-77
Rv0057	hypothetical protein(Rv0057)	0.64	3.93E-08	0.90	5.88E-16	0.85	5.45E-14	0.97	4.82E-14	0.97	2.45E-13	1.20	2.54E-20
Rv0060	hypothetical protein(Rv0060)	-0.74	1.10E-27	-0.45	3.94E-11	-1.13	1.46E-62	0.41	1.01E-03	0.02	9.12E-01	-0.19	1.70E-01
Rv0062	cellulase CelA(celA1)	0.76	9.85E-22	0.43	1.50E-07	1.46	2.12E-80	0.44	6.17E-07	0.38	2.31E-05	0.86	6.67E-23
Rv0066c	isocitrate dehydrogenase(icd2)	-0.56	7.31E-38	-0.60	1.61E-44	-1.10	144	-0.05	6.24E-01	-0.20	4.32E-02	-0.10	3.28E-01
Rv0069c	L-serine dehydratase(sdaA)	0.25	2.89E-02	1.00	1.95E-23	1.09	1.10E-27	0.65	4.61E-08	0.77	1.82E-10	1.10	9.69E-22
Rv0070c	serine hydroxymethyltransferase(glyA2)	0.78	1.34E-06	1.03	3.42E-11	1.72	6.77E-31	0.54	2.48E-03	0.48	9.79E-03	0.88	5.66E-07
Rv0072	glutamine ABC transporter permease(Rv0072)	0.51	4.19E-14	0.70	8.12E-27	1.17	6.45E-75	0.77	4.94E-22	0.61	2.17E-13	0.89	1.90E-27
Rv0073	glutamine ABC transporter ATP-binding protein(Rv0073)	0.11	1.20E-01	0.46	2.46E-12	0.53	3.82E-16	1.01	5.64E-33	0.75	6.70E-18	1.30	9.54E-51
Rv0074	hypothetical protein(Rv0074)	-0.16	1.46E-02	0.53	7.89E-19	0.23	1.83E-04	0.89	8.00E-26	0.80	1.72E-20	1.29	1.46E-51
Rv0076c	membrane protein(Rv0076c)	-0.38	1.17E-01	-1.10	4.27E-06	-0.57	1.42E-02	-0.36	1.47E-01	-0.43	9.64E-02	-0.65	7.22E-03
Rv0077c	oxidoreductase(Rv0077c)	-0.11	4.68E-01	-1.14	7.08E-17	-0.63	2.20E-06	-0.96	6.36E-11	-1.26	2.25E-16	-1.15	4.20E-14
Rv0078B	hypothetical protein(Rv0078B)	-2.53	1.70E-27	-1.37	4.81E-09	-2.34	3.85E-24	0.45	1.33E-01	0.55	6.87E-02	0.09	7.91E-01
Rv0079	hypothetical protein(Rv0079)	-0.98	3.76E-09	-0.30	8.39E-02	-1.33	4.62E-16	-0.55	6.31E-04	0.67	2.98E-05	-0.08	6.83E-01
Rv0080	hypothetical protein(Rv0080)	-1.99	1.07E-39	-0.96	2.49E-10	-2.46	1.22E-60	0.06	8.57E-01	0.44	8.93E-02	0.17	5.54E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0081	HTH-type transcriptional regulator(Rv0081)	-1.13	4.55E-08	-0.27	1.73E-01	-0.79	6.43E-05	-0.10	6.83E-01	0.13	5.84E-01	0.37	7.95E-02
Rv0082	oxidoreductase(Rv0082)	-0.99	3.30E-08	-0.59	1.06E-03	-1.11	3.36E-10	-0.15	5.17E-01	0.43	3.93E-02	0.23	3.06E-01
Rv0087	formate hydrogenase HycE(hycE)	-0.08	6.16E-01	1.16	9.42E-18	-0.11	4.78E-01	-0.10	6.32E-01	0.33	7.20E-02	0.32	7.33E-02
Rv0088	polyketide cyclase/dehydrodrase(Rv0088)	-0.99	5.00E-27	-0.84	4.24E-20	-1.67	1.64E-75	0.07	7.05E-01	-0.06	7.57E-01	-0.12	5.21E-01
Rv0091	5'-methylthioadenosine/S-adenosylhomocysteine nucleosidase(mtn)	0.33	7.26E-02	0.40	2.49E-02	1.24	9.36E-15	0.46	6.71E-03	0.38	3.67E-02	0.28	1.18E-01
Rv0095c	hypothetical protein(Rv0095c)	-1.07	2.80E-06	-0.60	5.20E-03	-1.45	7.63E-10	0.53	1.05E-01	0.52	1.21E-01	-0.06	8.72E-01
Rv0096	PPE family protein PPE1(PPE1)	-1.37	7.13E-04	-1.05	4.51E-03	0.13	7.09E-01	-0.45	3.90E-01	-0.28	6.24E-01	0.24	6.35E-01
Rv0104	hypothetical protein(Rv0104)	-1.05	2.09E-07	-0.33	9.57E-02	-0.22	2.70E-01	-0.05	8.09E-01	-0.53	8.11E-03	-0.05	7.97E-01
Rv0106	hypothetical protein(Rv0106)	0.78	7.08E-10	0.55	1.42E-05	1.13	2.22E-20	0.63	2.00E-06	0.90	5.42E-12	0.92	1.45E-12
Rv0108c	hypothetical protein(Rv0108c)	-1.26	1.00E-28	-1.35	3.78E-33	-1.20	1.10E-26	0.52	9.48E-02	-0.10	7.83E-01	0.71	2.53E-02
Rv0111	acyltransferase(Rv0111)	0.39	3.42E-04	0.69	5.83E-11	0.71	1.08E-11	0.93	6.98E-13	0.82	8.39E-10	1.23	5.47E-21
Rv0112	GDP-mannose 4,6-dehydratase(gca)	0.12	2.72E-01	-0.08	4.75E-01	1.13	5.16E-32	0.67	6.99E-12	0.34	1.46E-03	0.89	3.32E-19
Rv0113	phosphoheptose isomerase(gmhA)	0.05	7.31E-01	-0.25	3.86E-02	0.83	4.70E-14	0.93	1.41E-12	0.53	1.94E-04	1.35	2.28E-25
Rv0115a	hypothetical protein(Rv0115a)	0.61	3.63E-05	0.11	4.92E-01	1.18	2.94E-17	1.09	5.23E-13	0.32	7.37E-02	1.31	4.22E-18
Rv0122	hypothetical protein(Rv0122)	-0.61	8.57E-04	-1.05	7.61E-09	-0.41	2.15E-02	-1.12	1.31E-12	-0.56	5.71E-04	-0.88	4.69E-08
Rv0123	hypothetical protein(Rv0123)	-0.42	4.18E-04	-1.41	2.04E-32	-1.01	4.92E-18	-1.15	1.69E-17	-1.04	2.90E-14	-1.06	8.53E-14
Rv0124	PE-PGRS family protein PE_PGRS2(PE_PGRS2)	-0.03	8.81E-01	-0.59	2.36E-04	-0.11	5.15E-01	-1.49	1.73E-15	-0.58	3.78E-03	-1.10	2.40E-08
Rv0138	hypothetical protein(Rv0138)	0.62	5.30E-04	0.98	4.45E-09	1.47	9.08E-20	0.45	3.46E-02	0.25	2.99E-01	0.31	1.64E-01
Rv0139	oxidoreductase(Rv0139)	0.36	1.21E-04	0.76	2.62E-17	0.73	4.37E-16	0.87	4.50E-14	0.72	1.83E-09	1.24	1.90E-27
Rv0140	hypothetical protein(Rv0140)	-0.36	1.45E-03	-1.04	1.60E-20	-1.10	4.84E-23	-0.18	1.99E-01	-0.70	7.17E-08	-0.71	1.59E-07
Rv0150c	hypothetical protein(Rv0150c)	1.97	2.93E-02	2.05	2.01E-02	1.52	9.68E-02	0.27	7.52E-01	0.45	5.89E-01	0.57	4.43E-01
Rv0152c	PE family protein PE2(PE2)	0.46	9.26E-02	1.20	7.45E-07	1.48	5.08E-10	-0.26	4.22E-01	0.37	2.30E-01	-0.12	7.33E-01
Rv0155	NAD(P) transhydrogenase subunit alpha PntAa(pntAa)	0.40	1.92E-04	1.08	2.17E-28	1.04	4.51E-26	1.12	1.23E-14	0.99	4.51E-11	1.20	3.67E-16
Rv0156	NAD(P) transhydrogenase subunit alpha PntAb(pntAb)	0.74	7.97E-06	1.34	2.98E-17	1.29	3.20E-16	1.48	1.23E-14	0.78	2.28E-04	1.74	3.15E-19
Rv0157	NAD(P) transhydrogenase subunit beta PntB(pntB)	0.27	7.59E-04	0.71	7.24E-20	0.28	3.86E-04	0.78	1.38E-16	0.76	1.54E-15	1.22	6.91E-37
Rv0157A	hypothetical protein(Rv0157A)	-0.76	6.27E-17	-1.08	1.41E-32	-0.76	3.59E-17	-0.14	3.93E-01	-0.12	4.95E-01	-0.03	8.77E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0161	oxidoreductase(Rv0161)	0.40	1.03E-03	0.15	2.42E-01	0.66	1.01E-08	0.95	1.36E-09	0.88	6.81E-08	1.35	9.20E-19
Rv0164	hypothetical protein(TB18.5)	0.65	4.13E-14	0.58	1.14E-11	1.66	1.73E-94	1.55	2.24E-45	1.32	6.69E-32	2.27	4.40E-92
	fatty-acid--CoA ligase												
Rv0166	FadD5(fadD5)	-1.22	6.00E-68	-0.35	3.89E-07	-0.47	9.74E-12	0.19	5.91E-02	-0.04	7.52E-01	0.82	3.36E-18
Rv0186A	metallothionein(mymT)	-2.33	3.91E-50	-2.51	4.89E-58	-2.96	9.97E-81	-0.94	1.29E-07	-1.32	6.83E-14	-1.86	4.25E-24
	transmembrane protein(Rv0188)												
Rv0188	hypothetical protein(Rv0190)	-0.61	2.83E-18	-1.42	7.70E-95	-1.42	1.36E-95	-0.08	5.06E-01	-0.62	1.74E-10	-0.73	4.98E-13
Rv0190	two component transcriptional regulator(Rv0195)	-1.16	4.58E-30	-0.86	2.51E-17	-1.40	9.08E-44	0.10	5.28E-01	-0.41	2.86E-03	-0.63	4.17E-06
Rv0195	HTH-type transcriptional regulator(Rv0196)	0.78	2.24E-01	0.41	5.32E-01	0.79	2.01E-01	1.50	1.20E-02	0.93	1.67E-01	1.79	1.90E-03
Rv0196	oxidoreductase(Rv0197)	1.21	2.04E-23	-0.23	8.76E-02	0.65	1.51E-07	0.38	5.20E-03	0.00	9.80E-01	-0.24	1.03E-01
Rv0197	hypothetical protein(Rv0203)	1.07	1.95E-46	-0.55	9.84E-13	-0.05	5.40E-01	0.41	1.60E-06	-0.03	8.23E-01	-0.27	3.58E-03
Rv0203	transmembrane transport protein MmpL3(mmpL3)	0.35	3.32E-04	-0.19	5.70E-02	0.87	6.52E-21	0.51	1.03E-04	0.33	2.05E-02	1.17	1.21E-18
Rv0206c	fatty-acid--CoA ligase	0.83	4.19E-57	0.61	9.52E-32	1.00	1.09E-83	0.20	6.03E-02	0.15	1.77E-01	0.33	1.92E-03
Rv0214	FadD4(fadD4)	0.42	1.25E-03	0.60	1.29E-06	0.64	2.62E-07	0.72	1.56E-05	0.74	1.74E-05	1.05	1.10E-10
Rv0224c	methyltransferase(Rv0224c)	0.89	5.65E-07	1.16	9.83E-12	1.64	4.14E-23	0.70	1.47E-03	0.65	5.02E-03	0.86	5.39E-05
Rv0227c	membrane protein(Rv0227c)	0.17	3.33E-02	0.05	5.25E-01	0.78	3.30E-27	0.51	2.47E-09	0.31	5.37E-04	1.10	5.22E-38
	acyl-CoA dehydrogenase												
Rv0231	FadE4(fadE4)	-0.20	6.97E-02	-0.31	3.32E-03	0.23	2.49E-02	0.45	5.11E-04	0.19	1.91E-01	1.28	3.77E-25
	ribonucleoside-diphosphate reductase subunit beta												
Rv0233	NrdB(nrdB)	-0.56	5.39E-17	-0.85	2.58E-37	-1.13	1.74E-65	-0.77	9.61E-08	-0.96	3.19E-11	-0.84	2.84E-08
Rv0236A	hypothetical protein(Rv0236A)	0.37	2.47E-01	1.24	1.47E-05	1.49	1.02E-07	1.54	5.90E-05	2.01	9.31E-08	1.95	1.30E-07
	alpha-(1->3)-arabinofuranosyl-transferase(afuD)												
Rv0236c	lipoprotein LpqI(lpqI)	1.31	6.10E-50	1.71	1.33E-86	1.68	7.78E-84	0.39	4.88E-05	0.83	6.43E-19	0.76	1.87E-15
Rv0237	acyl-CoA dehydrogenase	0.68	7.21E-14	0.89	8.04E-24	1.54	4.90E-71	1.54	1.21E-20	1.55	2.24E-20	2.08	1.64E-34
Rv0244c	FadE5(fadE5)	0.28	1.45E-02	-1.18	5.34E-28	-0.36	9.36E-04	-0.45	8.06E-04	-0.15	3.15E-01	-1.23	6.54E-20
Rv0251c	heat shock protein(hsp)	-0.21	3.44E-01	-0.80	1.35E-04	-0.50	1.73E-02	-0.62	7.68E-04	-0.58	1.91E-03	-1.45	4.42E-15
	nitrite reductase large subunit												
Rv0252	NirB(nirB)	0.32	7.24E-05	0.55	3.07E-13	1.02	1.89E-43	0.89	1.84E-16	0.77	4.11E-12	1.24	1.99E-29
	nitrite reductase small subunit												
Rv0253	NirD(nirD)	0.53	4.01E-02	0.41	1.09E-01	0.53	3.35E-02	0.67	1.69E-02	0.58	5.18E-02	1.18	5.61E-06

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0254c	bifunctional cobinamide kinase/cobinamide phosphate guanylyltransferase(cobU)	0.74	4.59E-12	-0.26	2.83E-02	1.24	5.83E-34	0.03	8.38E-01	-0.17	1.45E-01	0.38	1.33E-04
Rv0260c	transcriptional regulator(Rv0260c)	1.50	2.08E-02	1.26	5.22E-02	1.53	1.55E-02	-0.16	7.88E-01	-0.20	7.59E-01	-0.29	6.12E-01
Rv0268c	hypothetical protein(Rv0268c)	-1.08	8.70E-31	-1.33	1.46E-46	-0.99	6.80E-27	-0.47	2.83E-02	-0.98	1.29E-06	-0.85	6.35E-05
Rv0270	fatty-acid--CoA ligase FadD2(fadD2)	-0.58	5.12E-39	-0.37	3.80E-17	-1.05	7.78E-127	-0.23	4.58E-04	-0.37	1.24E-08	-0.21	1.93E-03
Rv0273c	transcriptional regulator(Rv0273c)	0.09	3.78E-01	-0.09	3.23E-01	-0.43	1.62E-06	-0.31	2.12E-02	-0.20	1.65E-01	-1.01	1.77E-14
Rv0274	hypothetical protein(Rv0274)	-0.96	1.65E-33	-0.77	3.55E-22	-1.75	1.15E-103	-0.63	3.35E-10	-0.59	8.43E-09	-1.03	1.10E-22
Rv0275c	transcriptional regulator(Rv0275c)	0.17	7.25E-02	-0.07	4.84E-01	-0.30	1.06E-03	-0.66	2.02E-05	-0.46	4.90E-03	-1.02	1.70E-10
Rv0276	hypothetical protein(Rv0276)	-0.54	3.91E-42	-0.72	4.42E-74	-1.09	1.12E-162	-0.88	1.25E-88	-0.81	1.26E-73	-1.19	1.16E-140
Rv0279c	PE-PGRS family protein PE_PGRS4(PE_PGRS4)	0.38	2.09E-01	0.17	5.88E-01	0.52	7.02E-02	-1.04	1.99E-04	0.35	2.64E-01	-0.40	2.06E-01
Rv0290	ESX-3 secretion system protein EccD(eccD3)	1.08	1.33E-30	0.30	1.98E-03	0.93	1.89E-23	-0.61	5.94E-07	0.45	4.28E-04	-0.21	1.35E-01
Rv0291	membrane-anchored mycosin MycP(mycP3)	1.04	8.45E-30	0.28	3.76E-03	1.05	7.98E-31	-0.34	1.98E-03	0.42	1.29E-04	-0.03	8.46E-01
Rv0297	PE-PGRS family protein PE_PGRS5(PE_PGRS5)	-0.33	6.33E-02	-0.14	4.46E-01	-0.15	4.04E-01	-1.14	4.41E-09	-0.22	3.25E-01	-0.56	8.24E-03
Rv0298	antitoxin(Rv0298)	-1.14	2.28E-20	-0.55	8.29E-06	-0.93	1.93E-14	0.58	5.21E-04	0.34	6.17E-02	0.52	3.17E-03
Rv0311	hypothetical protein(Rv0311)	-0.29	7.45E-05	-0.20	5.34E-03	-1.26	1.39E-62	0.13	2.37E-01	-0.32	2.77E-03	-0.24	2.48E-02
Rv0313	hypothetical protein(Rv0313)	-1.40	1.71E-19	-1.04	2.28E-11	-1.38	2.23E-19	0.05	8.74E-01	-0.17	5.76E-01	-0.37	1.90E-01
Rv0315	beta-1,3-glucanase(Rv0315)	0.92	1.26E-18	1.27	7.71E-36	1.67	4.08E-62	0.91	2.48E-10	1.12	4.44E-15	1.29	3.39E-18
Rv0316	muconolactone isomerase(Rv0316)	1.14	2.77E-09	1.67	1.79E-20	1.97	1.00E-28	0.98	1.39E-06	1.14	2.45E-08	1.46	7.66E-14
Rv0322	UDP-glucose 6-dehydrogenase UdgA(udgA)	0.67	2.11E-03	1.24	6.32E-10	1.40	1.95E-12	0.03	9.13E-01	-0.02	9.27E-01	0.33	1.06E-01
Rv0327c	cytochrome P450 Cyp135A1(cyp135A1)	0.05	8.40E-01	-0.56	9.78E-03	0.00	9.93E-01	-0.62	1.20E-02	-1.05	4.40E-05	-1.25	9.54E-07
Rv0331	dehydrogenase/reductase(Rv0331)	0.81	5.57E-10	0.39	3.54E-03	0.40	3.02E-03	-1.09	7.58E-16	-0.69	6.16E-07	-1.22	6.72E-18
Rv0335c	PE family protein PE6(PE6)	0.02	9.73E-01	-0.16	7.93E-01	-0.39	4.91E-01	-1.19	2.07E-02	-0.18	7.61E-01	-1.15	2.52E-02
Rv0340	hypothetical protein(Rv0340)	-1.20	2.26E-24	-1.05	3.44E-19	-1.65	7.37E-45	-0.53	3.42E-03	-0.83	3.89E-06	-1.16	2.24E-10

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0341	isoniazid inducible protein IniB(iniB)	-0.98	4.00E-28	-2.45	1.65E-170	-1.82	2.53E-95	-1.16	7.57E-30	-2.36	3.58E-124	-1.48	2.61E-43
Rv0342	isoniazid inducible protein IniA(iniA)	-0.67	3.30E-27	-1.43	2.85E-120	-1.70	4.49E-168	-0.69	1.24E-37	-0.89	6.96E-62	-1.18	3.18E-96
Rv0343	isoniazid inducible protein IniC(iniC)	-0.12	8.65E-02	-0.71	8.11E-27	-1.00	1.17E-51	-0.71	1.57E-18	-0.44	1.33E-07	-1.31	7.62E-54
Rv0344c	lipoprotein LpqJ(lpqJ)	0.40	3.18E-01	0.96	5.82E-03	1.02	2.99E-03	-0.37	4.59E-01	0.41	4.06E-01	0.24	6.27E-01
Rv0350	chaperone protein DnaK(dnaK) stress response protein	-0.83	6.18E-12	-1.14	1.98E-21	-1.29	1.40E-27	-0.69	3.57E-05	-0.43	1.42E-02	-0.60	5.68E-04
Rv0351	GrpE(grpE)	-0.71	1.61E-10	-0.94	1.37E-17	-1.10	6.55E-24	-0.78	2.18E-07	-0.18	3.05E-01	-0.54	8.16E-04
Rv0359	zinc metalloprotease(Rv0359)	0.25	1.37E-01	0.73	1.30E-06	1.07	6.00E-13	1.14	5.89E-06	1.43	1.52E-08	1.64	2.11E-11
Rv0360c	hypothetical protein(Rv0360c)	-0.63	3.06E-04	0.00	9.82E-01	-0.38	2.42E-02	0.19	5.23E-01	0.56	3.88E-02	1.09	4.71E-06
Rv0362	Mg2+ transport transmembrane protein MgtE(mgtE)	1.20	4.12E-12	1.27	4.08E-14	1.75	1.29E-26	0.20	2.76E-01	0.71	1.27E-05	0.20	2.56E-01
Rv0364	transmembrane protein(Rv0364)	0.84	1.68E-20	1.16	9.10E-41	2.11	5.65E-141	1.12	4.50E-16	1.09	9.02E-15	1.34	3.31E-21
Rv0379	protein translocase subunit SecE(secE2)	0.13	7.29E-01	0.24	4.85E-01	0.96	2.10E-03	2.40	2.01E-08	1.85	4.20E-05	3.30	7.26E-15
Rv0384c	chaperone protein ClpB(clpB)	-0.39	1.36E-02	-0.92	1.95E-09	-0.64	2.83E-05	-1.03	2.00E-28	-0.86	7.42E-20	-1.40	2.62E-46
Rv0385	monooxygenase(Rv0385)	0.92	3.58E-14	0.81	1.97E-11	1.01	3.09E-17	-0.01	9.66E-01	0.16	3.86E-01	0.08	6.88E-01
Rv0397A	hypothetical protein(Rv0397A)	-1.18	4.64E-32	-1.27	7.54E-38	-1.43	5.17E-47	0.32	3.80E-02	-0.18	3.06E-01	0.13	4.75E-01
Rv0402c	transmembrane transport protein MmpL1(mmpL1) membrane protein	0.84	3.47E-07	0.91	1.64E-08	1.71	1.00E-28	0.49	2.44E-02	0.49	2.95E-02	0.45	4.26E-02
Rv0403c	MmpS1(mmpS1)	0.67	1.31E-03	0.86	1.83E-05	1.16	2.99E-09	0.38	1.04E-01	0.45	6.23E-02	0.29	2.27E-01
Rv0412c	membrane protein(Rv0412c)	0.18	3.95E-02	0.27	1.56E-03	-0.55	4.51E-11	-0.58	5.18E-07	0.01	9.26E-01	-1.08	5.54E-20
Rv0414c	thiamine-phosphate synthase(thiE)	0.66	7.09E-05	0.32	6.02E-02	0.44	9.26E-03	-1.04	3.75E-06	-0.84	3.46E-04	-1.11	1.56E-06
Rv0420c	transmembrane protein(Rv0420c)	1.32	1.72E-18	0.90	4.23E-09	1.58	2.20E-27	0.32	1.40E-01	0.06	8.26E-01	0.30	1.70E-01
Rv0421c	hypothetical protein(Rv0421c)	1.31	2.21E-34	0.65	4.81E-09	0.96	1.00E-18	0.12	4.44E-01	0.39	5.02E-03	0.00	9.99E-01
Rv0428c	GCN5-like N-acetyltransferase(Rv0428c)	0.56	1.33E-03	0.93	1.09E-08	0.74	7.61E-06	0.50	7.73E-02	0.61	3.49E-02	1.13	1.25E-05
Rv0429c	polypeptide deformylase(def)	1.33	7.89E-21	1.40	1.03E-23	1.88	5.62E-43	0.69	4.41E-04	0.88	8.07E-06	1.01	1.16E-07
Rv0440	molecular chaperone GroEL(groEL2)	-0.59	2.89E-05	-2.09	5.92E-53	-0.94	8.89E-12	-0.76	1.70E-12	-0.64	3.94E-09	-0.59	2.61E-07
Rv0444c	anti-sigma-K factor RskA(rskA)	0.12	2.88E-01	0.82	2.56E-17	0.68	2.55E-12	0.50	1.07E-04	1.18	2.00E-21	0.46	6.63E-04

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0445c	ECF RNA polymerase sigma factor SigK(sigK)	0.50	2.09E-09	1.08	6.31E-44	0.89	1.00E-28	0.53	1.41E-05	1.16	8.05E-23	0.47	2.39E-04
Rv0446c	transmembrane protein(Rv0446c)	0.95	2.27E-30	1.96	2.03E-136	1.20	6.43E-50	1.34	7.02E-24	2.76	5.61E-103	1.55	8.50E-30
Rv0447c	cyclopropane-fatty-acyl-phospholipid synthase UfaA(ufaA1)	1.80	6.93E-43	2.98	1.84E-121	2.11	5.37E-60	1.45	1.12E-20	3.19	2.18E-101	1.76	4.00E-28
Rv0448c	hypothetical protein(Rv0448c)	2.31	1.41E-16	3.53	1.57E-39	2.13	2.11E-14	1.60	8.35E-07	3.91	1.61E-37	2.15	4.18E-11
Rv0449c	hypothetical protein(Rv0449c)	1.56	1.51E-54	2.40	5.57E-134	1.62	1.74E-60	1.18	3.55E-13	2.62	2.10E-62	1.40	1.68E-16
Rv0456A	toxin MazF1(mazF1)	-0.07	7.45E-01	-0.32	1.19E-01	0.19	3.50E-01	-0.26	2.29E-01	-0.44	4.43E-02	-1.09	2.19E-07
Rv0456B	antitoxin MazE1(mazE1)	-1.70	9.71E-05	-0.81	4.24E-02	-0.63	1.13E-01	1.27	5.03E-03	0.18	7.72E-01	0.30	5.80E-01
Rv0459	hypothetical protein(Rv0459)	-0.66	2.64E-13	-0.37	3.89E-05	-1.02	1.13E-29	-0.24	1.10E-01	-0.35	2.07E-02	-0.01	9.38E-01
Rv0465c	HTH-type transcriptional regulator(Rv0465c)	1.18	6.83E-22	0.19	1.62E-01	1.17	4.68E-22	-0.30	1.39E-01	-0.08	7.48E-01	-0.75	1.16E-04
Rv0471c	hypothetical protein(Rv0471c)	0.89	9.68E-09	0.35	3.17E-02	0.83	5.88E-08	-0.56	4.56E-03	-0.57	5.51E-03	-1.10	5.00E-08
Rv0474	HTH-type transcriptional regulator(Rv0474)	-0.08	3.26E-01	-0.87	6.13E-32	-0.11	1.56E-01	-0.61	5.46E-04	-0.54	2.67E-03	-1.17	7.36E-11
Rv0488	amino acid transporter(Rv0488)	0.62	3.89E-04	0.27	1.43E-01	1.08	4.65E-11	0.41	3.53E-02	0.15	5.19E-01	0.01	9.80E-01
Rv0492A	hypothetical protein(Rv0492A)	1.22	5.00E-05	0.77	1.26E-02	1.15	1.05E-04	0.07	8.39E-01	0.13	6.99E-01	0.19	5.19E-01
Rv0498	hypothetical protein(Rv0498)	1.30	1.73E-42	0.86	7.89E-19	1.49	3.95E-57	0.02	9.16E-01	0.17	1.65E-01	0.44	4.67E-05
Rv0499	hypothetical protein(Rv0499)	1.03	1.66E-24	0.89	6.25E-19	1.29	1.74E-39	0.21	9.29E-02	0.46	1.27E-04	0.52	7.51E-06
Rv0508	hypothetical protein(Rv0508)	-0.63	4.05E-24	-1.31	3.20E-96	-0.73	3.25E-32	0.06	5.06E-01	-0.59	5.06E-15	0.04	6.82E-01
Rv0516c	anti-anti-sigma factor(Rv0516c)	1.44	3.65E-21	1.26	1.57E-16	2.49	3.51E-62	1.30	5.15E-16	1.49	1.47E-20	2.32	1.19E-44
Rv0518	hypothetical protein(Rv0518)	0.47	3.41E-04	0.80	8.47E-11	0.66	1.44E-07	0.91	1.10E-06	0.76	9.94E-05	1.03	2.60E-08
Rv0522	GABA permease GabP(gabP)	0.08	3.31E-01	0.18	2.20E-02	-0.15	6.43E-02	-0.56	1.95E-08	-0.54	1.33E-07	-1.06	2.37E-24
Rv0530A	hypothetical protein(Rv0530A)	-1.44	1.83E-10	-1.00	1.09E-05	-1.41	2.26E-10	0.25	5.46E-01	0.42	3.05E-01	0.06	9.01E-01
Rv0540	hypothetical protein(Rv0540)	0.95	5.33E-08	0.90	1.86E-07	1.32	2.49E-15	0.37	2.19E-02	0.05	8.21E-01	0.33	4.44E-02
Rv0542c	2-succinylbenzoic acid--CoA ligase(menE)	1.22	2.15E-46	1.27	1.74E-51	1.56	8.27E-79	0.23	6.21E-02	0.62	7.69E-08	0.15	2.39E-01
Rv0544c	transmembrane protein(Rv0544c)	1.24	3.01E-10	1.09	2.38E-08	1.64	2.98E-18	0.01	9.76E-01	0.55	6.80E-02	0.68	1.92E-02
Rv0550c	antitoxin VapB3(vapB3)	-0.66	9.98E-07	-1.25	1.51E-20	-0.77	6.05E-09	-0.08	7.77E-01	-0.13	6.13E-01	-0.88	1.22E-04
Rv0554	non-heme bromoperoxidase BpoC(bpoC)	1.10	6.02E-17	0.81	7.58E-10	1.35	4.55E-26	0.61	1.66E-03	0.62	1.64E-03	1.04	6.24E-08
Rv0563	protease HtpX(htpX)	0.39	4.15E-05	0.01	9.47E-01	-0.07	5.05E-01	-0.26	1.28E-01	-0.31	6.26E-02	-1.38	7.40E-18
Rv0566c	hypothetical protein(Rv0566c)	-1.32	2.27E-26	-0.96	3.51E-16	-0.87	1.58E-13	0.75	3.66E-05	0.47	1.71E-02	1.34	1.82E-14

		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
BDQ+													
Rv0569	hypothetical protein(Rv0569)	-2.00	6.53E-75	-0.42	1.26E-04	-2.12	2.35E-84	-0.12	5.30E-01	1.19	1.66E-14	0.23	2.07E-01
Rv0570	vitamin B12-dependent ribo-	-1.79	6.18E-76	-0.29	3.14E-03	-2.04	2.15E-98	-0.49	1.51E-02	1.00	2.04E-07	-0.40	6.19E-02
Rv0572c	nucleoside-diphosphate re-	-2.13	1.02E-81	-1.74	1.69E-61	-2.32	3.61E-95	-0.97	8.78E-10	0.07	7.05E-01	-0.58	5.32E-04
	ductase(nrdZ)												
Rv0573c	hypothetical protein(Rv0572c)	-0.59	6.39E-03	0.03	8.99E-01	-0.65	2.22E-03	-1.19	7.89E-07	0.10	7.35E-01	-1.08	1.23E-05
Rv0575c	nicotinic acid phosphoribosyl-	-1.37	9.52E-10	-1.30	4.37E-09	-2.19	4.31E-21	0.95	2.53E-04	0.58	3.83E-02	2.29	8.18E-20
Rv0577	transferase PncB2(pncB2)	-0.84	6.74E-30	-1.03	4.32E-45	-1.22	7.18E-63	-0.25	1.42E-01	-0.28	9.68E-02	0.21	2.42E-01
	oxidoreductase(Rv0575c)												
Rv0578c	hypothetical protein(TB27.3)	-0.32	7.07E-02	-0.15	4.08E-01	-0.33	5.78E-02	-1.11	3.24E-05	0.22	4.92E-01	-0.33	2.86E-01
Rv0581	PE-PGRS family protein	-1.19	8.39E-09	-0.67	8.31E-04	-1.04	2.52E-07	0.06	8.50E-01	0.20	5.06E-01	-0.43	1.34E-01
Rv0583c	PE_PGRS7(PE_PGRS7)	-0.21	9.76E-02	-0.14	2.59E-01	0.52	2.45E-06	0.20	2.75E-01	0.40	2.41E-02	1.10	3.61E-12
Rv0599c	antitoxin VapB26(vapB26)	-0.73	2.70E-14	-0.54	9.11E-09	-1.10	3.00E-29	0.25	1.80E-01	0.27	1.61E-01	0.10	6.42E-01
Rv0608	lipoprotein LpqN(lpqN)	-1.23	4.22E-44	-1.17	9.59E-41	-1.46	2.28E-61	-0.09	6.98E-01	-0.22	2.98E-01	-0.05	8.26E-01
Rv0609A	antitoxin VapB27(vapB27)	-1.21	1.46E-46	-0.86	1.94E-27	-1.37	3.14E-59	-0.54	3.59E-06	-0.68	1.55E-08	-0.57	2.44E-06
Rv0611c	antitoxin VapB28(vapB28)	0.59	8.63E-08	0.45	5.19E-05	1.09	3.08E-24	0.16	2.34E-01	0.25	5.85E-02	0.16	2.30E-01
Rv0614	hypothetical protein(Rv0609A)	1.25	3.29E-02	1.62	3.50E-03	1.85	6.34E-04	0.29	5.87E-01	0.07	9.08E-01	0.09	8.69E-01
Rv0623	hypothetical protein(Rv0611c)	-0.69	4.29E-10	-0.88	1.03E-15	-1.10	1.70E-23	0.05	8.33E-01	-0.02	9.47E-01	-0.38	4.46E-02
	hypothetical protein(Rv0614)												
Rv0625c	antitoxin VapB30(vapB30)	0.80	4.27E-07	0.73	3.55E-06	1.21	1.35E-15	0.18	4.01E-01	0.41	3.92E-02	0.17	4.18E-01
Rv0634A	transmembrane pro-	-1.29	3.60E-70	-0.69	1.37E-21	-0.90	3.66E-36	0.83	6.40E-14	0.35	3.33E-03	1.05	4.64E-20
	tein(Rv0625c)												
Rv0636	hypothetical protein(Rv0634A)	-0.18	6.25E-03	-0.10	1.30E-01	0.38	4.73E-10	0.72	5.02E-21	0.46	1.04E-08	1.16	3.35E-50
	(3R)-hydroxyacyl-ACP dehy-												
Rv0637	dratase subunit HadB(hadB)	-0.37	2.69E-07	-0.11	1.43E-01	0.26	3.20E-04	0.88	7.03E-24	0.64	2.57E-12	1.29	9.32E-47
	(3R)-hydroxyacyl-ACP dehy-												
Rv0645c	dratase subunit HadC(hadC)	-1.26	1.27E-65	-0.94	2.54E-38	-0.99	1.09E-42	0.28	1.27E-01	-0.32	9.09E-02	0.02	9.40E-01
Rv0646c	mycolic acid methyltransfer-	0.98	8.10E-20	1.14	1.20E-26	1.16	9.00E-28	0.35	1.73E-02	0.64	6.55E-06	0.94	2.05E-11
Rv0647c	ase MmaA1(mmaA1)	1.01	2.00E-28	1.44	4.18E-58	1.58	1.68E-70	0.79	6.38E-14	0.79	1.88E-13	1.09	4.57E-24
	malonyl CoA-acyl carrier pro-												
Rv0649	tein transacylase(fabD2)	-0.18	8.10E-01	-0.04	9.57E-01	-0.04	9.56E-01	-2.24	1.34E-02	-1.71	6.11E-02	-1.37	1.04E-01
	50S ribosomal protein												
Rv0651	L10(rplJ)	0.73	1.76E-11	1.17	1.15E-28	1.24	6.91E-32	0.49	7.68E-04	0.66	7.04E-06	1.24	6.52E-19
	50S ribosomal protein												
Rv0652	L7/L12(rplL)	0.91	2.45E-15	0.94	1.75E-16	1.30	6.72E-31	0.53	4.82E-04	0.33	4.12E-02	1.58	8.50E-27

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0654	carotenoid cleavage oxygenase(Rv0654)	-0.05	5.94E-01	-0.02	8.26E-01	-0.04	5.94E-01	-1.10	5.00E-28	-0.82	8.35E-16	-0.49	3.44E-06
Rv0655	ABC transporter ATP-binding protein(mkl)	-1.14	7.94E-114	-0.24	1.78E-06	-0.94	7.60E-80	-0.69	3.57E-23	-0.40	3.28E-08	-0.33	1.50E-05
Rv0661c	ribonuclease VapC7(vapC7)	1.03	6.71E-02	1.68	1.17E-03	1.06	5.08E-02	-0.15	8.16E-01	-0.10	8.92E-01	0.13	8.41E-01
Rv0662c	antitoxin VapB7(vapB7)	1.11	1.75E-02	1.72	7.38E-05	1.22	6.78E-03	-0.33	6.30E-01	-0.10	9.01E-01	0.26	7.07E-01
Rv0667	DNA-directed RNA polymerase subunit beta(rpoB)	0.11	2.09E-01	0.10	2.18E-01	0.49	6.45E-10	0.49	1.28E-07	0.42	6.53E-06	1.10	2.72E-31
Rv0668	DNA-directed RNA polymerase subunit beta'(rpoC)	0.40	1.97E-06	0.75	3.79E-20	0.68	8.61E-17	0.48	3.54E-06	0.44	3.42E-05	1.33	1.96E-36
Rv0669c	neutral ceramidase(Rv0669c)	0.13	3.50E-01	0.58	2.20E-06	0.91	1.74E-14	0.45	7.19E-03	0.76	3.28E-06	1.04	1.56E-11
Rv0676c	transmembrane transport protein MmpL5(mmpL5)	1.05	4.37E-126	0.54	5.28E-35	0.53	1.38E-33	-0.06	5.71E-01	0.30	1.16E-03	-0.23	1.96E-02
Rv0678	transcription regulator MmpR5 (Rv0678)	-0.54	1.37E-03	-5.15	6.69E-225	-1.21	9.22E-14	0.43	6.37E-03	-3.93	3.09E-167	-0.40	1.48E-02
Rv0679c	hypothetical protein(Rv0679c)	-0.34	3.46E-03	-0.71	6.21E-10	0.26	1.96E-02	-0.85	3.93E-09	-1.06	8.27E-13	-0.49	1.05E-03
Rv0680c	transmembrane protein(Rv0680c)	-0.50	6.76E-10	-0.94	7.57E-31	-0.41	3.93E-07	-0.69	2.34E-10	-1.22	8.80E-27	-0.39	6.70E-04
Rv0681	transcriptional regulator(Rv0681)	-0.26	3.02E-04	-1.02	5.85E-47	-0.43	2.04E-09	-0.02	9.32E-01	-1.31	9.45E-17	-0.06	7.57E-01
Rv0685	elongation factor Tu(tuf)	0.57	9.04E-18	0.20	3.51E-03	0.59	3.18E-19	0.83	3.78E-19	0.06	6.41E-01	1.25	3.70E-38
Rv0686	membrane protein(Rv0686)	-0.78	9.54E-46	-1.03	7.98E-80	-0.73	1.16E-41	-0.05	5.62E-01	-0.70	6.32E-25	-0.11	1.61E-01
Rv0687	NAD-dependent oxidoreductase(Rv0687)	-0.64	1.54E-18	-1.42	5.53E-85	-0.68	3.50E-21	-0.36	1.76E-05	-1.07	8.93E-40	-0.50	4.13E-09
Rv0689c	hypothetical protein(Rv0689c)	1.29	3.65E-01	2.61	3.69E-02	1.42	2.93E-01	-0.30	7.63E-01	-0.76	4.59E-01	0.13	8.95E-01
Rv0694	mycofactocin system heme/flavin oxidoreductase MftD(Rv0694)	0.51	4.15E-10	1.22	3.32E-54	1.30	2.24E-61	0.42	2.16E-05	0.11	3.65E-01	1.38	7.44E-46
Rv0695	mycofactocin system creatinine amidohydrolase family protein MftE(Rv0695)	0.25	5.03E-03	1.25	2.05E-52	0.95	5.16E-30	0.32	6.33E-03	0.07	6.24E-01	0.99	2.79E-19
Rv0699	hypothetical protein(Rv0699)	0.34	2.88E-01	1.35	4.55E-07	0.58	4.90E-02	0.96	6.01E-04	0.65	3.52E-02	0.77	7.66E-03
Rv0701	50S ribosomal protein L3(rplC)	0.38	5.40E-06	0.18	3.27E-02	0.88	2.40E-27	0.53	1.32E-07	0.70	2.04E-12	1.13	3.00E-28
Rv0702	50S ribosomal protein L4(rplD)	-0.12	1.26E-01	-0.04	6.68E-01	0.43	7.18E-09	0.72	3.82E-12	0.96	9.03E-21	1.39	1.40E-38
Rv0703	50S ribosomal protein L23(rplW)	0.15	9.03E-02	-0.14	1.36E-01	0.69	2.81E-16	0.55	3.71E-06	0.73	5.59E-10	1.15	1.08E-21
Rv0704	50S ribosomal protein L2(rplB)	-0.12	7.87E-02	0.02	8.16E-01	0.43	1.30E-10	1.03	1.03E-25	1.00	3.48E-24	1.74	4.44E-65

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0705	30S ribosomal protein S19(rpsS)	-0.06	5.03E-01	0.32	1.96E-04	0.61	3.51E-13	1.31	7.00E-28	1.11	1.14E-19	2.06	1.32E-62
Rv0706	50S ribosomal protein L22(rplV)	0.65	4.61E-12	0.78	1.01E-16	1.13	1.62E-34	0.69	1.77E-08	0.86	2.96E-12	1.56	3.62E-36
Rv0707	30S ribosomal protein S3(rpsC)	0.01	9.06E-01	0.36	1.57E-06	0.65	3.04E-19	1.06	8.75E-20	0.77	7.61E-11	1.89	1.84E-55
Rv0708	50S ribosomal protein L16(rplP)	0.06	5.07E-01	0.50	2.15E-09	0.61	8.70E-14	1.16	3.09E-24	0.91	6.57E-15	2.16	1.39E-76
Rv0709	50S ribosomal protein L29(rpmC)	-0.41	1.72E-07	0.26	7.71E-04	0.27	4.73E-04	1.44	1.47E-42	1.04	5.78E-22	2.25	4.15E-94
Rv0710	30S ribosomal protein S17(rpsQ)	-0.35	3.61E-05	0.38	5.73E-06	0.21	1.11E-02	1.38	8.17E-50	0.79	1.14E-15	2.22	1.48E-121
Rv0711	arylsulfatase AtsA(atsA)	-0.53	2.68E-11	-1.11	2.85E-46	-0.80	6.24E-25	-0.45	8.17E-04	-1.04	4.67E-16	-0.27	5.84E-02
Rv0713	transmembrane protein(Rv0713)	0.33	1.59E-04	0.53	2.00E-10	1.00	1.92E-35	0.37	2.44E-03	0.33	9.12E-03	0.52	1.79E-05
Rv0715	50S ribosomal protein L24(rplX)	-0.11	1.28E-01	0.21	2.53E-03	0.55	2.99E-16	0.71	3.30E-14	0.59	1.26E-09	1.29	1.90E-41
Rv0716	50S ribosomal protein L5(rplE)	0.51	6.83E-13	0.40	2.90E-08	1.13	8.79E-60	0.42	1.49E-05	0.38	1.26E-04	1.19	3.30E-35
Rv0717	30S ribosomal protein S14(rpsN1)	-0.33	5.24E-04	-0.31	9.92E-04	0.56	3.96E-10	0.87	1.10E-11	0.89	5.71E-12	1.63	1.30E-35
Rv0718	30S ribosomal protein S8(rpsH)	-0.18	3.54E-02	-0.05	5.56E-01	0.54	3.38E-11	1.11	9.14E-18	0.84	2.68E-10	2.00	6.28E-51
Rv0719	50S ribosomal protein L6(rplF)	0.27	9.82E-04	0.51	1.04E-10	0.84	7.50E-27	0.99	2.50E-21	0.62	1.07E-08	1.81	2.18E-63
Rv0720	50S ribosomal protein L18(rplR)	-0.04	6.73E-01	0.41	2.57E-06	0.76	2.06E-19	0.87	3.36E-11	0.63	3.23E-06	1.81	6.79E-42
Rv0721	30S ribosomal protein S5(rpsE)	-0.19	8.28E-03	0.41	2.23E-09	0.47	1.05E-11	0.91	6.44E-15	0.65	6.08E-08	1.83	3.45E-52
Rv0722	50S ribosomal protein L30(rpmD)	-0.14	1.50E-01	0.72	4.32E-17	0.55	1.35E-10	1.11	8.50E-19	0.75	8.91E-09	2.03	7.98E-58
Rv0723	50S ribosomal protein L15(rplO)	0.43	1.35E-11	0.72	1.15E-30	0.60	2.77E-22	0.85	9.29E-26	0.52	1.15E-09	1.64	1.96E-85
Rv0732	preprotein translocase SecY(secY)	0.76	4.10E-27	0.74	5.78E-26	1.41	6.23E-93	0.33	1.07E-04	0.36	2.89E-05	0.54	7.24E-10
Rv0733	adenylate kinase(adk)	0.15	4.66E-02	0.22	2.82E-03	0.39	6.07E-08	0.78	4.44E-23	0.44	1.31E-07	1.12	3.95E-43
Rv0744c	transcriptional regulator(Rv0744c)	-1.40	2.38E-34	-0.83	2.84E-13	-1.45	4.13E-37	0.43	1.24E-02	-0.03	8.93E-01	0.21	2.72E-01
Rv0749A	hypothetical protein(Rv0749A)	0.36	3.65E-02	0.38	2.39E-02	1.06	1.08E-11	-0.50	6.13E-03	-0.79	1.89E-05	-0.68	2.36E-04
Rv0750	hypothetical protein(Rv0750)	-0.70	7.10E-21	-1.28	3.43E-65	-0.67	2.47E-19	-0.02	9.29E-01	-0.57	1.11E-03	-0.21	2.72E-01
Rv0755c	PPE family protein PPE12(PPE12)	-1.14	1.89E-23	-0.80	1.66E-12	-1.25	2.00E-28	-0.26	3.76E-02	-0.14	2.98E-01	-0.28	3.14E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0757	two component system response regulator PhoP(phoP)	-0.78	1.43E-39	-0.76	1.92E-38	-1.36	4.20E-117	-0.28	1.99E-04	-0.24	2.29E-03	-0.51	8.49E-11
Rv0762c	hypothetical protein(Rv0762c)	-1.10	3.91E-26	-0.65	8.27E-11	-1.38	1.91E-39	-0.64	2.27E-08	-0.14	3.09E-01	-0.75	2.57E-10
Rv0763c	ferredoxin(Rv0763c)	-1.05	2.45E-10	-0.61	9.04E-05	-1.30	9.19E-15	-0.73	3.17E-05	-0.17	3.89E-01	-0.71	8.46E-05
Rv0765c	oxidoreductase(Rv0765c)	-0.36	1.47E-04	-0.48	2.27E-07	-0.82	2.76E-18	-0.77	2.20E-16	-0.13	2.25E-01	-1.03	2.10E-25
Rv0766c	cytochrome P450 Cyp123(cyp123)	0.00	9.86E-01	0.11	2.98E-01	-0.40	1.42E-04	-0.82	7.76E-10	-0.37	9.52E-03	-1.09	2.84E-15
Rv0770	oxidoreductase(Rv0770)	-0.84	2.74E-08	-0.93	3.92E-10	-1.04	2.75E-12	-1.07	1.11E-16	-0.82	6.41E-10	-0.73	2.79E-08
Rv0771	4-carboxymuconolactone decarboxylase(Rv0771)	-1.21	2.95E-07	-1.01	6.71E-06	-1.26	5.45E-08	-0.39	1.43E-01	-0.23	4.38E-01	-0.09	7.48E-01
Rv0773c	bifunctional cephalosporin acylase/gamma-glutamyltranspeptidase(ggtA)	0.51	2.82E-09	0.94	1.43E-30	1.25	4.09E-54	1.02	3.15E-17	0.87	4.09E-12	1.50	1.69E-35
Rv0779c	transmembrane protein(Rv0779c)	1.37	5.82E-03	0.65	2.26E-01	1.55	1.26E-03	0.74	2.26E-01	1.10	6.36E-02	0.94	9.99E-02
Rv0780	phosphoribosylaminoimidazole-succinocarboxamide synthase(purC)	0.22	2.22E-01	0.40	1.48E-02	1.03	1.84E-11	0.66	8.47E-04	0.59	4.34E-03	0.71	2.74E-04
Rv0787	hypothetical protein(Rv0787)	1.12	1.07E-05	1.40	1.22E-08	1.84	1.49E-14	0.72	1.44E-02	0.87	3.38E-03	1.00	3.57E-04
Rv0787A	hypothetical protein(Rv0787A)	-1.35	2.53E-12	-0.78	5.81E-05	-1.08	1.53E-08	0.63	2.47E-02	0.47	1.12E-01	0.39	1.98E-01
Rv0791c	hypothetical protein(Rv0791c)	0.01	9.40E-01	-0.03	8.77E-01	-0.29	6.34E-02	-1.12	3.24E-11	-0.83	1.72E-06	-0.90	4.04E-07
Rv0792c	transcriptional regulator(Rv0792c)	-0.10	5.14E-01	-0.02	8.83E-01	-0.14	3.08E-01	-0.80	3.12E-07	-0.88	3.03E-08	-1.06	7.25E-11
Rv0797	insertion sequence element IS1547 transposase(Rv0797)	0.58	2.87E-02	0.43	1.02E-01	1.33	1.48E-08	0.27	2.98E-01	-0.06	8.65E-01	0.15	5.77E-01
Rv0814c	hypothetical protein(Rv0814c)	1.06	1.28E-17	-0.14	3.07E-01	1.46	1.54E-33	-0.06	6.75E-01	-0.14	3.37E-01	0.25	7.43E-02
Rv0816c	thioredoxin ThiX(thiX)	0.79	5.88E-03	1.14	2.17E-05	1.56	2.31E-09	0.52	2.71E-02	0.27	3.14E-01	0.66	3.60E-03
Rv0817c	hypothetical protein(Rv0817c)	0.21	4.10E-01	0.48	3.29E-02	1.08	3.49E-07	0.29	3.09E-01	0.54	5.03E-02	0.70	5.37E-03
Rv0824c	acyl-ACP desaturase DesA(desA1)	-1.05	8.87E-70	-0.67	1.37E-29	-1.47	7.75E-138	0.00	9.99E-01	0.08	5.60E-01	0.03	8.53E-01
Rv0827c	HTH-type transcriptional regulator KmtR(kmtR)	-1.80	8.74E-37	-1.03	4.06E-13	-2.66	2.16E-76	0.27	4.15E-01	0.41	1.94E-01	0.37	2.58E-01
Rv0828c	deaminase(Rv0828c)	0.70	2.70E-01	0.88	1.37E-01	1.54	5.42E-03	-0.35	4.60E-01	-1.05	3.08E-02	-0.22	6.51E-01
Rv0830	S-adenosylmethionine-dependent methyltransferase(Rv0830)	-0.20	2.12E-02	-0.26	1.87E-03	-0.48	1.16E-08	-0.21	6.14E-02	-0.29	1.07E-02	-1.17	1.13E-25

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0833	PE-PGRS family protein PE_PGRS13(PE_PGRS13)	-0.27	4.07E-01	0.38	2.30E-01	0.20	5.22E-01	-0.61	1.90E-01	1.10	1.35E-02	0.57	2.40E-01
Rv0836c	hypothetical protein(Rv0836c)	-0.30	5.53E-01	-0.31	5.19E-01	-0.80	1.01E-01	-1.20	1.36E-03	-0.36	3.84E-01	-1.42	2.18E-04
Rv0841	transmembrane protein(Rv0841)	2.47	1.83E-02	2.33	2.49E-02	2.10	4.47E-02	0.38	6.73E-01	0.89	2.78E-01	0.99	2.02E-01
Rv0846c	oxidase(Rv0846c)	-1.12	7.55E-22	-1.45	1.04E-35	-1.86	7.51E-57	-1.43	8.31E-24	-1.71	2.17E-32	-2.35	2.90E-54
Rv0847	lipoprotein LpqS(lpqS)	-1.03	4.62E-19	-1.37	1.14E-32	-1.77	6.63E-54	-1.28	2.90E-20	-1.39	2.29E-23	-2.10	3.04E-47
Rv0848	cysteine synthase CysK(cysK2)	-1.38	5.45E-37	-1.59	7.74E-49	-2.34	104	-1.18	7.01E-14	-1.42	1.94E-19	-1.75	1.67E-26
Rv0849	MFS-type transporter(Rv0849)	-1.16	3.83E-20	-1.59	1.66E-36	-2.14	5.02E-65	-1.34	5.63E-13	-1.70	3.26E-20	-1.92	4.22E-23
Rv0854	hypothetical protein(Rv0854)	-1.06	4.00E-28	-0.72	2.28E-14	-0.99	2.02E-25	0.08	5.43E-01	-0.27	3.09E-02	0.05	7.13E-01
Rv0856	hypothetical protein(Rv0856)	-1.40	3.25E-39	-1.27	2.05E-33	-1.29	3.41E-34	0.27	7.82E-02	-0.23	1.49E-01	-0.19	2.37E-01
Rv0858c	N-succinyldiaminopimelate aminotransferase DapC(dapC)	1.27	1.46E-11	0.96	4.12E-07	1.71	2.13E-21	0.65	1.79E-02	0.89	1.12E-03	0.91	5.05E-04
Rv0860	fatty oxidation protein FadB(fadB)	-0.14	3.33E-02	-1.19	3.09E-79	-0.72	6.93E-30	-0.12	1.79E-01	0.03	7.83E-01	0.00	9.91E-01
Rv0862c	hypothetical protein(Rv0862c)	0.57	1.15E-06	1.21	1.10E-26	0.51	9.66E-06	-0.07	6.47E-01	0.33	1.93E-02	-0.12	4.19E-01
Rv0863	hypothetical protein(Rv0863)	-0.71	2.16E-22	-0.52	9.72E-13	-1.39	4.00E-82	0.00	9.69E-01	-0.24	2.88E-03	-0.20	1.62E-02
Rv0864	cyclic pyranopterin monophosphate synthase accessory protein(moaC2)	-0.59	2.38E-19	-0.49	5.74E-14	-1.16	1.23E-71	0.23	1.13E-02	-0.15	1.13E-01	0.07	4.84E-01
Rv0867c	resuscitation-promoting factor RpfA(rpfA)	1.46	6.57E-53	1.40	1.31E-49	2.75	7.12E-197	1.20	5.00E-28	1.27	7.02E-31	1.75	2.47E-55
Rv0871	cold shock-like protein CspB(cspB)	0.40	5.75E-07	0.45	1.22E-08	1.22	5.33E-58	0.65	4.82E-06	0.68	3.19E-06	0.92	2.99E-10
Rv0872c	PE-PGRS family protein PE_PGRS15(PE_PGRS15)	-0.70	2.47E-05	-0.60	2.64E-04	-0.91	1.92E-08	-1.02	9.06E-08	-0.11	6.51E-01	-0.59	4.33E-03
Rv0873	acyl-CoA dehydrogenase(fadE10)	-0.78	1.56E-43	-0.55	2.20E-22	-1.10	3.64E-85	-0.07	4.01E-01	-0.23	4.92E-03	-0.11	1.97E-01
Rv0878c	PPE family protein PPE13(PPE13)	0.01	9.63E-01	0.42	3.17E-03	0.76	2.29E-08	0.63	1.58E-04	0.57	1.10E-03	1.38	2.27E-18
Rv0880	HTH-type transcriptional regulator(Rv0880)	0.97	3.61E-07	0.80	2.59E-05	1.58	1.06E-18	-0.31	1.22E-01	-0.20	3.81E-01	0.10	6.37E-01
Rv0882	transmembrane protein(Rv0882)	1.11	2.81E-04	1.08	3.17E-04	1.90	1.94E-11	-0.01	9.80E-01	0.02	9.68E-01	0.14	6.68E-01
Rv0890c	HTH-type transcriptional regulator(Rv0890c)	0.71	3.19E-19	1.04	1.79E-42	1.39	1.27E-76	0.66	2.65E-12	0.78	2.57E-16	1.00	2.07E-26
Rv0891c	hypothetical protein(Rv0891c)	0.57	1.35E-02	1.17	3.21E-08	1.81	5.89E-19	0.44	8.52E-02	0.51	5.36E-02	0.42	1.02E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0896	citrate synthase 1(gltA2)	-0.72	2.01E-21	-0.11	1.90E-01	-1.13	1.12E-51	-0.02	9.02E-01	-0.19	5.50E-02	0.24	1.48E-02
Rv0898c	hypothetical protein(Rv0898c)	-1.42	4.80E-27	-0.73	3.30E-08	-1.80	2.85E-42	0.57	1.44E-03	0.40	3.42E-02	0.63	6.63E-04
Rv0904c	acetyl-CoA carboxylase carboxyl transferase subunit beta(accD3)	0.86	6.50E-09	0.78	1.11E-07	1.32	8.22E-21	-0.07	7.42E-01	0.19	3.67E-01	0.14	4.68E-01
Rv0909	antitoxin(Rv0909)	-1.16	1.53E-14	-0.58	8.71E-05	-0.42	4.38E-03	0.54	1.70E-02	0.29	2.47E-01	0.23	3.49E-01
Rv0919	GCN5-like N-acetyltransferase(Rv0919)	-0.57	7.74E-03	-0.46	2.83E-02	-0.51	1.47E-02	-0.52	4.77E-03	-0.48	1.09E-02	-1.27	1.12E-11
Rv0923c	hypothetical protein(Rv0923c)	1.24	1.39E-13	0.94	2.44E-08	1.37	6.89E-17	-0.23	3.44E-01	-0.24	3.43E-01	-0.17	5.03E-01
Rv0932c	phosphate ABC transporter substrate-binding lipoprotein PstS(pstS2)	-0.74	2.12E-37	-0.59	3.41E-24	-1.11	1.42E-82	0.14	1.87E-01	0.02	8.95E-01	-0.31	2.37E-03
Rv0933	phosphate ABC transporter ATP-binding protein PstB(pstB)	0.80	1.69E-15	0.93	5.90E-21	1.49	3.93E-54	0.27	7.01E-02	0.33	2.85E-02	0.35	1.52E-02
Rv0934	phosphate ABC transporter substrate-binding lipoprotein PstS(pstS1)	0.16	3.00E-02	0.10	1.91E-01	0.28	7.65E-05	0.76	4.68E-16	0.60	3.83E-10	1.38	3.69E-48
Rv0935	phosphate ABC transporter permease PstC(pstC1)	0.41	3.60E-07	0.36	6.66E-06	0.42	9.13E-08	0.66	8.95E-10	0.69	2.20E-10	1.12	6.00E-24
Rv0936	phosphate ABC transporter permease PstA(pstA2)	0.44	3.77E-07	0.64	2.01E-14	0.68	2.93E-16	0.69	9.25E-07	0.58	6.52E-05	1.09	9.75E-15
Rv0937c	non-homologous end joining protein Ku(mku)	-0.86	7.63E-18	-0.21	3.85E-02	-1.06	9.30E-27	0.05	6.83E-01	-0.38	6.35E-04	-0.30	7.45E-03
Rv0939	bifunctional 2-hydroxyhepta-2,4-diene-1,7-dioate isomerase/cyclase/dehydrogenase(Rv0939)	-0.59	4.27E-39	-0.53	3.49E-32	-1.20	7.73E-158	-0.06	4.04E-01	-0.01	8.97E-01	-0.32	6.36E-08
Rv0941c	hypothetical protein(Rv0941c)	-1.06	4.15E-39	-1.08	1.96E-41	-1.25	2.38E-54	-0.03	8.63E-01	-0.07	7.37E-01	-0.37	2.39E-02
Rv0942	hypothetical protein(Rv0942)	0.36	7.39E-01	1.49	8.39E-02	1.37	1.14E-01	2.12	1.66E-01	3.17	2.83E-02	2.88	3.82E-02
Rv0943c	monooxygenase(Rv0943c)	3.70	3.11E-03	2.28	8.94E-02	2.67	3.85E-02	0.43	6.10E-01	0.10	9.24E-01	-0.67	4.58E-01
Rv0944	formamidopyrimidine-DNA glycosylase(Rv0944)	0.74	2.63E-02	0.69	3.40E-02	1.36	6.20E-06	0.75	1.09E-01	0.89	6.18E-02	0.83	6.49E-02
Rv0946c	glucose-6-phosphate isomerase(pgi)	-0.58	1.15E-24	-0.40	4.27E-13	-1.25	1.02E-103	-0.01	9.13E-01	-0.12	2.47E-01	0.02	8.82E-01
Rv0948c	chorismate mutase(Rv0948c)	0.91	7.67E-18	0.54	5.76E-07	1.64	2.65E-60	0.16	2.96E-01	-0.07	7.04E-01	0.04	8.13E-01
Rv0950c	hypothetical protein(Rv0950c)	0.48	2.79E-10	0.10	2.25E-01	1.21	3.89E-61	0.39	3.18E-03	0.51	8.88E-05	0.81	3.68E-10
Rv0964c	hypothetical protein(Rv0964c)	0.57	3.34E-02	0.94	1.33E-04	1.06	1.51E-05	0.84	1.82E-04	0.53	3.25E-02	0.86	1.02E-04

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv0967	copper-sensing transcriptional repressor CsoR(csoR)	-1.99	1.95E-86	-1.44	2.61E-46	-2.70	2.91E-157	-0.75	5.83E-12	-0.66	2.39E-09	-1.13	5.88E-23
Rv0968	hypothetical protein(Rv0968)	-2.14	8.28E-50	-1.74	1.47E-33	-2.80	2.91E-85	-0.51	1.08E-02	-0.46	2.35E-02	-0.97	1.14E-06
Rv0969	copper-exporting ATPase(ctpV)	-1.35	1.84E-45	-1.14	5.04E-33	-2.07	1.52E-106	-1.01	1.14E-30	-0.44	1.86E-06	-1.27	4.12E-43
Rv0970	integral membrane protein(Rv0970)	-1.15	3.70E-37	-0.97	1.47E-27	-1.51	2.62E-63	-0.82	1.88E-25	-0.38	4.69E-06	-1.16	8.09E-45
Rv0974c	acetyl-/propionyl-CoA carboxylase subunit beta(accD2)	1.39	2.73E-31	0.22	9.04E-02	0.95	1.72E-15	-0.69	2.01E-06	-0.31	5.24E-02	-0.27	1.02E-01
Rv0979A	50S ribosomal protein L32(rpmF)	1.21	2.50E-27	1.00	2.14E-19	2.16	2.00E-90	0.50	2.23E-05	0.84	2.62E-13	0.94	3.78E-16
Rv0979c	hypothetical protein(Rv0979c)	-1.34	7.71E-04	-1.43	2.31E-04	-1.10	3.94E-03	-0.66	2.34E-01	-0.44	4.66E-01	-0.55	3.27E-01
Rv0990c	hypothetical protein(Rv0990c)	0.02	9.00E-01	-0.17	3.42E-01	0.05	7.62E-01	-0.97	7.52E-12	-0.53	4.10E-04	-1.08	3.53E-13
Rv0991c	hypothetical protein(Rv0991c)	-0.78	3.20E-09	-1.81	7.76E-45	-0.96	1.20E-13	-0.71	1.47E-06	-0.88	3.07E-09	-1.30	1.54E-17
Rv0996	transmembrane protein(Rv0996)	-0.58	1.91E-16	-0.86	8.53E-35	-1.61	7.01E-120	-0.01	9.58E-01	0.02	9.18E-01	-0.74	9.89E-09
Rv0997	hypothetical protein(Rv0997)	0.35	1.48E-06	0.55	1.73E-14	0.28	1.63E-04	1.15	2.70E-27	0.71	3.23E-10	1.55	1.08E-45
Rv1006	hypothetical protein(Rv1006)	0.88	1.78E-58	1.19	1.44E-109	1.67	2.46E-219	0.64	1.18E-13	0.47	1.50E-07	1.19	1.56E-41
Rv1008	deoxyribonuclease TatD(tatD)	0.56	5.45E-04	0.40	1.49E-02	1.34	1.90E-19	0.51	2.83E-03	0.17	4.03E-01	0.76	5.68E-06
Rv1009	resuscitation-promoting factor RpfB(rpfB)	0.61	4.32E-19	0.22	1.51E-03	1.32	7.53E-87	0.28	5.41E-03	0.20	6.93E-02	0.52	2.50E-07
Rv1010	rRNA small subunit methyltransferase A(ksgA)	0.70	2.45E-18	0.70	2.23E-18	1.35	2.37E-68	0.49	1.16E-05	0.23	6.04E-02	0.75	3.53E-11
Rv1019	transcriptional regulator(Rv1019)	-0.95	4.14E-35	-0.54	1.78E-12	-1.18	1.39E-53	0.13	4.55E-01	-0.26	1.21E-01	-0.09	6.20E-01
Rv1028A	membrane protein KdpF(kdpF)	0.24	5.29E-01	0.39	2.69E-01	0.87	8.07E-03	1.14	4.44E-03	0.86	4.89E-02	1.01	1.18E-02
Rv1033c	two component transcriptional regulator TrcR(trcR)	-1.39	1.44E-23	-1.96	7.94E-44	-1.54	1.00E-28	0.32	3.85E-01	-0.56	1.22E-01	0.16	6.81E-01
Rv1038c	ESAT-6 like protein EsxJ(esxJ)	-1.44	2.98E-15	-1.09	1.32E-09	-1.29	6.37E-13	-0.10	6.25E-01	-0.28	1.65E-01	0.28	1.39E-01
Rv1039c	PPE family protein PPE15(PPE15)	0.93	1.59E-02	1.09	3.43E-03	0.58	1.42E-01	-0.65	1.14E-01	-0.27	5.56E-01	-0.37	3.72E-01
Rv1040c	PE family protein PE8(PE8)	0.26	5.50E-01	0.50	2.09E-01	0.43	2.76E-01	-0.23	6.01E-01	-0.04	9.31E-01	-1.09	7.77E-03
Rv1042c	IS2-like transposase(Rv1042c)	-1.20	1.47E-19	-1.28	2.98E-22	-0.52	6.07E-05	0.45	1.32E-02	-0.04	8.65E-01	0.10	6.53E-01
Rv1048c	hypothetical protein(Rv1048c)	-0.17	1.95E-01	-0.43	4.24E-04	0.08	5.33E-01	-0.99	3.67E-19	-0.67	3.58E-09	-1.41	1.18E-33
Rv1049	transcriptional repressor(Rv1049)	-0.79	9.69E-06	-0.75	1.63E-05	-1.10	1.10E-09	-0.59	1.27E-03	-0.30	1.26E-01	-1.02	4.40E-08
Rv1052	hypothetical protein(Rv1052)	-1.04	2.28E-38	-0.54	3.52E-12	-1.09	7.14E-43	0.33	1.92E-02	0.01	9.47E-01	-0.06	7.08E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1075c	hypothetical protein(Rv1075c)	-0.37	3.34E-04	-0.49	1.30E-06	-1.26	2.50E-32	-0.43	4.23E-03	-0.64	2.35E-05	-1.26	1.32E-16
Rv1076	lipase LipU(lipU)	-0.66	2.37E-11	-0.74	7.80E-14	-1.71	2.01E-68	-0.38	4.03E-03	-0.52	6.52E-05	-1.10	7.09E-17
Rv1081c	membrane protein(Rv1081c)	1.65	6.30E-12	1.67	1.59E-12	2.12	4.00E-20	0.11	6.79E-01	0.45	5.04E-02	0.05	8.62E-01
Rv1082	mycothiol S-conjugate amidase(mca)	-0.85	4.48E-19	-0.73	1.54E-14	-1.09	6.77E-31	0.13	5.76E-01	-0.07	7.83E-01	-0.16	4.96E-01
Rv1085c	hemolysin-like protein(Rv1085c)	0.62	1.55E-07	0.67	7.98E-09	1.32	1.02E-32	0.36	3.24E-02	0.87	1.78E-08	0.92	1.34E-09
Rv1088a	hypothetical protein(Rv1088a)	0.82	2.88E-01	0.99	1.75E-01	1.26	7.30E-02	-1.63	3.38E-02	-1.07	1.68E-01	-0.60	3.97E-01
Rv1096	glycosyl hydrolase(Rv1096)	0.52	9.04E-07	0.80	2.35E-15	1.05	5.33E-26	0.25	5.16E-02	0.31	1.70E-02	0.18	1.73E-01
Rv1097c	hypothetical protein(Rv1097c)	1.19	3.17E-25	1.42	3.28E-37	1.77	2.79E-58	0.74	4.61E-06	0.72	1.56E-05	1.42	1.47E-19
Rv1098c	fumarate hydratase(fum)	0.76	7.74E-26	1.07	3.04E-52	1.21	1.03E-65	0.72	4.70E-20	0.86	8.00E-28	1.36	1.66E-69
Rv1099c	fructose 1,6-bisphosphatase(glpX)	0.62	6.78E-16	1.01	2.37E-43	1.28	2.49E-70	1.00	5.24E-24	1.00	2.81E-23	1.32	1.98E-39
Rv1108c	exodeoxyribonuclease VII large subunit(xseA)	0.80	6.22E-19	1.13	2.97E-37	0.68	3.47E-14	-0.28	4.08E-03	-0.05	6.88E-01	-0.06	5.94E-01
Rv1113	antitoxin VapB32(vapB32)	-0.64	2.03E-02	0.26	3.18E-01	0.30	2.44E-01	0.99	2.17E-02	1.08	1.40E-02	1.58	1.08E-04
Rv1116	hypothetical protein(Rv1116)	1.58	9.51E-02	1.57	9.09E-02	2.41	5.35E-03	-0.65	4.23E-01	0.37	6.48E-01	-0.95	2.35E-01
Rv1117	hypothetical protein(Rv1117)	0.61	1.58E-07	1.10	6.83E-23	1.34	1.33E-33	0.28	7.07E-02	0.33	3.21E-02	-0.10	5.60E-01
Rv1118c	hypothetical protein(Rv1118c)	0.49	8.65E-03	0.74	2.33E-05	1.15	8.52E-12	0.57	3.10E-03	0.69	4.15E-04	1.16	3.88E-11
Rv1120c	hypothetical protein(Rv1120c)	-1.14	9.80E-03	-0.80	5.65E-02	-1.15	7.70E-03	-0.20	6.88E-01	0.46	3.17E-01	0.04	9.38E-01
Rv1129c	transcriptional regulator(Rv1129c)	0.38	1.68E-02	1.37	2.46E-21	0.74	7.66E-07	0.45	1.16E-02	-0.58	2.90E-03	0.38	3.69E-02
Rv1130	2-methylcitrate dehydratase(prpD)	1.22	3.93E-10	3.72	1.10E-96	1.92	5.12E-25	1.49	1.03E-09	2.81	1.70E-33	2.17	1.53E-19
Rv1131	methylcitrate synthase PrpC(prpC)	1.72	1.28E-10	4.17	1.22E-62	2.71	2.26E-26	1.97	1.66E-12	2.46	7.63E-19	2.48	6.00E-19
Rv1132	hypothetical protein(Rv1132)	0.55	4.00E-08	1.15	9.53E-34	1.05	4.00E-28	1.07	6.13E-16	0.86	4.49E-10	1.30	7.26E-22
Rv1134	hypothetical protein(Rv1134)	0.72	2.91E-02	0.24	4.99E-01	1.10	3.46E-04	-0.04	9.25E-01	-0.55	1.23E-01	-0.18	6.12E-01
Rv1138a	hypothetical protein(Rv1138a)	0.51	2.90E-01	1.00	1.92E-02	1.10	9.12E-03	0.36	4.35E-01	0.05	9.25E-01	0.23	6.16E-01
Rv1146	transmembrane transport protein(Rv1146)	-0.43	4.09E-05	-0.13	2.38E-01	-0.20	5.63E-02	0.65	8.66E-06	0.49	1.65E-03	1.06	2.82E-13
Rv1149	transposase(Rv1149)	-1.29	3.93E-54	-0.81	9.63E-23	-1.26	5.66E-53	0.09	5.67E-01	-0.27	8.06E-02	-0.21	1.78E-01
Rv1152	transcriptional regulator(Rv1152)	-0.72	1.86E-14	-0.75	4.57E-16	-1.38	3.13E-49	-0.08	6.31E-01	-0.33	2.35E-02	-0.53	2.12E-04
Rv1155a	hypothetical protein(Rv1155a)	-1.67	2.98E-25	-1.09	9.00E-12	-1.74	1.50E-27	0.70	2.18E-02	0.11	7.68E-01	0.71	2.55E-02
Rv1156	hypothetical protein(Rv1156)	-0.81	1.55E-46	-0.62	1.14E-27	-1.31	120	0.01	9.26E-01	-0.20	4.69E-03	-0.01	9.23E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1159A	4a-hydroxytetrahydrobiop- terin dehydratase(Rv1159A)	-1.78	4.30E-48	-1.40	1.86E-30	-2.55	6.48E-98	0.30	1.21E-01	-0.18	4.02E-01	0.04	8.68E-01
Rv1170	1D-myo-inositol 2-acetamido- 2-deoxy-alpha-D-glucopyra- noside deacetylase(mshB) NADPH dependent 2,4- dienoyl-CoA reductase	0.64	3.12E-08	0.83	1.47E-13	1.07	3.16E-22	0.47	3.04E-04	0.36	9.74E-03	0.80	2.93E-10
Rv1175c	FadH(fadH)	0.98	2.10E-31	1.09	2.74E-40	1.46	1.22E-73	0.80	2.32E-15	0.75	6.47E-13	1.42	4.72E-46
Rv1176c	hypothetical protein(Rv1176c)	1.25	8.19E-06	1.36	6.31E-07	2.18	5.63E-17	0.06	8.39E-01	0.42	1.10E-01	0.38	1.29E-01
Rv1179c	hypothetical protein(Rv1179c)	0.92	8.70E-37	1.09	1.38E-52	1.36	1.45E-82	0.23	3.10E-03	0.09	3.05E-01	0.35	7.57E-06
Rv1184c	hypothetical protein(Rv1184c)						1.22E-						
	fatty-acid--CoA ligase	1.36	4.65E-63	1.33	7.89E-62	2.18	170	0.61	2.91E-06	0.43	1.64E-03	0.88	3.53E-11
Rv1185c	FadD21(fadD21)	0.27	2.28E-05	0.45	1.34E-13	0.59	3.10E-22	0.84	4.49E-21	0.48	1.58E-07	1.12	3.31E-33
Rv1194c	hypothetical protein(Rv1194c)	0.89	1.73E-38	0.79	2.87E-31	1.39	1.44E-95	0.35	5.91E-03	0.17	2.13E-01	-0.04	7.86E-01
Rv1195	PE family protein PE13(PE13)	0.84	3.35E-18	1.03	1.55E-26	1.54	1.35E-58	1.87	1.12E-68	1.57	1.10E-48	2.59	9.60E-118
	PPE family protein												
Rv1196	PPE18(PPE18)	0.59	3.90E-09	1.07	3.97E-27	1.30	1.27E-39	2.22	3.05E-48	1.94	5.50E-37	3.32	1.52E-95
Rv1197	ESAT-6 like protein EsxK(esxK)	0.00	9.79E-01	0.64	8.52E-08	0.59	7.23E-07	2.05	7.32E-60	1.50	5.01E-32	3.39	7.34E-145
Rv1198	ESAT-6 like protein EsxL(esxL)	0.38	3.64E-04	1.05	2.45E-24	0.95	1.51E-20	1.94	5.69E-94	1.40	3.99E-49	3.35	2.69E-250
Rv1201c	(dapD)	-0.03	6.88E-01	-0.08	1.99E-01	0.62	2.73E-31	0.64	4.40E-16	0.21	1.76E-02	1.29	4.33E-58
	integral membrane pro- tein(Rv1216c)	-1.09	5.08E-37	0.20	1.87E-02	-1.32	6.66E-54	-0.55	6.05E-05	-0.10	5.30E-01	-0.58	5.45E-05
	tetronasin ABC transporter in- tegral membrane pro- tein(Rv1217c)	-0.62	1.01E-14	0.82	2.51E-26	-0.71	4.06E-19	-1.27	2.10E-21	-0.09	5.74E-01	-1.24	1.40E-18
Rv1218c	tetronasin ABC transporter ATP-binding protein(Rv1218c)	-1.16	1.41E-44	0.51	4.15E-11	-1.33	1.10E-58	-1.02	4.25E-13	-0.20	2.01E-01	-1.07	4.03E-13
Rv1219c	transcriptional regula- tor(Rv1219c)	-0.97	1.56E-19	0.75	2.47E-15	-0.67	1.20E-10	-1.11	2.04E-14	-0.53	5.32E-04	-1.62	6.92E-26
	ECF RNA polymerase sigma factor SigE(sigE)	0.22	4.76E-03	-0.19	1.79E-02	-0.12	1.33E-01	-0.61	8.73E-08	-0.53	3.95E-06	-1.10	8.34E-21
Rv1221	ribonuclease VapC33(vapC33)	0.61	9.46E-03	0.82	2.56E-04	1.09	6.08E-07	1.24	4.15E-06	1.17	3.19E-05	1.42	7.42E-08
Rv1244	lipoprotein LpqZ(lpqZ)	1.21	1.46E-19	1.10	1.05E-16	1.82	8.77E-46	-0.11	5.12E-01	0.01	9.49E-01	0.14	3.84E-01
	serine/threonine-protein ki- nase PknH(pknH)	-0.85	2.24E-33	-0.55	2.25E-15	-1.22	2.17E-67	-0.12	3.35E-01	-0.25	3.51E-02	-0.38	9.97E-04
Rv1268c	hypothetical protein(Rv1268c)	1.02	4.11E-02	-0.09	8.85E-01	1.54	8.66E-04	-1.11	1.45E-02	-0.81	8.83E-02	-0.92	4.51E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1282c	oligopeptide ABC transporter permease OppC(oppC)	1.09	1.11E-19	0.93	1.20E-14	0.99	1.13E-16	0.18	3.83E-01	0.30	1.23E-01	0.61	6.22E-04
Rv1284	beta-carbonic anhydrase(canA)	-1.32	7.40E-25	-1.85	6.15E-47	-1.79	9.78E-44	-0.98	5.05E-12	-0.99	9.85E-12	-1.07	5.17E-13
Rv1285	sulfate adenylyltransferase subunit 2(cysD)	-0.48	3.19E-07	-0.23	1.45E-02	-1.23	8.83E-42	-0.64	1.25E-07	-0.19	1.61E-01	-0.88	3.52E-12
Rv1292	arginine--tRNA ligase(argS)	0.74	7.79E-17	0.63	8.87E-13	1.12	1.34E-38	-0.20	2.34E-01	0.01	9.59E-01	-0.42	8.05E-03
Rv1302	decaprenyl-phosphate N-acetylglucosaminephosphotransferase(rfe)	1.09	4.54E-40	0.73	1.51E-18	1.56	3.51E-84	-0.03	8.11E-01	0.17	9.09E-02	0.30	1.24E-03
Rv1304	ATP synthase subunit A(atpB)	-0.36	5.14E-08	-1.01	7.59E-54	-0.66	4.64E-24	-0.58	3.22E-06	-0.55	1.31E-05	-0.63	8.92E-07
Rv1305	ATP synthase subunit C(atpE)	-0.72	9.66E-24	-1.11	4.25E-55	-0.85	3.61E-33	-0.38	1.66E-04	-0.27	8.46E-03	-0.24	2.75E-02
Rv1319c	adenylate cyclase(Rv1319c)	0.65	1.29E-07	0.98	2.59E-17	1.07	1.49E-20	0.20	3.08E-01	0.12	5.99E-01	0.40	3.46E-02
Rv1320c	adenylate cyclase(Rv1320c)	0.84	1.29E-17	1.14	4.75E-33	1.54	4.91E-61	0.32	2.44E-02	0.37	9.79E-03	0.26	7.95E-02
Rv1331	ATP-dependent Clp protease adapter protein ClpS(Rv1331)	-0.85	4.00E-28	-0.41	2.11E-07	-1.04	4.98E-42	0.20	1.65E-01	-0.13	3.81E-01	-0.63	2.65E-06
Rv1343c	lipoprotein LprD(lprD)	0.82	7.60E-07	0.49	3.82E-03	1.37	5.50E-18	-0.04	8.78E-01	0.17	4.71E-01	0.44	3.55E-02
Rv1344	acyl carrier protein MbtL(mbtL)	0.18	3.72E-01	0.80	5.22E-06	1.31	1.83E-14	1.56	8.37E-13	1.36	1.90E-09	2.40	8.40E-30
Rv1347c	lysine N-acetyltransferase MbtK(mbtK)	1.32	1.35E-11	1.25	1.11E-10	2.49	8.16E-43	0.20	5.24E-01	0.64	2.10E-02	1.04	5.01E-05
Rv1348	iron ABC transporter ATP-binding protein/permease IrtA(irtA)	1.26	5.27E-46	1.64	1.85E-79	2.21	2.64E-146	0.69	3.50E-06	0.87	3.15E-09	1.42	2.54E-21
Rv1349	iron ABC transporter ATP-binding protein/permease IrtB(irtB)	0.93	1.58E-23	1.12	1.40E-34	1.92	4.41E-103	0.85	6.43E-08	1.11	1.22E-12	1.72	3.00E-28
Rv1352	hypothetical protein(Rv1352)	-1.26	1.14E-26	-0.78	2.18E-11	-1.57	3.06E-41	0.07	7.63E-01	-0.09	6.87E-01	0.24	2.53E-01
Rv1353c	HTH-type transcriptional regulator(Rv1353c)	1.03	3.73E-04	1.49	4.68E-08	1.29	3.38E-06	0.23	3.97E-01	0.28	3.27E-01	-0.32	2.44E-01
Rv1360	oxidoreductase(Rv1360)	0.63	1.44E-10	1.38	7.30E-49	1.16	1.46E-34	0.28	1.41E-02	0.31	7.11E-03	0.14	2.51E-01
Rv1361c	PPE family protein PPE19(PPE19)	0.22	1.77E-02	0.49	4.59E-08	0.69	1.90E-15	0.71	3.36E-09	0.65	8.85E-08	1.09	1.45E-18
Rv1366A	hypothetical protein(Rv1366A)	-1.02	2.00E-28	-0.84	1.24E-20	-1.02	2.41E-29	0.20	2.38E-01	-0.15	3.94E-01	0.08	6.77E-01
Rv1378c	hypothetical protein(Rv1378c)	-0.59	2.07E-18	-0.68	6.29E-24	-1.26	1.22E-78	-0.06	6.68E-01	0.13	3.55E-01	-0.58	3.62E-06
Rv1387	PPE family protein PPE20(PPE20)	0.29	1.49E-04	0.69	2.79E-20	0.28	2.30E-04	0.79	3.31E-26	0.81	2.60E-27	1.15	4.16E-49

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
	integration host factor												
Rv1388	MihF(mihF)	0.22	1.79E-04	0.13	2.80E-02	0.31	6.42E-08	0.79	1.41E-26	0.71	3.02E-21	1.24	1.12E-59
Rv1397c	ribonuclease VapC10(vapC10)	0.06	4.21E-01	-0.13	7.58E-02	-0.37	3.37E-08	-0.81	2.07E-12	-0.68	8.04E-09	-1.56	3.90E-38
Rv1398c	antitoxin VapB10(vapB10)	-0.40	2.44E-03	-0.62	1.87E-06	-0.27	3.69E-02	-0.93	3.59E-18	-0.64	4.50E-09	-1.34	5.81E-33
Rv1402	primosomal protein N'(priA)	0.66	1.76E-13	1.20	4.20E-46	1.17	9.74E-44	0.39	2.44E-03	0.47	3.28E-04	0.65	4.04E-07
Rv1403c	methyltransferase(Rv1403c)	-0.36	2.60E-01	-1.68	2.99E-07	-1.02	1.02E-03	0.10	7.63E-01	-0.10	7.84E-01	0.39	1.96E-01
Rv1415	(ribA2)	-0.68	1.00E-28	-0.39	3.23E-10	-1.04	1.42E-66	-0.07	5.53E-01	-0.06	5.70E-01	-0.25	1.47E-02
Rv1419	hypothetical protein(Rv1419)	-0.87	5.27E-22	-0.80	5.53E-19	-1.04	3.12E-31	-0.14	3.80E-01	-0.25	8.92E-02	-0.31	3.81E-02
Rv1439c	hypothetical protein(Rv1439c)	0.73	2.75E-03	1.01	1.04E-05	1.45	7.50E-11	0.18	4.54E-01	-0.74	3.19E-03	0.24	2.91E-01
Rv1457c	antibiotic ABC transporter permease(Rv1457c)	0.96	1.24E-16	0.81	2.54E-12	1.20	4.13E-26	-0.18	4.07E-01	0.27	1.97E-01	0.08	7.26E-01
Rv1460	transcriptional regulator(Rv1460)	0.48	4.35E-09	0.01	9.12E-01	-0.16	6.72E-02	-0.84	4.72E-08	-0.11	5.47E-01	-1.25	6.32E-15
Rv1461	hypothetical protein(Rv1461)	-0.24	4.87E-03	-0.74	1.66E-18	-1.12	1.65E-41	-0.32	1.36E-03	-0.02	8.54E-01	-0.74	1.84E-13
Rv1462	hypothetical protein(Rv1462)	0.12	6.87E-02	-0.68	1.38E-26	-0.68	9.80E-27	-0.63	5.44E-09	0.03	8.11E-01	-1.01	4.10E-19
Rv1463	ABC transporter ATP-binding protein(Rv1463)	0.63	1.58E-16	-0.32	3.67E-05	-0.40	1.95E-07	-0.92	1.54E-09	-0.08	6.71E-01	-1.11	3.26E-12
Rv1464	cysteine desulfurase(csd)	0.76	8.01E-22	-0.31	9.88E-05	-0.08	3.59E-01	-0.76	7.08E-10	-0.11	4.73E-01	-1.11	7.80E-18
Rv1466	hypothetical protein(Rv1466)	0.13	7.91E-02	-1.03	6.61E-49	-0.38	9.40E-08	-0.59	8.51E-06	-0.21	1.61E-01	-0.89	6.60E-11
Rv1470	thioredoxin TrxA(trxA)	-0.64	2.72E-04	-1.23	3.03E-12	-0.42	1.56E-02	-0.01	9.80E-01	-0.43	2.39E-02	-0.17	3.80E-01
Rv1473A	transcriptional regulator(Rv1473A)	-1.54	1.02E-04	-0.19	6.37E-01	-0.99	8.84E-03	2.01	1.56E-03	2.15	8.82E-04	1.92	2.79E-03
Rv1476	membrane protein(Rv1476)	-0.81	6.28E-24	-0.82	5.42E-25	-1.03	1.00E-38	0.30	6.24E-02	-0.06	7.58E-01	0.33	4.31E-02
Rv1482c	hypothetical protein(Rv1482c)	0.33	7.19E-01	1.54	3.49E-02	0.79	3.16E-01	0.71	4.37E-01	0.88	3.38E-01	0.93	2.68E-01
Rv1483	3-oxoacyl-ACP reductase FabG(fabG1)	0.63	2.00E-28	0.48	7.38E-17	1.09	5.96E-86	0.56	1.91E-16	0.45	1.79E-10	0.62	1.43E-18
Rv1485	ferrochelatase(hemZ)	0.73	3.64E-19	0.65	1.09E-15	1.06	6.90E-41	0.75	7.26E-15	0.50	1.20E-06	1.10	1.32E-29
Rv1497	esterase LipL(lipL)	-0.75	3.10E-27	-0.81	4.35E-32	-1.10	2.23E-57	-0.16	3.39E-01	-0.45	3.37E-03	-0.50	1.33E-03
Rv1498A	hypothetical protein(Rv1498A)	-1.07	2.19E-15	-0.94	3.67E-13	-0.64	5.10E-07	0.04	8.40E-01	-0.14	5.13E-01	0.05	8.16E-01
Rv1501	hypothetical protein(Rv1501)	-0.90	8.47E-26	-1.02	1.93E-33	-0.74	1.37E-18	-0.06	5.89E-01	-0.50	1.44E-07	-0.83	2.08E-17
Rv1502	hypothetical protein(Rv1502)	-0.97	7.57E-46	-1.05	3.45E-54	-1.17	2.77E-67	-0.05	6.52E-01	-0.57	3.64E-10	-0.68	3.34E-13
Rv1507A	hypothetical protein(Rv1507A)	0.34	5.14E-01	0.97	3.07E-02	1.36	1.63E-03	-0.74	6.04E-02	-0.18	6.88E-01	-0.85	2.90E-02
Rv1509	hypothetical protein(Rv1509)	-0.93	5.97E-26	-0.82	6.44E-21	-1.17	2.70E-40	-0.07	5.22E-01	-0.46	5.07E-06	-0.54	6.75E-08
Rv1514c	glycosyltransferase(Rv1514c)	-0.14	3.55E-01	-0.01	9.73E-01	0.56	1.33E-05	1.16	9.48E-11	0.53	1.01E-02	1.32	1.35E-13
Rv1516c	sugar transferase(Rv1516c)	0.90	2.13E-07	0.68	9.09E-05	1.23	1.34E-13	0.47	4.74E-02	0.39	1.24E-01	0.50	3.65E-02
Rv1517	hypothetical protein(Rv1517)	-1.41	2.92E-26	-1.23	1.52E-21	-1.36	1.43E-25	-0.48	2.39E-02	-0.79	1.92E-04	-0.46	3.46E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1519	hypothetical protein(Rv1519)	1.33	1.63E-14	1.33	6.69E-15	2.50	6.00E-53	0.22	2.58E-01	0.06	8.09E-01	0.15	4.53E-01
Rv1532c	hypothetical protein(Rv1532c)	0.81	2.42E-03	0.80	2.30E-03	1.10	1.40E-05	1.11	1.49E-03	0.61	1.23E-01	1.44	2.28E-05
Rv1534	transcriptional regulator(Rv1534)	0.46	2.00E-05	0.76	6.58E-14	1.09	1.00E-27	1.17	1.06E-16	1.00	1.04E-11	1.60	8.93E-31
Rv1545	hypothetical protein(Rv1545)	-0.06	4.94E-01	0.08	3.53E-01	0.23	2.78E-03	1.09	2.00E-28	0.54	5.26E-07	1.36	2.08E-42
Rv1546	hypothetical protein(Rv1546)	-0.30	2.78E-03	0.21	2.78E-02	0.51	4.32E-08	0.85	1.10E-06	0.82	5.05E-06	1.35	1.28E-14
Rv1552	fumarate reductase flavoprotein subunit(frdA)	-1.53	1.94E-21	-1.06	2.63E-12	-1.47	1.17E-20	-0.63	6.65E-03	-1.22	3.24E-07	-0.21	4.06E-01
Rv1558	hypothetical protein(Rv1558)	0.36	1.44E-03	0.65	1.08E-09	1.22	9.32E-33	0.93	1.66E-12	0.80	4.04E-09	1.21	1.95E-20
Rv1567c	membrane protein(Rv1567c)	0.48	1.33E-02	1.15	6.88E-11	1.35	9.94E-15	0.85	2.05E-05	0.62	4.31E-03	0.65	1.62E-03
Rv1573	phage protein(Rv1573)	0.98	1.05E-03	1.54	3.16E-08	1.21	2.48E-05	0.87	6.29E-03	0.51	1.60E-01	1.26	2.58E-05
Rv1584c	phage protein(Rv1584c)	-0.53	7.61E-02	-1.86	1.41E-10	-0.50	8.51E-02	-0.84	5.17E-04	-0.83	7.81E-04	-0.81	1.28E-03
Rv1585c	phage protein(Rv1585c)	-0.75	2.71E-12	-1.50	4.15E-45	-1.20	5.11E-30	-0.99	1.72E-16	-0.92	2.27E-14	-1.17	1.91E-20
Rv1586c	phage integrase(Rv1586c)	-0.69	1.31E-11	-1.29	3.23E-36	-1.08	2.35E-26	-1.17	1.91E-18	-0.70	3.74E-07	-1.10	7.10E-15
Rv1587c	hypothetical protein(Rv1587c)	0.94	1.59E-01	1.32	3.72E-02	0.41	5.53E-01	-0.18	7.52E-01	0.34	5.47E-01	-0.84	1.17E-01
Rv1588c	hypothetical protein(Rv1588c)	-0.97	2.52E-13	-0.53	5.18E-05	-1.37	1.43E-24	-0.03	9.13E-01	-0.11	6.26E-01	-0.37	5.01E-02
Rv1611	indole-3-glycerol phosphate synthase(trpC)	0.33	4.29E-08	0.87	1.11E-51	1.11	9.35E-83	0.85	4.18E-43	0.80	6.77E-37	1.16	6.98E-75
Rv1612	tryptophan synthase subunit beta(trpB)	0.90	9.26E-47	1.49	5.58E-134	1.83	3.52E-204	0.79	5.23E-12	0.84	5.43E-13	1.36	1.29E-30
Rv1613	tryptophan synthase subunit alpha(trpA)	0.08	2.90E-01	1.01	8.85E-50	0.83	3.16E-34	1.27	8.88E-20	1.13	8.70E-16	2.00	1.42E-43
Rv1614	prolipoprotein diacylglycerol transferase(lgt)	0.20	5.19E-04	1.00	1.15E-76	1.01	8.01E-79	1.06	7.45E-31	0.83	1.66E-18	1.61	2.13E-64
Rv1617	pyruvate kinase(pykA)	0.47	2.23E-16	0.60	1.08E-26	0.89	2.23E-57	0.63	1.18E-30	0.64	8.00E-30	1.01	4.72E-75
Rv1618	acyl-CoA thioesterase II(tesB1)	0.18	2.12E-02	0.75	7.77E-27	0.77	1.00E-27	0.64	6.44E-15	0.49	9.73E-09	1.23	1.73E-51
Rv1620c	cytochrome biosynthesis ABC transporter ATP-binding protein/permease CydC(cydC)	0.14	1.57E-01	-1.31	2.80E-48	0.08	4.12E-01	-0.69	4.69E-11	-0.90	8.05E-18	-0.90	2.68E-16
Rv1621c	cytochrome biosynthesis ABC transporter ATP-binding protein/permease CydD(cydD)	0.28	1.26E-03	-0.78	5.10E-21	-0.09	2.99E-01	-1.06	2.23E-32	-0.65	9.78E-13	-1.29	7.41E-43
Rv1622c	cytochrome D ubiquinol oxidase subunit II CydB(cydB)	-0.49	9.04E-18	-1.12	3.97E-89	-0.94	3.39E-63	-1.01	9.80E-19	-0.70	1.96E-09	-1.12	1.15E-20
Rv1623c	cytochrome D ubiquinol oxidase subunit I CydA(cydA)	-0.05	5.32E-01	-0.66	2.74E-22	-0.45	3.79E-11	-0.99	2.61E-11	-0.88	4.75E-09	-1.25	1.32E-15
Rv1632c	hypothetical protein(Rv1632c)	1.39	1.78E-06	1.69	1.31E-09	2.02	1.75E-13	-0.22	4.84E-01	-0.07	8.50E-01	-0.33	2.69E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
	iron-regulated universal stress protein(TB15.3)	-0.42	3.46E-15	-1.04	2.10E-85	-0.66	2.72E-35	-0.18	7.30E-03	-0.52	7.74E-16	-0.52	6.88E-15
Rv1636	hypothetical protein(Rv1638A)	0.08	6.16E-01	0.02	8.92E-01	0.27	6.71E-02	0.37	2.96E-02	0.11	6.03E-01	1.16	4.50E-14
Rv1649	phenylalanine--tRNA ligase subunit alpha(pheS)	0.75	2.47E-18	0.60	3.42E-12	1.27	3.26E-54	0.11	3.26E-01	0.14	2.23E-01	0.46	3.60E-06
Rv1650	phenylalanine--tRNA ligase subunit beta(pheT)	0.78	2.32E-36	0.76	9.07E-36	1.23	1.16E-92	0.37	6.28E-08	0.40	6.23E-09	1.00	7.64E-51
Rv1669	hypothetical protein(Rv1669)	0.88	2.35E-01	0.66	3.73E-01	1.71	9.62E-03	0.75	2.22E-01	-0.47	5.52E-01	0.12	8.72E-01
Rv1671	membrane protein(Rv1671)	-0.55	1.72E-10	-1.28	1.05E-47	-1.34	3.14E-50	0.27	2.34E-02	-0.53	1.05E-05	-0.29	1.71E-02
Rv1685c	hypothetical protein(Rv1685c)	-0.96	2.93E-13	-2.72	1.20E-95	-1.67	8.62E-38	-2.46	7.80E-42	-2.51	3.02E-43	-2.64	1.22E-42
Rv1686c	ABC transporter per-mease(Rv1686c)	-1.09	1.00E-28	-2.63	3.89E-157	-1.65	3.73E-64	-2.48	125	-2.56	1.34E-131	-2.67	1.68E-126
Rv1687c	ABC transporter ATP-binding protein(Rv1687c)	-1.59	2.07E-71	-2.85	1.51E-224	-2.02	115	-2.40	1.24E-72	-2.72	1.73E-93	-2.65	1.38E-78
Rv1688	3-methyladenine DNA glyco-sylase(mpg)	-0.45	1.34E-02	-0.39	2.83E-02	-0.24	1.77E-01	-1.11	1.44E-09	-1.24	1.37E-10	-1.06	2.07E-08
Rv1704c	D-serine/alanine/glycine trans- porter protein CycA(cycA)	0.68	1.73E-09	0.69	6.68E-10	1.26	2.86E-31	0.31	1.23E-01	0.12	5.83E-01	0.82	1.42E-05
Rv1706A	hypothetical protein(Rv1706A)	2.68	6.78E-02	2.51	8.50E-02	3.36	1.38E-02	-0.12	9.17E-01	-0.19	8.80E-01	-0.21	8.48E-01
Rv1709	segregation and condensation protein ScpA(scpA)	0.88	2.33E-25	1.26	7.59E-54	1.13	1.59E-43	0.36	8.27E-04	0.34	2.52E-03	0.36	9.95E-04
Rv1710	segregation and condensation protein ScpB(scpB)	0.84	7.25E-17	1.14	3.79E-31	0.99	1.04E-23	0.15	2.75E-01	0.15	2.65E-01	0.17	1.99E-01
Rv1712	cytidylate kinase(cmk)	0.63	1.12E-15	1.08	6.26E-46	0.68	2.21E-18	0.39	4.45E-07	0.17	4.53E-02	0.51	5.26E-11
Rv1715	3-hydroxybutyryl-CoA dehy- drogenase FadB(fadB3)	0.68	2.46E-03	1.47	4.21E-13	0.80	2.36E-04	-0.23	3.74E-01	-0.18	4.97E-01	-0.14	5.97E-01
Rv1716	hypothetical protein(Rv1716)	0.16	3.58E-01	1.54	3.66E-29	0.26	9.92E-02	-0.28	9.24E-02	-0.37	3.05E-02	0.01	9.55E-01
Rv1729c	S-adenosylmethionine-de- pendent methyltransfer- ase(Rv1729c)	0.48	1.13E-03	0.32	3.33E-02	1.18	2.78E-18	0.31	3.03E-02	0.31	3.75E-02	0.01	9.36E-01
Rv1733c	transmembrane pro- tein(Rv1733c)	-2.71	1.00E-100	-1.02	7.25E-18	-3.43	145	-1.21	3.24E-09	0.62	3.82E-03	-0.64	3.60E-03
Rv1734c	hypothetical protein(Rv1734c)	-0.93	4.40E-01	1.75	4.10E-02	0.50	6.05E-01	0.04	9.56E-01	0.35	5.48E-01	-1.96	2.58E-03
Rv1735a	hypothetical protein(Rv1735a)	-2.03	5.42E-13	-1.10	3.02E-05	-2.40	7.04E-17	-0.97	1.36E-03	-0.04	9.16E-01	-1.39	8.13E-06
Rv1735c	membrane protein(Rv1735c)	-1.09	2.06E-02	0.65	9.53E-02	-0.54	2.17E-01	-0.61	1.38E-01	1.29	2.49E-04	-0.33	4.36E-01
Rv1736c	nitrate reductase-like protein NarX(narX)	-1.49	1.44E-18	-0.67	9.52E-05	-2.05	1.96E-34	-0.88	1.98E-05	0.53	1.47E-02	-0.52	1.89E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1737c	nitrate/nitrite transporter(narK2)	-1.40	2.40E-27	-0.57	1.60E-05	-2.07	5.27E-59	-0.39	8.08E-02	0.53	1.77E-02	-0.09	7.29E-01
Rv1738	hypothetical protein(Rv1738)	-3.63	2.42E-66	-1.66	6.20E-15	-5.14	3.73E-131	-0.33	2.41E-01	1.08	2.99E-05	0.26	3.87E-01
Rv1749c	integral membrane protein(Rv1749c)	-1.05	9.49E-31	-0.53	3.38E-09	-0.72	9.18E-16	0.67	1.76E-06	0.27	8.33E-02	0.67	3.63E-06
Rv1751	oxidoreductase(Rv1751)	0.37	3.87E-08	1.14	1.59E-67	0.83	1.68E-36	0.47	6.54E-08	0.72	1.63E-16	0.68	4.43E-14
Rv1752	hypothetical protein(Rv1752)	0.35	5.87E-03	1.09	9.97E-21	0.68	1.56E-08	0.72	3.01E-07	0.63	1.93E-05	0.85	1.62E-09
Rv1762c	hypothetical protein(Rv1762c)	-1.08	1.64E-23	-0.63	1.64E-09	-1.06	2.97E-23	0.09	5.11E-01	-0.16	2.35E-01	-0.10	4.31E-01
Rv1765c	hypothetical protein(Rv1765c)	-0.43	6.34E-08	-0.22	5.73E-03	-1.08	3.30E-43	0.01	9.38E-01	0.20	1.54E-01	-0.10	4.87E-01
Rv1788	PE family protein PE18(PE18)	1.11	1.58E-02	1.63	1.29E-04	1.92	3.87E-06	0.01	9.89E-01	0.41	3.06E-01	0.32	4.13E-01
Rv1792	pseudo(esxM)	-1.24	3.92E-45	-0.79	2.70E-19	-0.95	9.00E-28	-0.11	3.53E-01	-0.24	4.17E-02	0.37	1.15E-03
Rv1799	lipoprotein LppT(lppT)	-0.76	2.31E-11	-0.91	4.90E-16	-1.22	1.40E-26	0.21	3.78E-01	-0.05	8.68E-01	0.34	1.30E-01
Rv1811	Mg2+ transport P-type ATPase MgtC(mgtC)	-0.54	2.06E-09	-0.35	1.13E-04	-0.40	6.46E-06	0.72	1.08E-07	0.40	6.20E-03	1.06	1.85E-14
Rv1812c	dehydrogenase(Rv1812c)	-1.54	1.32E-91	-0.68	2.37E-19	-1.84	1.40E-130	-0.61	3.68E-06	0.58	1.67E-05	-0.97	9.89E-13
Rv1813c	hypothetical protein(Rv1813c)	-3.18	3.83E-223	-1.54	5.59E-54	-3.99	0.00E+00	-0.43	3.45E-02	0.35	9.80E-02	-0.98	9.44E-07
Rv1817	flavoprotein(Rv1817)	-0.67	1.29E-13	-0.56	5.55E-10	-1.17	4.43E-38	-0.28	1.23E-02	0.17	1.72E-01	-0.47	4.07E-05
Rv1819c	vitamin B12 transport ATP-binding protein BacA(bacA)	0.85	6.94E-15	0.87	3.46E-16	1.28	3.80E-34	0.88	9.48E-11	0.87	3.60E-10	1.14	1.11E-16
Rv1826	glycine cleavage system protein H(gcvH)	0.65	1.05E-12	0.49	7.17E-08	1.20	9.03E-42	0.04	8.03E-01	-0.06	7.30E-01	0.23	1.37E-01
Rv1839c	antitoxin VapB13(vapB13)	-1.02	5.86E-12	-0.75	2.03E-07	-0.83	9.42E-09	0.94	4.23E-06	0.13	6.24E-01	0.81	1.54E-04
Rv1849	urease subunit beta(ureB)	0.96	1.89E-08	1.09	3.95E-11	1.34	1.60E-16	0.14	5.44E-01	0.64	1.85E-03	0.75	1.18E-04
Rv1850	urease subunit alpha(ureC)	0.41	4.98E-06	1.00	1.02E-32	0.79	4.13E-20	0.29	1.05E-03	0.49	2.36E-08	0.80	1.99E-21
Rv1851	urease accessory protein UreF(ureF)	1.08	4.06E-07	1.60	1.14E-15	1.58	4.11E-15	0.13	6.75E-01	0.39	1.55E-01	0.48	6.30E-02
Rv1855c	oxidoreductase(Rv1855c)	-0.60	8.00E-28	-0.66	8.54E-34	-1.03	2.35E-79	0.27	8.86E-04	-0.08	3.92E-01	-0.01	8.98E-01
Rv1857	molybdate ABC transporter substrate-binding lipoprotein ModA(modA)	0.68	1.13E-20	0.46	2.63E-10	1.23	2.60E-67	0.74	7.02E-09	0.40	3.91E-03	1.05	2.45E-15
Rv1858	molybdenum ABC transporter permease ModB(modB)	0.97	2.22E-10	1.01	1.61E-11	1.12	5.45E-14	0.48	1.16E-02	0.57	2.89E-03	0.79	6.58E-06
Rv1870c	hypothetical protein(Rv1870c)	0.41	4.39E-09	0.63	9.40E-20	1.09	2.43E-58	0.45	3.03E-05	0.31	6.45E-03	0.34	3.18E-03
Rv1880c	cytochrome P450 Cyp140(cyp140)	-0.65	9.76E-37	-0.57	3.20E-28	-1.38	1.53E-156	-0.27	2.57E-04	-0.42	1.79E-08	-0.42	2.71E-08

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1881c	lipoprotein LppE(lppE)	-1.14	1.33E-31	-0.69	6.80E-13	-1.26	2.11E-38	0.33	4.58E-04	-0.20	5.56E-02	0.20	4.76E-02
Rv1883c	hypothetical protein(Rv1883c) resuscitation-promoting factor	-0.89	1.59E-41	-0.55	2.21E-17	-1.13	6.91E-66	0.30	4.43E-02	0.58	4.66E-05	-0.06	7.54E-01
Rv1884c	RpfC(rpfC)	-0.64	3.58E-16	-0.04	6.33E-01	-1.07	1.59E-43	0.20	2.42E-01	0.73	1.54E-06	-0.17	3.45E-01
Rv1885c	chorismate mutase(Rv1885c)	0.53	2.13E-08	-0.38	1.22E-04	1.01	1.00E-28	0.02	9.13E-01	-0.03	8.92E-01	0.56	1.08E-04
Rv1887a	pseudo(Rv1887a)	0.09	2.38E-01	-0.16	3.03E-02	0.55	1.28E-16	0.76	6.91E-19	0.51	1.95E-08	1.18	9.02E-42
Rv1891	hypothetical protein(Rv1891)	-1.24	9.87E-22	-0.68	1.97E-07	-1.29	5.02E-24	-0.14	3.08E-01	-0.23	9.11E-02	-0.24	7.90E-02
Rv1893	hypothetical protein(Rv1893)	-0.72	5.53E-15	-0.71	4.19E-15	-1.30	5.98E-45	-0.45	8.57E-03	-0.70	3.47E-05	-0.27	1.41E-01
	hypothetical protein(Rv1894c)						1.36E-						
Rv1894c		-1.01	5.05E-40	-1.45	4.21E-81	-1.81	125	-0.40	1.09E-05	-0.40	1.49E-05	-0.74	2.70E-15
Rv1903	membrane protein(Rv1903)	-0.16	4.01E-01	-0.19	2.74E-01	0.65	4.04E-05	0.59	3.18E-03	0.05	8.51E-01	1.06	1.89E-08
Rv1909c	ferric uptake regulation protein FurA(furA)	-0.01	9.27E-01	-0.61	5.46E-07	-0.41	7.37E-04	-0.98	9.05E-08	-0.98	1.32E-07	-1.52	1.76E-15
Rv1912c	oxidoreductase FadB(fadB5)	-0.41	3.10E-08	-0.63	3.56E-18	-1.04	2.27E-44	-0.21	2.56E-02	-0.25	7.20E-03	-0.38	3.00E-05
	immunogenic protein												
Rv1926c	Mpt63(mpt63)	-0.01	9.30E-01	-1.04	1.25E-22	-0.04	6.92E-01	-0.78	5.24E-10	-1.19	2.20E-20	-0.66	5.51E-07
Rv1932	2-Cys peroxiredoxin(tpx)	-0.12	5.94E-02	-0.01	8.73E-01	0.35	5.05E-09	0.85	1.01E-15	0.43	1.52E-04	1.29	1.14E-31
Rv1936	monooxygenase(Rv1936)	-1.02	1.39E-22	-1.22	1.93E-32	-1.18	5.82E-30	-0.99	2.35E-16	-0.56	7.46E-06	-0.80	3.39E-10
Rv1937	oxygenase(Rv1937)	-0.04	7.47E-01	-0.66	8.37E-12	-0.28	3.99E-03	-1.12	5.00E-26	-0.64	6.32E-09	-0.82	2.73E-13
Rv1939	oxidoreductase(Rv1939)	-0.12	4.56E-01	-0.74	8.15E-07	-0.27	6.81E-02	-1.07	4.25E-13	-0.87	1.11E-08	-1.07	4.51E-12
	riboflavin biosynthesis protein												
Rv1940	RibA(ribA1)	0.09	5.49E-01	-0.52	9.11E-05	-0.22	1.11E-01	-1.03	8.75E-20	-0.97	6.25E-17	-0.96	4.40E-16
Rv1944c	hypothetical protein(Rv1944c)	1.18	4.38E-08	0.95	1.05E-05	1.76	6.05E-18	0.55	1.28E-02	0.71	1.40E-03	0.64	3.31E-03
Rv1946c	lipoprotein(lppG)	0.38	1.46E-01	0.15	5.72E-01	1.49	3.71E-11	0.41	1.57E-01	-0.55	8.62E-02	0.10	7.74E-01
Rv1947	hypothetical protein(Rv1947)	0.69	1.16E-12	0.33	8.93E-04	1.36	5.58E-51	0.74	9.70E-14	0.43	6.99E-05	1.09	2.00E-28
Rv1950c	hypothetical protein(Rv1950c)	-0.25	5.53E-02	0.05	7.16E-01	-1.00	4.98E-16	0.51	2.61E-03	0.69	4.43E-05	0.55	1.81E-03
Rv1953	ribonuclease VapC14(vapC14)	-0.89	3.20E-13	-0.70	1.40E-08	-1.43	1.65E-32	-0.21	1.98E-01	-0.58	9.00E-05	-0.49	1.42E-03
Rv1954c	hypothetical protein(Rv1954c)	0.84	2.45E-01	1.38	3.55E-02	1.78	4.71E-03	-0.33	5.10E-01	-0.31	5.56E-01	-0.17	7.33E-01
Rv1955	toxin HigB(higB)	-0.10	3.45E-01	-0.51	4.36E-07	-1.00	1.18E-23	-1.08	1.08E-12	-1.20	3.12E-15	-1.32	1.46E-16
Rv1956	antitoxin HigA(higA)	0.49	2.00E-06	0.05	6.72E-01	-0.25	1.74E-02	-1.31	1.46E-12	-1.05	2.31E-08	-1.25	1.21E-10
Rv1957	SecB-like chaperone(Rv1957)	0.46	1.18E-04	0.37	1.71E-03	0.05	6.75E-01	-1.13	1.46E-17	-0.78	8.61E-09	-0.89	2.75E-10
Rv1960c	antitoxin ParD1(parD1)	-0.73	1.14E-08	-1.29	1.35E-23	-0.48	1.50E-04	0.30	2.89E-01	-0.33	2.46E-01	-0.14	6.55E-01
	integral membrane protein(yrbE3B)												
Rv1965		0.05	7.56E-01	-0.60	3.08E-05	0.07	6.35E-01	-1.15	9.10E-20	-0.81	3.62E-10	-0.91	6.40E-12
	Mce family protein												
Rv1966	Mce3A(mce3A)	-0.08	6.82E-01	-0.50	3.63E-03	0.03	8.75E-01	-1.17	4.01E-18	-0.62	7.35E-06	-0.79	2.38E-08

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1967	Mce family protein Mce3B(mce3B)	0.28	3.91E-02	-0.43	1.99E-03	0.02	8.79E-01	-1.25	1.29E-22	-0.81	6.87E-10	-0.94	1.99E-12
Rv1968	Mce family protein Mce3C(mce3C)	0.40	2.68E-02	-0.44	1.83E-02	0.34	5.55E-02	-1.45	3.24E-19	-0.88	1.08E-07	-0.95	2.76E-08
Rv1969	Mce family protein Mce3D(mce3D)	0.64	1.30E-02	-0.19	5.04E-01	0.56	2.68E-02	-1.58	3.81E-16	-0.67	8.50E-04	-0.90	7.08E-06
Rv1970	Mce family lipoprotein LprM(lprM)	0.53	1.00E-02	-0.23	2.98E-01	0.47	2.16E-02	-1.21	2.04E-11	-0.76	5.21E-05	-0.97	2.92E-07
Rv1971	Mce family protein Mce3F(mce3F)	0.43	7.91E-03	-0.27	1.05E-01	0.28	8.85E-02	-1.10	1.42E-24	-0.69	3.14E-10	-0.82	2.73E-13
Rv1972	Mce associated membrane protein(Rv1972)	-0.36	2.12E-02	-1.17	1.18E-13	-0.40	8.09E-03	-0.82	1.21E-04	-0.91	2.89E-05	-0.86	1.09E-04
Rv1973	Mce associated membrane protein(Rv1973)	-0.32	2.83E-01	-1.11	2.11E-04	-0.43	1.32E-01	-1.07	2.72E-04	-0.99	1.27E-03	-1.13	2.07E-04
Rv1974	membrane protein(Rv1974)	-0.19	6.58E-01	-1.05	8.55E-03	-0.31	4.22E-01	-1.02	9.03E-04	-1.16	3.93E-04	-0.91	3.51E-03
Rv1975	hypothetical protein(Rv1975)	0.55	3.31E-01	0.71	1.86E-01	1.06	3.85E-02	-0.89	3.59E-02	-0.63	1.62E-01	-0.62	1.46E-01
Rv1976c	hypothetical protein(Rv1976c)	-0.98	3.87E-17	-0.73	2.94E-10	-1.04	2.59E-19	-0.03	9.08E-01	-0.23	2.15E-01	0.03	9.10E-01
Rv1979c	permease(Rv1979c)	0.69	3.98E-16	1.05	8.64E-39	2.08	1.35E-160	0.63	1.90E-14	0.72	1.13E-17	0.81	4.52E-22
Rv1980c	immunogenic protein Mpt64(mpt64)	0.04	7.29E-01	-0.21	2.63E-02	1.23	2.31E-45	0.33	6.17E-03	0.25	5.23E-02	0.63	1.81E-07
Rv1982A	antitoxin VapB36(vapB36)	-1.32	7.94E-32	-1.43	1.23E-37	-2.08	2.68E-77	-0.09	7.14E-01	0.08	7.48E-01	-0.19	4.16E-01
Rv1985c	HTH-type transcriptional regulator(Rv1985c)	0.17	1.06E-01	0.10	3.53E-01	-0.28	5.68E-03	-0.43	4.10E-05	-0.40	1.94E-04	-1.01	4.33E-21
Rv1988	23S rRNA (adenine(2058)-N(6))-methyltransferase Erm(37)(erm(37))	-1.52	2.77E-19	-0.96	1.38E-08	-1.46	2.71E-18	0.31	2.54E-01	0.02	9.53E-01	0.02	9.40E-01
Rv1989c	hypothetical protein(Rv1989c)	-0.79	1.21E-20	-0.85	1.87E-24	-1.31	1.89E-52	-0.21	1.53E-01	-0.02	9.13E-01	-0.41	3.39E-03
Rv1990c	transcriptional regulator(Rv1990c)	-1.10	9.72E-36	-1.35	6.50E-53	-1.41	9.05E-57	-0.15	2.87E-01	-0.32	1.92E-02	-0.57	2.26E-05
Rv1991A	antitoxin MazE6(mazE6)	-1.36	6.06E-43	-1.19	3.33E-35	-2.08	5.74E-89	-0.14	3.38E-01	-0.04	8.10E-01	-0.51	1.19E-04
Rv1992c	cation transporter ATPase G(ctpG)	-0.42	3.46E-10	-0.71	5.72E-27	-1.07	1.23E-58	-0.86	3.83E-24	0.00	9.97E-01	-1.01	3.80E-30
Rv1993c	hypothetical protein(Rv1993c)	-1.22	8.81E-20	-1.12	2.70E-17	-1.80	5.26E-41	-0.67	1.07E-06	0.20	2.04E-01	-0.86	1.70E-09
Rv1994c	HTH-type transcriptional regulator CmtR(cmtR)	-1.31	4.28E-45	-1.33	8.70E-47	-1.95	4.16E-97	-0.29	7.04E-04	0.00	9.80E-01	-0.77	2.21E-19
Rv1996	universal stress protein(Rv1996)	-1.16	1.89E-29	0.30	5.01E-03	-2.22	1.18E-102	-0.10	6.67E-01	1.01	5.36E-08	-0.14	5.44E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv1997	cation transporter ATPase F(ctpF)	-1.74	1.06E-57	-0.15	2.07E-01	-2.15	1.79E-87	-0.47	6.27E-02	0.93	1.40E-04	-0.64	1.27E-02
Rv1999c	transporter(Rv1999c)	0.48	2.63E-03	1.82	1.85E-37	0.76	4.06E-07	0.09	7.11E-01	0.66	2.27E-03	0.33	1.46E-01
Rv2001	hypothetical protein(Rv2001)	0.60	5.44E-04	0.27	1.32E-01	0.64	1.64E-04	0.85	1.13E-04	0.35	1.74E-01	1.06	9.71E-07
Rv2005c	universal stress protein(Rv2005c)	-1.44	2.49E-36	-1.19	1.49E-25	-1.68	4.57E-50	0.15	5.84E-01	-0.13	6.50E-01	0.28	3.04E-01
Rv2007c	ferredoxin(fdxA)	-3.43	6.04E-99	-1.84	5.94E-30	-4.10	140	-0.43	3.14E-02	0.40	5.12E-02	-0.27	2.05E-01
Rv2015c	hypothetical protein(Rv2015c)	-0.40	1.67E-07	-0.28	2.20E-04	-1.11	5.92E-50	0.01	9.33E-01	0.15	2.68E-01	-0.16	2.59E-01
Rv2022c	hypothetical protein(Rv2022c)	-0.74	2.00E-13	-1.23	1.87E-34	-0.35	4.13E-04	0.13	5.65E-01	-0.40	5.72E-02	-0.02	9.27E-01
Rv2025c	cation efflux system protein(Rv2025c)	0.39	7.67E-03	0.96	1.53E-12	-0.78	5.80E-07	-0.61	1.73E-03	1.40	6.41E-15	-0.84	1.96E-05
Rv2028c	universal stress protein(Rv2028c)	-1.85	3.46E-26	-0.57	1.34E-03	-2.35	1.99E-41	-0.68	1.39E-03	0.17	5.02E-01	-0.40	8.55E-02
Rv2029c	6-phosphofructokinase PfkB(pfkB)	-1.59	9.36E-26	-0.30	6.06E-02	-1.95	1.65E-38	-0.81	6.74E-04	0.04	8.86E-01	-0.50	4.93E-02
Rv2030c	hypothetical protein(Rv2030c)	-2.01	7.92E-49	-1.16	3.69E-17	-2.30	7.94E-65	-0.82	1.38E-04	-0.02	9.49E-01	-0.58	1.15E-02
Rv2031c	alpha-crystallin(hspX)	-3.42	4.10E-99	-1.62	1.47E-23	-3.85	126	-1.03	2.76E-05	0.23	4.27E-01	-0.58	3.04E-02
Rv2032	NAD(P)H nitroreductase(acg)	-2.04	1.56E-52	-0.61	5.74E-06	-2.69	6.24E-89	-1.17	3.87E-08	1.05	8.95E-07	-0.77	7.52E-04
Rv2034	ArsR family HTH-type transcriptional repressor(Rv2034)	0.44	3.33E-02	-0.62	4.21E-03	0.01	9.82E-01	-0.62	8.90E-04	-0.60	1.89E-03	-1.08	1.72E-08
Rv2035	hypothetical protein(Rv2035)	-0.58	4.29E-09	-1.16	2.21E-31	-0.94	2.05E-21	-0.83	1.02E-11	-0.80	1.59E-10	-1.03	6.69E-16
Rv2036	hypothetical protein(Rv2036)	-0.43	1.26E-04	-0.97	4.35E-18	-0.50	5.31E-06	-0.94	8.56E-15	-1.02	4.37E-16	-1.11	3.11E-18
Rv2044c	hypothetical protein(Rv2044c)	0.47	3.56E-01	1.33	1.86E-03	1.26	3.64E-03	0.26	6.07E-01	0.73	9.69E-02	0.28	5.58E-01
Rv2046	lipoprotein Lppl(lppl)	1.11	3.31E-04	1.24	3.34E-05	2.34	5.92E-17	0.59	9.48E-02	0.74	3.64E-02	1.07	7.77E-04
Rv2048c	polyketide synthase(pks12)	-0.24	2.41E-03	0.47	9.24E-10	-0.12	1.38E-01	0.22	1.29E-01	0.61	5.87E-06	1.22	8.13E-19
Rv2054	hypothetical protein(Rv2054)	0.22	1.58E-02	0.64	5.28E-14	0.39	6.92E-06	0.38	6.57E-03	0.26	8.07E-02	1.23	1.85E-20
Rv2055c	30S ribosomal protein S18(rpsR2)	0.57	1.10E-01	0.83	1.24E-02	1.82	2.65E-09	0.29	4.84E-01	-0.01	9.86E-01	0.47	2.22E-01
Rv2056c	30S ribosomal protein S14(rpsN2)	1.05	3.20E-02	0.93	5.44E-02	1.80	6.04E-05	-0.07	9.22E-01	0.35	5.77E-01	0.27	6.48E-01
Rv2057c	50S ribosomal protein L33(rpmG1)	-0.12	8.74E-01	0.39	5.41E-01	1.21	3.44E-02	0.81	3.03E-01	0.71	4.01E-01	1.41	4.48E-02
Rv2058c	50S ribosomal protein L28(rpmB2)	2.47	7.10E-04	2.23	2.24E-03	3.56	2.43E-07	-0.46	5.67E-01	0.79	2.57E-01	-0.06	9.41E-01
Rv2059	hypothetical protein(Rv2059)	0.63	1.20E-03	1.02	2.02E-08	1.08	3.21E-09	0.18	3.32E-01	0.58	7.48E-04	0.07	7.31E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2063	antitoxin MazE7(mazE7) mRNA interferase	0.59	1.06E-01	0.21	5.89E-01	-0.01	9.90E-01	-1.21	6.17E-03	-0.37	4.54E-01	-0.95	3.47E-02
Rv2063A	MazF7(mazF7)	0.59	2.52E-03	0.26	1.97E-01	0.11	6.18E-01	-0.62	6.47E-03	-0.08	7.88E-01	-1.01	1.28E-05
Rv2067c	hypothetical protein(Rv2067c)	0.19	5.24E-02	0.66	3.54E-13	1.16	1.21E-38	0.65	2.35E-10	0.35	1.54E-03	0.84	6.30E-16
Rv2068c	beta-lactamase(blaC)	0.13	2.36E-01	0.62	7.08E-11	0.82	3.06E-18	0.64	2.98E-07	0.58	7.27E-06	1.03	2.61E-17
Rv2069	ECF RNA polymerase sigma factor SigC(sigC)	0.26	8.36E-02	0.79	9.37E-09	1.29	2.47E-22	0.98	1.39E-07	1.28	4.49E-12	1.49	1.13E-16
Rv2094c	Sec-independent protein translocase membrane-bound protein TatA(tatA)	-1.34	3.47E-37	-0.64	1.48E-09	-1.50	1.18E-46	0.40	2.69E-02	0.31	9.81E-02	0.55	2.61E-03
Rv2102	hypothetical protein(Rv2102)	0.09	5.15E-01	0.59	1.81E-06	0.16	2.33E-01	0.70	3.04E-05	1.05	2.93E-10	1.19	2.18E-13
Rv2107	PE family protein PE22(PE22)	0.02	9.21E-01	-0.19	2.32E-01	-1.11	8.62E-13	-0.28	1.86E-01	-0.38	7.84E-02	-0.37	7.79E-02
Rv2108	PPE family protein PPE36(PPE36)	-0.75	1.72E-14	-0.70	5.55E-13	-1.41	9.59E-46	0.21	1.71E-01	-0.02	9.33E-01	0.32	3.39E-02
Rv2111c	ubiquitin-like protein Pup(pup)	-1.35	5.80E-27	-0.90	7.06E-13	-1.19	1.25E-21	0.07	8.00E-01	0.25	3.00E-01	-0.38	1.12E-01
Rv2113	integral membrane protein(Rv2113)	0.99	8.81E-12	1.14	6.99E-16	1.94	1.21E-45	0.44	1.89E-02	0.49	1.05E-02	0.92	1.77E-07
Rv2114	hypothetical protein(Rv2114)	0.89	3.60E-07	1.00	4.39E-09	2.12	8.79E-41	1.24	5.07E-08	0.99	4.13E-05	1.62	4.34E-13
Rv2117	hypothetical protein(Rv2117)	1.04	2.91E-02	1.24	6.75E-03	0.91	5.63E-02	-0.04	9.40E-01	0.84	4.80E-02	0.37	4.05E-01
Rv2123	PPE family protein PPE37(PPE37)	0.86	2.33E-03	1.26	4.02E-06	1.33	9.73E-07	0.03	9.23E-01	0.75	6.53E-03	-0.02	9.53E-01
Rv2142A	antitoxin ParD2(parD2)	-1.03	1.16E-14	-0.92	3.08E-12	-1.08	3.51E-16	0.37	6.73E-02	-0.14	5.38E-01	0.10	6.57E-01
Rv2142c	toxin ParE2(parE2)	-0.05	8.13E-01	-0.53	5.05E-03	-0.14	4.79E-01	-0.08	7.31E-01	-0.46	2.70E-02	-1.04	2.04E-07
Rv2153c	(murG)	1.32	3.34E-33	0.73	5.22E-11	0.70	3.11E-10	-0.54	2.01E-05	0.16	2.79E-01	-0.86	3.64E-11
Rv2158c	UDP-N-acetylmuramoylalanine-D-glutamate--2,6-diaminopimelate ligase(murE)	0.50	2.01E-05	-0.46	8.48E-05	0.38	1.17E-03	-1.34	2.57E-32	-0.76	4.74E-11	-0.94	5.21E-15
Rv2159c	hypothetical protein(Rv2159c)	0.31	1.21E-02	-0.22	8.20E-02	0.09	4.87E-01	-1.59	1.00E-28	-0.88	1.63E-09	-0.88	8.23E-09
Rv2161c	hypothetical protein(Rv2161c)	-0.24	2.31E-02	-0.61	1.25E-09	-0.33	1.09E-03	-1.03	1.42E-23	-0.38	4.73E-04	-0.56	4.34E-07
Rv2166c	transcriptional regulator MraZ(Rv2166c)	-0.75	1.86E-17	-0.89	3.43E-24	-1.04	1.03E-32	-0.18	2.09E-01	0.11	4.83E-01	-0.78	3.22E-09
Rv2169c	transmembrane protein(Rv2169c)	-1.03	5.58E-36	-0.88	4.44E-27	-1.14	1.36E-44	-0.13	1.72E-01	-0.45	2.80E-07	-0.87	5.54E-22
Rv2170	GCN5-like N-acetyltransferase(Rv2170)	0.20	4.50E-01	0.65	5.70E-03	0.57	1.75E-02	-0.50	9.26E-02	-0.41	1.97E-01	-1.05	4.28E-04
Rv2179c	3'-5' exoribonuclease(Rv2179c)	0.78	3.53E-04	0.43	5.86E-02	1.12	7.22E-08	-0.13	6.54E-01	0.05	8.80E-01	-0.04	8.95E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2185c	hypothetical protein(TB16.3)	-1.15	7.71E-24	-1.53	2.06E-41	-1.23	1.70E-27	0.04	8.44E-01	-0.28	8.99E-02	-0.07	7.10E-01
Rv2186c	hypothetical protein(Rv2186c)	-1.34	7.57E-46	-0.94	5.02E-25	-1.44	5.66E-53	0.03	8.54E-01	-0.38	7.96E-03	-0.01	9.53E-01
Rv2187	long-chain-fatty-acid--CoA ligase FadD15(fadD15)	-0.53	2.50E-31	-0.81	8.41E-74	-1.05	119	-0.58	6.87E-12	-0.69	6.54E-16	-0.80	1.35E-19
Rv2189c	hypothetical protein(Rv2189c)	0.81	2.69E-16	0.77	3.01E-15	1.85	1.94E-86	1.03	7.94E-18	0.94	1.74E-14	1.45	4.03E-33
Rv2190c	endopeptidase(Rv2190c)	0.61	5.11E-38	0.49	2.56E-25	1.41	203	0.85	1.24E-20	0.68	3.53E-13	1.07	1.00E-28
Rv2201	asparagine synthetase(asnB)	0.89	4.93E-17	0.53	9.82E-07	1.34	3.49E-39	-0.32	1.22E-02	-0.25	6.25E-02	-0.07	6.22E-01
Rv2206	transmembrane protein(Rv2206)	0.74	3.08E-12	0.52	9.32E-07	1.02	1.49E-22	-0.36	1.46E-02	-0.13	4.43E-01	-0.23	1.43E-01
Rv2211c	aminomethyltransferase(gcvT)	0.48	1.36E-09	0.58	1.46E-13	1.14	3.16E-50	0.79	2.82E-12	1.01	5.73E-19	1.27	3.00E-28
Rv2213	cytosol aminopeptidase(pepB)	0.40	2.62E-10	0.38	6.48E-10	0.71	5.53E-32	0.71	1.14E-22	0.72	1.79E-22	1.02	7.72E-44
Rv2220	glutamine synthetase(glnA1)	0.28	2.96E-03	0.00	9.94E-01	0.53	4.37E-09	0.28	1.22E-02	-0.02	8.74E-01	1.08	9.45E-23
Rv2237A	hypothetical protein(Rv2237A)	-1.95	3.25E-32	-1.41	1.32E-17	-1.55	4.06E-21	0.66	2.59E-02	0.18	6.00E-01	0.58	6.23E-02
Rv2241	pyruvate dehydrogenase E1 component(aceE)	-0.64	1.62E-52	-0.15	4.46E-04	-1.05	139	-0.14	7.69E-04	-0.09	4.59E-02	-0.03	5.41E-01
Rv2243	malonyl CoA-acyl carrier protein transacylase(fabD)	0.28	1.35E-02	-0.22	5.55E-02	-0.06	6.41E-01	-0.86	1.29E-13	-0.10	4.75E-01	-1.03	1.54E-17
Rv2249c	glycerol-3-phosphate dehydrogenase(glpD1)	0.51	2.36E-04	0.07	6.27E-01	-0.18	2.15E-01	-0.57	5.60E-06	-0.40	2.35E-03	-1.01	5.71E-15
Rv2256a	hypothetical protein(Rv2256a)	-0.85	4.25E-13	-0.93	1.91E-15	-1.43	3.41E-34	0.39	4.74E-02	0.06	7.95E-01	-0.33	1.06E-01
Rv2256c	hypothetical protein(Rv2256c)	-0.70	4.11E-25	-0.68	7.18E-24	-1.46	105	-0.08	4.89E-01	-0.09	4.50E-01	-0.39	2.63E-04
Rv2257c	hypothetical protein(Rv2257c)	0.71	7.01E-13	1.41	1.20E-52	1.54	3.39E-63	1.28	4.01E-14	1.26	5.30E-13	2.14	5.64E-38
Rv2258c	transcriptional regulator(Rv2258c)	0.43	5.45E-10	1.06	2.23E-57	1.28	5.27E-84	1.55	3.41E-31	1.41	1.45E-25	2.30	9.18E-63
Rv2265	integral membrane protein(Rv2265)	0.65	4.84E-03	1.20	8.21E-09	1.36	5.45E-11	0.27	4.22E-01	0.22	5.51E-01	0.79	6.06E-03
Rv2270	lipoprotein LppN(lppN)	1.18	1.67E-04	0.40	2.36E-01	1.02	1.04E-03	0.05	8.46E-01	-0.16	5.11E-01	-0.25	2.43E-01
Rv2271	hypothetical protein(Rv2271)	0.51	1.92E-05	0.27	2.49E-02	1.44	2.74E-39	1.32	2.20E-16	0.93	3.52E-08	1.92	7.08E-32
Rv2272	transmembrane protein(Rv2272)	0.91	4.63E-05	1.00	4.61E-06	1.67	1.79E-15	0.82	2.03E-03	0.75	6.66E-03	1.50	2.61E-09
Rv2273	transmembrane protein(Rv2273)	0.30	3.05E-01	0.88	5.34E-04	1.25	3.61E-07	0.34	3.73E-01	0.57	1.20E-01	0.73	3.12E-02
Rv2275	cyclo(L-tyrosyl-L-tyrosyl) synthase(Rv2275)	0.91	1.35E-09	1.20	9.38E-17	2.28	5.13E-62	1.41	3.31E-15	1.21	7.92E-11	1.81	3.06E-24
Rv2276	cytochrome P450 Cyp121(cyp121)	0.95	5.38E-10	1.35	3.00E-20	2.13	6.85E-52	1.94	4.36E-25	1.80	6.95E-21	2.69	7.44E-48

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2282c	LysR family HTH-type transcriptional regulator(Rv2282c)	0.89	1.10E-02	1.25	1.30E-04	1.49	3.34E-06	0.12	7.92E-01	0.50	2.00E-01	0.66	6.04E-02
Rv2288	hypothetical protein(Rv2288)	-0.05	8.42E-01	0.39	3.93E-02	1.13	3.52E-11	0.88	1.60E-04	0.54	3.67E-02	1.16	3.06E-07
Rv2289	CDP-diacylglycerol pyrophosphatase(cdh)	0.32	7.78E-03	0.49	1.55E-05	1.45	2.76E-42	1.36	5.15E-16	0.91	3.34E-07	1.67	1.32E-22
Rv2294	cystathionine beta-lyase(Rv2294)	0.65	2.41E-07	0.74	1.07E-09	1.34	1.41E-30	1.00	1.07E-11	0.77	9.35E-07	1.63	5.37E-30
Rv2301	cutinase(cut2)	0.76	2.29E-09	0.18	1.99E-01	1.03	1.59E-16	-0.36	6.93E-02	-0.35	8.67E-02	0.28	1.76E-01
Rv2302	hypothetical protein(Rv2302)	-0.92	1.39E-40	-1.30	1.51E-79	-1.35	4.55E-85	-0.33	2.59E-03	-0.71	3.41E-11	-0.02	8.80E-01
Rv2304c	hypothetical protein(Rv2304c)	-1.31	2.00E-15	-0.84	1.49E-07	-1.22	5.13E-14	0.03	9.13E-01	-0.27	2.29E-01	-0.25	2.59E-01
Rv2307A	hypothetical protein(Rv2307A)	-0.31	1.84E-01	-0.34	1.37E-01	-0.05	8.48E-01	-0.52	2.94E-02	-1.03	3.71E-05	-0.74	2.16E-03
Rv2307B	hypothetical protein(Rv2307B)	-1.08	4.30E-12	-1.07	1.48E-12	-1.26	8.63E-16	0.51	2.13E-02	-0.29	2.68E-01	-0.09	7.34E-01
Rv2311	hypothetical protein(Rv2311)	-0.55	1.08E-01	-0.33	3.16E-01	-0.20	5.44E-01	1.12	2.20E-02	1.14	2.42E-02	1.17	1.35E-02
Rv2317	sugar ABC transporter permease UspB(uspB)	1.05	1.14E-01	1.76	3.72E-03	1.18	6.45E-02	0.67	3.88E-01	-0.41	6.79E-01	0.38	6.35E-01
Rv2329c	nitrate/nitrite transporter(narK1)	0.97	2.00E-28	1.05	1.96E-33	1.75	5.11E-93	1.40	1.60E-39	1.08	6.29E-23	1.90	1.17E-65
Rv2331A	hypothetical protein(Rv2331A)	1.06	4.54E-06	0.95	3.17E-05	1.45	4.59E-11	0.83	2.77E-04	0.54	3.23E-02	0.76	9.38E-04
Rv2335	serine acetyltransferase(cysE)	0.76	2.83E-12	0.76	1.80E-12	1.01	1.76E-21	-0.08	6.00E-01	0.19	1.73E-01	0.20	1.37E-01
Rv2337c	hypothetical protein(Rv2337c)	-0.69	1.01E-22	-0.98	5.96E-44	-1.09	6.57E-53	-0.79	9.46E-20	-0.81	4.66E-20	-1.32	1.49E-46
Rv2338c	molybdopterin biosynthesis protein MoeW(moeW)	-1.30	1.64E-69	-1.50	4.71E-93	-1.53	3.23E-95	-0.73	1.34E-14	-0.99	1.81E-25	-1.38	6.14E-44
Rv2340c	PE-PGRS family protein PE_PGRS39(PE_PGRS39)	-0.80	3.30E-17	-0.84	7.16E-19	-1.06	8.62E-30	-0.58	2.63E-08	-0.31	4.09E-03	-0.53	9.24E-07
Rv2347c	ESAT-6 like protein EsxP(esxP)	-0.90	1.50E-20	-1.01	1.25E-25	-0.89	1.27E-20	0.24	9.92E-02	-0.33	2.16E-02	0.86	5.04E-10
Rv2348c	hypothetical protein(Rv2348c)	-1.02	2.71E-26	-1.53	1.76E-57	-1.09	1.77E-30	-0.27	9.48E-02	-0.81	3.40E-08	-0.32	4.59E-02
Rv2351c	membrane-associated phospholipase A(plcA)	-0.54	1.72E-05	-0.47	1.17E-04	-1.25	1.08E-22	0.13	6.14E-01	0.31	1.95E-01	0.15	5.60E-01
Rv2352c	PPE family protein PPE38(PPE38)	-1.56	1.41E-09	-1.00	1.19E-04	-1.23	1.55E-06	-0.63	4.42E-02	-0.32	3.42E-01	0.09	8.08E-01
Rv2359	zinc uptake regulation protein(zur)	-0.72	7.46E-26	-0.76	3.79E-29	-1.37	5.28E-87	-0.25	1.02E-01	-0.63	1.46E-05	-0.85	1.05E-08
Rv2365c	hypothetical protein(Rv2365c)	1.27	3.95E-15	1.29	9.12E-16	1.07	4.65E-11	-0.53	2.19E-05	-0.22	1.05E-01	-0.82	1.82E-10
Rv2368c	phosphate starvation-inducible protein PhoH(phoH1)	0.01	8.55E-01	-0.04	5.41E-01	-0.19	4.81E-03	-0.42	3.87E-08	-0.39	7.30E-07	-1.15	6.66E-48
Rv2377c	hypothetical protein(Rv2377c)	-1.22	3.81E-19	-1.20	4.12E-19	-1.17	2.99E-18	-0.30	2.52E-01	0.48	5.56E-02	0.36	1.55E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2383c	phenyloxazoline synthase(mbtB)	1.11	3.35E-41	1.13	4.81E-43	1.29	1.63E-55	-0.55	8.34E-03	0.25	2.79E-01	-0.45	3.93E-02
Rv2384	2,3-dihydroxybenzoate-AMP ligase(mbtA)	0.46	3.82E-03	0.70	3.59E-06	1.24	1.17E-17	0.19	4.44E-01	0.58	9.84E-03	0.87	4.79E-05
Rv2386a	hypothetical protein(Rv2386a)	-1.03	3.80E-20	-1.07	1.84E-22	-0.15	1.59E-01	0.01	9.63E-01	-0.52	9.20E-03	0.25	2.25E-01
Rv2386c	salicylate synthase(mbtI)	1.01	1.63E-23	1.27	2.57E-37	1.63	9.04E-62	0.34	6.03E-02	0.22	2.71E-01	0.31	1.02E-01
Rv2390c	hypothetical protein(Rv2390c)	-0.06	7.28E-01	-0.11	4.71E-01	-0.08	5.77E-01	1.05	1.29E-11	0.53	1.68E-03	1.32	9.28E-17
Rv2401	hypothetical protein(Rv2401)	-1.18	9.63E-03	-0.88	3.72E-02	-0.32	4.44E-01	0.37	6.00E-01	0.29	7.02E-01	1.15	4.89E-02
Rv2406c	hypothetical protein(Rv2406c)	-1.18	7.45E-31	-0.40	9.57E-05	-0.82	2.39E-16	-0.17	3.08E-01	-0.01	9.42E-01	-0.08	6.33E-01
Rv2420c	hypothetical protein(Rv2420c)	0.42	1.63E-05	0.44	4.51E-06	1.03	3.05E-29	0.57	2.98E-08	0.51	1.94E-06	0.91	2.69E-19
Rv2421c	nicotinate-nucleotide adenyltransferase(nadD)	0.70	6.37E-10	0.61	5.95E-08	1.31	1.24E-34	0.00	9.79E-01	0.24	7.81E-02	0.15	2.64E-01
Rv2427A	pseudo(oxyR')	0.15	6.40E-01	0.91	6.16E-04	1.18	5.34E-06	0.59	3.10E-02	0.07	8.55E-01	0.40	1.65E-01
Rv2435c	cyclase(Rv2435c)	0.23	3.81E-02	1.20	1.18E-32	0.82	1.26E-15	0.92	2.20E-16	0.61	3.34E-07	1.06	7.37E-21
Rv2436	ribokinase RbsK(rbsK)	0.37	6.98E-03	1.19	6.28E-21	-0.05	7.56E-01	0.35	5.02E-03	0.47	1.42E-04	-0.56	1.36E-05
Rv2437	transmembrane protein(Rv2437)	0.49	3.02E-02	1.10	7.41E-08	0.06	7.94E-01	0.26	3.78E-01	0.41	1.52E-01	-0.50	8.30E-02
Rv2440c	GTPase Obg(obg)	-0.65	4.74E-13	-0.47	1.81E-07	-0.17	6.69E-02	0.64	1.14E-07	0.48	1.18E-04	1.28	1.52E-25
Rv2441c	50S ribosomal protein L27(rpmA)	-0.27	4.54E-04	-0.38	3.87E-07	0.13	8.79E-02	0.53	5.41E-05	0.56	2.25E-05	1.28	4.43E-22
Rv2442c	50S ribosomal protein L21(rplU)	-0.75	1.82E-18	-0.73	1.77E-17	-0.04	6.51E-01	0.66	1.19E-05	0.44	4.55E-03	1.01	4.25E-11
Rv2450c	resuscitation-promoting factor RpfE(rpfE)	1.62	7.27E-30	1.17	6.65E-16	2.85	1.28E-97	0.72	1.70E-04	1.30	3.22E-12	1.53	2.29E-16
Rv2466c	hypothetical protein(Rv2466c)	-0.73	3.62E-06	-1.56	3.58E-24	-0.96	4.63E-10	-0.52	1.12E-04	-0.59	1.24E-05	-0.68	9.35E-07
Rv2470	hemoglobin GlbO(glbO)	-0.92	2.67E-19	-0.64	3.15E-10	-1.14	3.00E-29	0.13	5.06E-01	0.01	9.76E-01	-0.62	3.38E-04
Rv2473	hypothetical protein(Rv2473)	0.79	5.02E-07	0.95	4.54E-10	1.18	4.37E-15	-0.13	4.89E-01	0.01	9.49E-01	0.04	8.35E-01
Rv2478c	hypothetical protein(Rv2478c)	0.03	8.57E-01	0.55	1.80E-04	1.16	8.22E-17	0.87	3.93E-06	0.65	1.14E-03	1.60	8.30E-18
Rv2503c	succinyl-CoA:3-ketoacid-CoA transferase subunit B(scoB)	-0.55	4.78E-12	-0.51	5.40E-11	-1.08	6.78E-42	-0.56	1.72E-06	-0.18	1.72E-01	-0.54	9.85E-06
Rv2509	short-chain type dehydrogenase/reductase(Rv2509)	0.70	1.99E-15	1.08	2.67E-36	0.87	7.54E-24	0.28	2.50E-02	0.37	2.54E-03	0.38	2.01E-03
Rv2510c	hypothetical protein(Rv2510c)	0.53	3.20E-05	1.27	4.88E-28	1.74	1.21E-53	0.82	2.78E-06	1.04	4.10E-09	1.17	1.34E-11
Rv2516c	hypothetical protein(Rv2516c)	-1.09	2.17E-29	-0.66	5.91E-12	-0.97	3.71E-24	-0.85	2.32E-22	-0.71	1.23E-15	-1.04	3.30E-29
Rv2517c	hypothetical protein(Rv2517c)	-1.23	3.52E-14	-0.51	1.43E-03	-0.88	2.82E-08	-0.63	1.19E-05	-0.58	9.53E-05	-1.20	6.04E-16
Rv2522c	hypothetical protein(Rv2522c)	0.75	7.72E-13	0.96	7.72E-21	1.15	4.92E-30	0.61	2.13E-06	0.72	3.42E-08	0.75	1.23E-08
Rv2525c	hypothetical protein(Rv2525c)	-0.41	2.77E-04	0.00	9.87E-01	-0.60	5.56E-08	-0.12	4.45E-01	0.12	4.31E-01	-1.08	1.91E-15

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2526	antitoxin VapB17(vapB17)	-0.73	4.12E-08	-0.54	3.61E-05	-1.36	1.20E-23	-0.58	3.05E-03	-0.10	6.74E-01	-0.80	6.79E-05
Rv2527	ribonuclease VapC17(vapC17)	-0.12	3.88E-01	-0.30	2.30E-02	-1.02	7.63E-15	-0.35	5.54E-02	-0.34	6.60E-02	-0.80	7.46E-06
Rv2528c	restriction system protein(mrr)	0.96	7.35E-03	0.88	1.20E-02	1.03	2.82E-03	0.59	1.23E-01	0.57	1.57E-01	1.34	6.96E-05
Rv2541	hypothetical protein(Rv2541)	0.89	1.93E-03	0.63	2.86E-02	1.66	5.91E-10	-1.03	1.24E-02	0.52	2.11E-01	0.27	5.33E-01
Rv2543	lipoprotein LppA(lppA)	0.97	3.91E-03	0.48	1.79E-01	1.29	5.02E-05	-0.49	1.79E-01	-0.23	5.74E-01	-0.39	2.87E-01
Rv2544	lipoprotein LppB(lppB)	1.33	1.39E-03	0.92	2.97E-02	1.38	6.79E-04	0.61	1.61E-01	0.81	6.42E-02	0.77	6.30E-02
Rv2548	ribonuclease VapC19(vapC19)	0.57	1.56E-02	1.14	1.08E-07	0.64	4.46E-03	-0.64	7.66E-03	-0.44	8.13E-02	-0.79	1.00E-03
Rv2550c	antitoxin VapB20(vapB20)	-0.86	2.99E-11	-0.98	2.55E-14	-0.85	3.49E-11	1.10	3.21E-07	0.57	1.54E-02	1.26	1.09E-08
	hypothetical protein(Rv2557)						5.43E-						
Rv2557		-1.36	3.39E-68	-1.72	2.36E-108	-1.80	118	-0.67	9.46E-08	-1.04	6.71E-17	-0.50	1.75E-04
Rv2558	hypothetical protein(Rv2558)	-0.90	7.89E-30	-1.57	2.90E-89	-1.43	2.09E-74	-0.73	7.10E-06	-1.03	9.22E-11	-0.75	9.91E-06
Rv2563	glutamine ABC transporter permease(Rv2563)	1.24	5.41E-23	1.13	1.89E-19	2.11	2.32E-68	0.91	1.40E-07	0.87	1.02E-06	1.35	7.53E-15
Rv2564	glutamine ABC transporter ATP-binding protein(glnQ)	1.12	6.11E-16	1.44	8.44E-27	2.15	4.91E-61	1.31	8.29E-16	0.91	1.64E-07	2.14	6.69E-41
Rv2578c	hypothetical protein(Rv2578c)	-0.60	2.17E-04	0.06	7.21E-01	-1.15	2.99E-12	-0.80	2.22E-04	-0.41	7.73E-02	-1.01	5.62E-06
Rv2582	peptidyl-prolyl cis-trans isomerase B(ppiB)	0.58	9.94E-04	0.49	4.71E-03	1.52	4.22E-21	1.22	2.20E-10	0.95	2.77E-06	1.69	2.75E-18
Rv2591	PE-PGRS family protein PE_PGRS44(PE_PGRS44)	0.18	7.70E-02	0.04	7.26E-01	0.37	1.24E-04	0.81	7.04E-12	0.45	3.41E-04	1.05	8.53E-18
Rv2594c	crossover junction endodeoxy-ribonuclease RuvC(ruvC)	-0.69	1.44E-10	-0.56	2.14E-07	-1.30	3.16E-34	0.58	4.82E-04	0.67	5.31E-05	-0.06	7.76E-01
Rv2606c	pyridoxine biosynthesis protein(snzP)	-0.99	3.24E-55	-0.77	2.76E-34	-1.30	7.87E-95	0.20	3.06E-01	0.14	4.97E-01	0.06	7.86E-01
Rv2623	universal stress protein(TB31.7)	-1.66	6.31E-34	-0.49	4.46E-04	-2.23	1.82E-61	-0.08	7.92E-01	0.71	5.54E-03	-0.26	3.58E-01
Rv2624c	universal stress protein(Rv2624c)	-2.77	1.61E-58	-1.07	3.93E-10	-3.30	4.33E-82	-0.83	2.71E-03	0.56	5.20E-02	-0.96	7.50E-04
Rv2625c	zinc metalloprotease Rip3(Rv2625c)	-2.33	7.22E-37	-1.09	4.61E-09	-2.89	1.61E-56	-0.61	5.37E-02	0.61	5.65E-02	-0.82	1.05E-02
Rv2626c	hypoxic response protein(hrp1)	-2.75	1.86E-92	-0.91	1.02E-11	-3.25	127	-0.70	9.66E-03	0.95	3.37E-04	-0.65	2.19E-02
Rv2627c	hypothetical protein(Rv2627c)	-1.08	4.79E-19	0.60	6.76E-07	-2.11	5.21E-69	-0.03	9.43E-01	1.47	8.57E-08	-0.17	6.19E-01
Rv2628	hypothetical protein(Rv2628)	-2.64	2.53E-85	-0.51	1.67E-04	-3.29	1.34E-127	-0.51	1.60E-01	1.44	1.71E-05	-0.61	1.03E-01
Rv2629	hypothetical protein(Rv2629)	-1.67	2.02E-81	-0.53	1.67E-09	-2.13	9.06E-133	-0.32	6.30E-02	1.14	6.32E-13	-0.73	1.53E-05
Rv2630	hypothetical protein(Rv2630)	-1.17	1.19E-42	-0.11	2.17E-01	-1.81	1.05E-98	-0.28	1.67E-01	0.82	7.93E-06	-0.59	2.45E-03

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2631	RNA-splicing ligase RtcB(Rv2631)	-1.14	1.90E-33	0.29	3.00E-03	-1.49	3.72E-56	-0.19	3.07E-01	0.91	2.07E-08	-0.46	8.68E-03
Rv2632c	hypothetical protein(Rv2632c)	-1.56	1.39E-68	-1.68	3.88E-80	-1.83	1.89E-93	-0.01	9.69E-01	-0.77	5.12E-05	-0.10	6.43E-01
Rv2633c	hypothetical protein(Rv2633c)	-0.74	1.65E-16	-1.18	2.45E-39	-0.83	8.92E-21	-0.36	9.07E-03	-1.11	4.67E-16	-0.23	1.13E-01
Rv2634c	PE-PGRS family protein PE_PGRS46(PE_PGRS46)	-1.00	1.34E-05	-0.62	7.81E-03	-1.17	1.94E-07	-0.74	1.89E-02	0.07	8.57E-01	-0.28	4.24E-01
Rv2639c	integral membrane protein(Rv2639c)	-1.33	1.56E-19	-0.95	6.15E-11	-1.08	1.13E-13	0.30	2.51E-01	0.15	6.08E-01	0.22	4.34E-01
Rv2641	cadmium inducible protein CadI(cadI)	-2.44	1.77E-42	-2.46	1.30E-43	-3.73	1.84E-97	-0.43	2.85E-05	-1.06	2.12E-25	-1.54	5.78E-48
Rv2642	ArsR family transcriptional regulator(Rv2642)	-0.67	1.88E-04	-1.03	4.49E-09	-1.48	6.36E-17	-1.40	3.13E-12	-0.98	2.19E-06	-1.76	6.81E-17
Rv2643	arsenic-transport integral membrane protein ArsC(arsC)	-1.10	9.95E-24	-1.27	1.20E-31	-1.85	1.76E-63	-0.89	1.11E-12	-0.72	1.99E-08	-1.30	2.31E-23
Rv2645	hypothetical protein(Rv2645)	1.09	3.49E-03	0.52	1.90E-01	1.80	1.89E-07	-0.62	1.06E-01	-0.17	6.97E-01	-0.55	1.53E-01
Rv2646	integrase(Rv2646)	1.62	1.45E-08	1.34	3.10E-06	1.85	2.25E-11	-0.73	3.25E-03	0.06	8.35E-01	-0.16	5.47E-01
Rv2647	hypothetical protein(Rv2647)	1.57	7.32E-04	1.07	2.53E-02	2.04	3.97E-06	0.15	7.31E-01	-0.39	3.81E-01	0.33	4.05E-01
Rv2656c	prophage protein(Rv2656c)	0.09	8.63E-01	-0.26	5.72E-01	-0.53	2.56E-01	-1.14	2.63E-03	-0.58	1.48E-01	-1.15	2.77E-03
Rv2657c	prophage protein(Rv2657c)	0.16	4.19E-01	-0.57	1.94E-03	0.33	6.00E-02	-0.80	4.00E-06	-1.21	1.65E-11	-0.93	1.95E-07
Rv2658c	prophage protein(Rv2658c)	-1.45	7.00E-28	-2.75	4.53E-90	-2.17	6.15E-59	-0.92	7.98E-08	-0.97	3.01E-08	-1.59	5.51E-19
Rv2659c	prophage integrase(Rv2659c)	-1.17	2.00E-20	-1.90	1.23E-50	-1.69	5.92E-41	-1.46	1.00E-28	-0.97	3.32E-13	-1.63	4.08E-32
Rv2660c	hypothetical protein(Rv2660c)	-1.03	3.09E-01	0.00	9.98E-01	-0.81	3.86E-01	1.91	1.09E-01	0.31	8.54E-01	2.76	1.25E-02
Rv2662	hypothetical protein(Rv2662)	0.14	6.90E-01	-0.73	2.79E-02	0.09	7.85E-01	-1.19	4.72E-08	-1.27	1.99E-08	-1.33	3.57E-09
Rv2663	hypothetical protein(Rv2663)	-1.55	8.97E-36	-1.66	1.36E-41	-1.88	2.29E-51	-0.44	5.68E-02	-0.70	2.45E-03	-0.97	2.32E-05
Rv2672	protease(Rv2672)	0.65	1.36E-10	1.01	2.49E-25	0.96	1.31E-22	0.10	5.42E-01	0.23	1.39E-01	0.45	1.76E-03
Rv2673	alpha-(1->3)-arabinofuranosyl-transferase(aftC)	0.63	1.03E-08	0.53	1.17E-06	1.22	2.12E-32	0.79	9.60E-08	0.58	2.60E-04	1.35	1.31E-20
Rv2679	enoyl-CoA hydratase EchA15(echA15)	0.89	4.12E-15	0.80	1.79E-12	1.01	9.83E-20	0.00	9.84E-01	0.44	2.49E-03	0.20	1.99E-01
Rv2687c	antibiotic ABC transporter permease(Rv2687c)	0.79	3.64E-15	0.31	2.42E-03	0.05	6.78E-01	-0.63	4.06E-04	-0.20	3.21E-01	-1.02	1.87E-08
Rv2700	hypothetical protein(Rv2700)	0.85	1.05E-10	0.41	2.51E-03	1.35	4.40E-27	-0.12	5.18E-01	-0.02	9.15E-01	-0.03	8.70E-01
Rv2707	hypothetical protein(Rv2707)	0.05	6.44E-01	-0.48	4.45E-08	-0.48	3.95E-08	-0.31	3.50E-02	-0.58	3.62E-05	-1.12	4.86E-15
Rv2709	transmembrane protein(Rv2709)	-0.79	1.47E-05	-0.24	1.99E-01	-1.07	2.12E-09	-0.16	3.16E-01	-0.14	4.21E-01	-0.47	1.60E-03
Rv2723	integral membrane protein(Rv2723)	0.54	1.39E-03	0.84	1.07E-07	1.81	1.34E-34	0.05	8.39E-01	0.17	4.62E-01	0.08	7.12E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2730	hypothetical protein(Rv2730) (dimethylallyl)adenosine tRNA methyltransferase(Rv2733c)	-2.35	9.51E-16	-1.18	3.16E-05	-1.94	8.80E-12	0.53	2.75E-01	0.25	6.52E-01	0.16	7.65E-01
Rv2733c		0.71	9.54E-19	0.78	7.42E-23	1.02	6.70E-39	-0.10	3.47E-01	0.07	5.46E-01	0.15	1.56E-01
Rv2734	hypothetical protein(Rv2734)	-0.11	3.44E-01	-0.53	2.18E-06	-0.12	3.13E-01	1.02	3.49E-11	0.50	3.75E-03	1.13	5.18E-13
Rv2738c	hypothetical protein(Rv2738c)	-0.32	4.19E-04	0.14	1.20E-01	0.07	4.57E-01	0.87	4.46E-06	0.74	1.69E-04	1.01	1.79E-07
Rv2760c	antitoxin VapB42(vapB42)	0.98	2.87E-02	0.71	1.20E-01	1.22	4.31E-03	0.34	4.31E-01	0.08	8.83E-01	0.13	7.77E-01
Rv2767c	membrane protein(Rv2767c)	0.62	1.97E-02	1.00	4.74E-05	0.20	4.91E-01	0.14	6.87E-01	0.47	1.22E-01	0.90	6.75E-04
Rv2769c	PE family protein PE27(PE27)	-1.24	1.46E-10	-0.51	6.90E-03	-0.76	5.06E-05	0.17	5.62E-01	0.39	1.55E-01	0.59	2.39E-02
Rv2780	L-alanine dehydrogenase(ald)	-0.63	8.04E-21	-0.87	5.10E-39	-1.28	1.15E-82	-1.61	6.00E-28	-0.88	6.77E-09	-1.63	1.12E-25
Rv2781c	oxidoreductase(Rv2781c)	1.10	3.00E-48	1.52	3.50E-96	1.40	2.00E-80	0.87	1.56E-17	0.98	1.45E-21	1.63	4.00E-59
Rv2782c	zinc protease(pepR)	1.14	1.04E-41	1.18	1.29E-44	1.08	3.51E-38	0.58	4.13E-08	0.88	6.42E-17	1.09	5.26E-24
Rv2783c	bifunctional guanosine pentaphosphate synthetase/polyribonucleotide nucleotidyltransferase(gpsI)	0.66	5.83E-33	0.74	3.53E-41	0.59	1.12E-26	0.82	4.53E-31	0.91	1.21E-37	1.42	9.21E-83
Rv2787	hypothetical protein(Rv2787) tRNA pseudouridine synthase B(truB)	-0.27	4.96E-02	-0.31	1.95E-02	0.06	6.58E-01	0.72	5.79E-06	0.32	7.90E-02	1.08	2.54E-12
Rv2793c		1.04	3.38E-25	0.88	8.26E-19	1.18	8.34E-33	-0.32	1.82E-02	0.27	5.41E-02	0.05	7.57E-01
Rv2797c	hypothetical protein(Rv2797c)	0.51	3.56E-04	1.01	5.30E-14	1.04	5.96E-15	0.25	1.53E-01	0.82	1.03E-07	0.77	3.56E-07
Rv2798c	hypothetical protein(Rv2798c)	-1.15	5.01E-23	-0.28	1.82E-02	-0.48	2.16E-05	0.73	6.98E-04	0.55	1.54E-02	1.09	6.34E-07
Rv2800	hydrolase(Rv2800)	1.05	1.40E-21	0.88	1.63E-15	0.89	5.17E-16	-0.27	5.63E-02	0.31	2.55E-02	-0.23	1.22E-01
Rv2802c	arginine/hypothetical protein(Rv2802c)	0.95	1.59E-21	0.64	1.80E-10	1.21	4.36E-36	0.21	1.10E-01	0.51	2.83E-05	0.43	4.53E-04
Rv2803	hypothetical protein(Rv2803)	0.68	1.78E-01	1.09	1.94E-02	1.15	1.30E-02	-1.04	1.07E-01	-0.05	9.43E-01	-0.17	7.99E-01
Rv2805	hypothetical protein(Rv2805)	-0.05	7.58E-01	0.57	3.78E-05	0.62	7.35E-06	0.68	3.78E-03	0.60	1.65E-02	1.38	4.84E-10
Rv2806	membrane protein(Rv2806)	0.22	7.03E-01	1.70	1.26E-04	1.56	5.08E-04	1.57	1.42E-02	1.64	1.21E-02	1.62	9.44E-03
Rv2809	hypothetical protein(Rv2809)	-1.39	5.24E-26	-0.99	5.54E-14	-1.33	1.89E-24	0.39	1.01E-02	-0.06	7.50E-01	0.24	1.53E-01
Rv2812	transposase(Rv2812)	1.03	8.54E-03	1.03	7.23E-03	0.48	2.44E-01	0.36	3.19E-01	0.27	5.01E-01	0.12	7.58E-01
Rv2816c	CRISPR-associated endoribonuclease Cas2(Rv2816c)	-1.69	1.76E-14	-1.89	6.60E-18	-1.94	7.84E-19	0.47	2.04E-01	-0.33	4.01E-01	-1.08	2.83E-03
Rv2824c	CRISPR-associated endoribonuclease Cas6(Rv2824c)	-0.62	7.77E-10	-0.99	8.43E-23	-0.78	7.22E-15	-0.63	2.00E-07	-0.69	3.23E-08	-1.11	1.14E-18
Rv2830c	antitoxin VapB22(vapB22)	-1.18	2.90E-27	-1.08	2.08E-23	-1.46	1.03E-40	0.06	6.71E-01	0.08	5.97E-01	-0.23	1.01E-01
Rv2833c	sn-glycerol-3-phosphate ABC transporter substrate-binding lipoprotein UgpB(ugpB)	0.60	3.38E-02	1.53	7.04E-10	1.17	5.14E-06	0.45	8.08E-02	0.52	5.03E-02	0.27	3.20E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
	sn-glycerol-3-phosphate ABC transporter permease												
Rv2835c	UgpA(ugpA)	0.66	9.78E-06	1.26	7.99E-20	0.94	3.46E-11	0.50	1.30E-03	0.57	3.83E-04	0.72	2.90E-06
Rv2865	antitoxin RelF(relF)	-0.83	4.49E-08	-0.72	9.64E-07	-1.35	3.31E-18	0.39	1.91E-02	0.09	6.67E-01	-0.20	2.85E-01
Rv2873	cell surface lipoprotein(mpt83)	1.88	1.27E-65	2.64	7.13E-131	1.54	9.67E-45	0.81	7.11E-06	2.13	2.49E-35	1.05	1.74E-08
	integral membrane C-type cytochrome biogenesis protein												
Rv2874	DipZ(dipZ)	2.09	7.42E-45	2.95	1.29E-92	2.08	4.25E-45	1.39	1.81E-11	3.01	7.88E-52	1.49	3.65E-12
	major secreted immunogenic protein Mpt70(mpt70)												
Rv2875	transmembrane protein	2.33	4.67E-53	3.48	9.42E-119	2.32	1.74E-53	1.86	2.91E-21	3.70	2.42E-83	2.06	2.06E-23
Rv2876	tein(Rv2876)	0.31	7.74E-04	1.16	8.27E-39	0.46	3.98E-07	0.79	9.34E-05	2.04	3.43E-26	0.94	6.78E-06
Rv2882c	ribosome recycling factor(frr)	-1.31	1.71E-31	-1.17	2.23E-25	-1.22	9.00E-28	0.06	7.72E-01	0.18	3.63E-01	0.02	9.38E-01
Rv2886c	resolvase(Rv2886c)	-0.83	5.89E-10	-0.33	1.76E-02	-1.49	2.22E-29	0.32	8.28E-02	0.45	1.10E-02	0.11	5.86E-01
Rv2897c	hypothetical protein(Rv2897c)	0.62	9.14E-06	0.85	1.69E-10	0.83	4.08E-10	0.63	1.02E-03	0.58	3.63E-03	1.02	7.67E-08
Rv2898c	hypothetical protein(Rv2898c)	-0.88	1.27E-15	-0.25	2.63E-02	-0.49	6.68E-06	1.00	1.42E-10	0.37	3.11E-02	1.48	2.76E-20
Rv2908c	hypothetical protein(Rv2908c)	-1.05	7.83E-20	-0.92	1.48E-15	-0.94	1.54E-16	0.86	1.14E-04	0.36	1.43E-01	1.17	3.16E-07
Rv2910c	hypothetical protein(Rv2910c)	-0.67	9.79E-05	-1.12	6.12E-11	-0.50	3.51E-03	0.25	3.52E-01	-0.12	6.96E-01	0.21	4.40E-01
	penicillin-binding protein												
Rv2911	DacB2(dacB2)	1.18	3.73E-18	1.16	4.47E-18	1.74	1.30E-40	0.18	4.17E-01	0.47	2.14E-02	0.60	2.14E-03
	TetR family HTH-type transcriptional regulator(Rv2912c)												
Rv2912c	D-amino acid aminohydro-	-1.76	3.24E-20	-1.37	4.54E-13	-1.81	1.05E-21	-1.00	2.07E-17	-0.71	3.58E-09	-1.10	2.33E-19
	lase(Rv2913c)												
Rv2913c	nitrogen regulatory protein P-II(glnB)	-1.71	6.05E-49	-1.52	1.57E-39	-2.07	1.90E-71	-1.01	2.21E-25	-1.06	3.80E-27	-1.22	8.57E-33
	ribonuclease III(rnc)												
Rv2919c		-1.00	2.15E-18	-0.35	2.97E-03	-1.18	2.92E-25	0.55	1.01E-05	0.20	1.43E-01	0.04	7.89E-01
	hypothetical protein(Rv2927c)												
Rv2925c		-0.64	4.31E-36	-0.57	1.40E-29	-1.13	5.10E-110	0.00	9.68E-01	0.00	9.65E-01	-0.29	1.36E-04
	daunorubicin ABC transporter												
Rv2927c	permease DrrB(drrB)	-0.87	2.16E-30	-0.61	1.18E-15	-1.77	3.71E-122	0.15	1.88E-01	0.06	6.48E-01	-0.48	1.57E-05
	multifunctional mycocerosic acid synthase(mas)												
Rv2937	transposase(Rv2943A)	1.10	9.45E-25	0.41	2.18E-04	0.76	1.05E-12	-0.09	6.62E-01	0.26	1.38E-01	-0.19	3.03E-01
	insertion sequence element												
Rv2940c	IS1533 transposase(Rv2944)	0.52	2.59E-30	0.98	7.26E-106	0.88	3.19E-86	0.39	1.78E-08	0.50	1.75E-13	1.10	2.42E-55
Rv2943A	lipoprotein LppX(lppX)	-1.65	3.34E-04	-1.64	1.81E-04	-0.44	2.70E-01	2.01	2.02E-04	0.77	2.45E-01	1.65	2.85E-03
Rv2944		1.08	7.50E-02	0.99	9.86E-02	1.18	4.49E-02	-0.52	2.95E-01	-0.75	1.50E-01	-0.39	4.25E-01
Rv2945c		-0.92	4.65E-32	-0.07	4.27E-01	-1.27	1.26E-59	-0.48	4.58E-04	-0.32	2.62E-02	0.57	3.80E-05

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2947c	polyketide synthase(pks15)	0.04	6.66E-01	1.46	1.72E-67	0.58	1.12E-11	1.14	4.19E-12	0.92	5.60E-08	2.64	1.26E-54
Rv2948c	p-hydroxybenzoyl--AMP ligase(fadD22)	0.81	4.52E-32	1.59	1.68E-124	1.51	8.30E-112	1.38	8.59E-44	0.95	1.11E-20	2.30	2.67E-109
Rv2949c	chorismate pyruvate-lyase(Rv2949c)	0.51	5.11E-11	1.16	8.33E-53	1.51	6.63E-90	1.80	5.22E-50	1.18	2.29E-21	2.73	9.47E-104
Rv2950c	long-chain-fatty-acid--AMP ligase FadD29(fadD29)	0.25	4.54E-06	0.72	2.18E-43	1.19	8.58E-118	1.50	2.62E-76	1.05	9.11E-37	2.23	4.84E-150
Rv2951c	phthiodiolone/phenolphthiodiolone dimycocerosates ketoreductase(Rv2951c)	0.33	7.61E-06	0.65	5.21E-20	0.85	1.68E-33	1.09	7.00E-36	0.79	3.41E-18	1.74	5.61E-84
Rv2952	phthiotriol/phenolphthiotriol dimycocerosates methyltransferase(Rv2952)	1.07	1.72E-22	1.38	5.51E-39	2.23	3.03E-105	1.66	5.00E-28	1.47	2.25E-21	2.46	1.99E-58
Rv2953	trans-acting enoyl reductase(Rv2953)	0.72	2.09E-19	1.06	1.38E-43	1.65	3.81E-108	1.52	3.84E-41	1.31	1.43E-29	2.11	1.95E-76
Rv2954c	hypothetical protein(Rv2954c)	0.07	3.32E-01	0.31	1.10E-06	0.89	4.41E-48	0.99	5.16E-30	0.71	4.76E-15	1.42	5.75E-57
Rv2955c	hypothetical protein(Rv2955c)	1.07	1.87E-10	1.34	8.16E-17	2.01	8.93E-38	0.54	9.38E-03	0.42	6.24E-02	1.15	1.59E-09
Rv2956	hypothetical protein(Rv2956)	0.02	8.54E-01	-0.51	7.09E-06	0.32	3.41E-03	0.66	1.59E-08	0.23	8.32E-02	1.00	1.25E-17
Rv2958c	PGL/p-HBAD biosynthesis glycosyltransferase(Rv2958c)	0.13	3.08E-01	0.34	2.78E-03	1.00	2.60E-21	0.68	2.12E-06	0.23	1.78E-01	1.57	2.11E-31
Rv2959c	rhamnosyl O-methyltransferase(Rv2959c)	-0.51	1.15E-06	-0.84	4.45E-16	0.05	6.78E-01	1.01	1.24E-21	0.17	1.97E-01	1.42	1.82E-39
Rv2961	transposase(Rv2961)	-1.17	2.09E-04	-1.85	9.99E-09	-1.69	1.39E-07	-0.16	7.81E-01	-1.04	4.76E-02	-0.93	6.71E-02
Rv2962c	PGL/p-HBAD biosynthesis rhamnosyltransferase(Rv2962c)	-1.77	5.90E-58	-1.66	4.87E-52	-1.87	1.66E-65	-1.00	5.51E-24	-1.17	8.13E-32	-1.30	8.05E-36
Rv2963	integral membrane protein(Rv2963)	-1.62	2.60E-75	-1.76	7.83E-90	-2.52	1.62E-179	-0.97	1.28E-19	-1.44	2.48E-42	-2.19	8.40E-86
Rv2964	formyltetrahydrofolate dehydrogenase(purU)	-0.39	1.22E-04	-0.80	6.64E-16	-1.05	2.62E-25	-0.77	1.50E-08	-1.00	5.54E-13	-1.34	5.47E-21
Rv2983	2-phospho-L-lactate guanylyltransferase(Rv2983)	1.04	1.98E-04	1.13	3.00E-05	1.37	2.16E-07	-0.79	3.18E-02	0.30	4.39E-01	-0.48	1.99E-01
Rv2986c	DNA-binding protein HU(hupB)	-0.03	7.91E-01	0.21	1.89E-02	-0.50	9.70E-09	0.60	8.66E-06	0.52	1.63E-04	1.03	1.09E-13
Rv2987c	3-isopropylmalate dehydratase small subunit(leuD)	1.19	4.19E-18	1.62	1.13E-35	2.31	8.33E-75	1.35	4.07E-16	1.41	6.56E-17	2.54	2.34E-53
Rv2988c	3-isopropylmalate dehydratase large subunit(leuC)	0.70	1.48E-14	1.22	3.22E-44	1.99	1.03E-120	1.12	1.48E-22	1.33	3.06E-31	2.05	5.64E-69

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv2989	transcriptional regulator(Rv2989)	1.01	4.08E-12	1.36	2.62E-22	1.62	1.12E-31	1.27	5.55E-15	1.22	3.08E-13	1.79	2.00E-28
Rv2992c	glutamate--tRNA ligase(gltS)	0.95	2.87E-17	1.72	7.66E-56	1.30	2.01E-32	0.93	6.64E-11	0.91	3.53E-10	1.33	4.12E-20
Rv2993c	2-hydroxyhepta-2,4-diene-1,7-dioate isomerase(Rv2993c)	0.74	1.41E-10	1.73	4.10E-57	1.26	3.46E-30	1.19	5.44E-20	1.29	9.76E-23	1.54	5.59E-33
Rv2994	MFS-type transporter(Rv2994)	0.96	5.33E-22	0.88	4.44E-19	1.11	6.30E-30	0.12	4.84E-01	0.58	1.47E-04	0.56	2.41E-04
Rv3007c	oxidoreductase(Rv3007c)	-0.93	1.31E-19	-0.72	3.02E-13	-1.30	2.40E-35	0.15	3.94E-01	-0.11	5.34E-01	0.17	3.26E-01
Rv3008	hypothetical protein(Rv3008)	-1.66	1.77E-37	-0.96	5.48E-14	-1.69	1.67E-39	0.16	2.81E-01	-0.01	9.63E-01	-0.24	1.00E-01
Rv3010c	6-phosphofructokinase(pfkA)	-0.69	2.39E-37	-0.30	3.99E-08	-1.10	1.50E-91	0.00	9.99E-01	-0.13	7.37E-02	-0.13	6.57E-02
Rv3011c	glutamyl-tRNA(GLN) amidotransferase subunit A(gatA)	0.85	2.08E-17	0.94	1.06E-21	1.24	8.38E-38	0.87	8.88E-13	1.03	6.25E-17	1.63	3.20E-43
Rv3012c	glutamyl-tRNA(GLN) amidotransferase subunit C(gatC)	1.65	5.93E-07	1.80	2.41E-08	2.54	3.09E-16	1.20	1.78E-03	1.56	4.27E-05	1.98	6.16E-08
Rv3013	hypothetical protein(Rv3013)	1.02	2.00E-08	0.81	8.17E-06	1.49	1.01E-17	0.92	4.11E-05	1.23	2.92E-08	1.39	1.05E-10
Rv3017c	ESAT-6 like protein EsxQ(esxQ)	-0.24	2.64E-01	-0.26	2.11E-01	0.12	5.57E-01	0.91	3.78E-04	0.51	7.55E-02	1.15	5.16E-06
Rv3018c	PPE family protein PPE46(PPE46)	-0.03	8.57E-01	0.66	4.11E-06	0.64	7.66E-06	0.54	3.73E-02	0.59	2.68E-02	1.11	6.10E-06
Rv3019c	ESAT-6 like protein EsxR(esxR)	-0.02	9.22E-01	0.11	5.92E-01	0.01	9.75E-01	0.41	1.12E-01	0.41	1.34E-01	1.13	7.78E-07
Rv3020c	ESAT-6 like protein EsxS(esxS)	-0.09	6.95E-01	0.55	5.08E-03	0.38	6.21E-02	0.71	3.51E-03	0.55	3.56E-02	1.79	1.81E-16
Rv3021c	pseudo(PPE47)	-0.04	8.83E-01	0.84	1.79E-04	0.84	1.86E-04	0.85	6.57E-03	0.86	7.97E-03	1.54	2.32E-07
Rv3022A	PE family protein PE29(PE29)	0.45	2.00E-01	0.09	8.13E-01	1.11	3.73E-04	0.80	9.48E-02	1.14	1.51E-02	1.45	9.83E-04
Rv3022c	pseudo(PPE48)	0.54	4.04E-01	0.54	3.83E-01	1.27	2.22E-02	1.67	4.05E-02	0.93	3.25E-01	1.47	7.22E-02
Rv3027c	GCN5-like N-acetyltransferase(Rv3027c)	0.60	4.08E-10	0.79	1.03E-17	0.97	3.55E-26	0.62	3.21E-09	0.76	7.01E-13	1.03	3.15E-23
Rv3030	S-adenosylmethionine-dependent methyltransferase(Rv3030)	0.42	3.06E-04	0.92	1.08E-16	0.57	5.40E-07	0.56	2.29E-04	0.77	3.59E-07	1.35	2.14E-20
Rv3031	1,4-alpha-glucan-branching protein(Rv3031)	1.01	3.28E-22	1.16	2.12E-29	1.11	3.10E-27	0.23	6.52E-02	0.64	2.09E-08	0.98	6.03E-19
Rv3032A	hypothetical protein(Rv3032A)	0.30	2.25E-01	0.98	6.31E-06	0.62	5.42E-03	0.99	6.99E-03	1.09	3.65E-03	2.05	2.64E-10
Rv3033	hypothetical protein(Rv3033)	0.17	4.15E-01	0.17	4.00E-01	0.68	2.08E-04	0.73	1.87E-03	0.54	3.35E-02	1.52	1.54E-12
Rv3036c	hypothetical protein(TB22.2)	1.52	8.88E-21	1.01	1.19E-09	1.97	6.93E-36	-0.14	5.93E-01	0.01	9.84E-01	-0.26	3.26E-01
Rv3037c	S-adenosylmethionine-dependent methyltransferase(Rv3037c)	0.72	1.18E-13	1.03	2.46E-27	0.78	6.60E-16	0.09	4.25E-01	0.38	2.48E-04	0.07	5.82E-01
Rv3047c	hypothetical protein(Rv3047c)	-1.09	2.94E-12	-0.78	1.77E-07	-0.98	1.33E-10	-0.19	2.62E-01	-0.15	4.36E-01	-0.38	2.09E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3051c	ribonucleoside-diphosphate reductase subunit alpha(nrdE)	-0.52	2.14E-14	-0.57	3.47E-17	-1.01	1.71E-51	-0.23	1.92E-02	-0.24	1.99E-02	-0.33	9.06E-04
Rv3052c	NrdI protein(nrdI)	0.15	3.63E-02	-0.17	1.72E-02	-0.16	2.84E-02	-0.93	1.06E-14	-0.36	4.72E-03	-1.09	4.99E-18
Rv3054c	hypothetical protein(Rv3054c)	-0.32	5.42E-02	-1.06	3.23E-11	-1.51	2.69E-20	0.27	1.75E-01	-0.37	6.97E-02	-0.75	1.30E-04
Rv3059	cytochrome P450 Cyp136(cyp136)	-0.62	3.02E-17	-0.46	1.98E-10	-1.02	3.73E-44	-0.31	5.85E-03	-0.53	1.10E-06	-0.62	2.93E-08
Rv3060c	GntR family transcriptional regulator(Rv3060c)	0.15	3.38E-02	0.00	9.83E-01	-0.31	4.91E-06	-0.55	3.85E-10	-0.40	1.09E-05	-1.35	1.49E-49
Rv3061c	acyl-CoA dehydrogenase FadE22(fadE22)	-0.40	4.46E-09	-0.80	6.01E-33	-1.06	9.59E-57	-0.67	7.29E-18	-0.36	6.05E-06	-1.09	1.90E-41
Rv3079c	hypothetical protein(Rv3079c)	-0.94	2.00E-28	-1.24	1.06E-48	-1.64	2.41E-78	-0.49	2.93E-04	-0.25	9.00E-02	-1.03	4.54E-14
Rv3080c	serine/threonine-protein kinase PknK(pknK)	-0.42	4.53E-18	-0.34	7.15E-13	-1.09	106	-0.62	1.05E-17	-0.03	7.50E-01	-0.80	7.83E-26
Rv3087	diacylglycerol O-acyltransferase(Rv3087)	0.37	1.77E-04	0.01	9.10E-01	-0.51	2.06E-07	-1.27	1.18E-13	-0.70	7.78E-05	-0.98	7.29E-08
Rv3088	diacylglycerol O-acyltransferase(tgs4)	-0.41	2.62E-08	-0.52	1.82E-12	-1.18	7.92E-58	-1.11	5.77E-19	-0.80	3.64E-10	-0.94	1.56E-12
Rv3089	long chain-fatty-acid--CoA ligase FadD13(fadD13)	-0.13	8.16E-02	-0.61	9.03E-19	-0.96	1.93E-44	-1.05	5.03E-12	-1.08	2.86E-12	-0.98	1.35E-09
Rv3101c	cell division protein FtsX(ftsX)	0.54	9.67E-12	0.42	9.24E-08	1.10	5.94E-48	0.40	4.19E-07	0.45	1.43E-08	0.28	6.81E-04
Rv3102c	cell division ATP-binding protein FtsE(ftsE)	0.84	1.86E-18	0.70	1.32E-13	1.28	1.46E-43	0.31	1.45E-02	0.53	2.58E-05	0.34	1.05E-02
Rv3110	pterin-4-alpha-carbinolamine dehydratase(moaB1)	-0.89	4.31E-02	-1.16	7.74E-03	-0.70	9.97E-02	0.52	3.06E-01	0.52	3.31E-01	0.06	9.23E-01
Rv3111	cyclic pyranopterin monophosphate synthase accessory protein(moaC1)	-1.14	7.25E-03	-0.76	5.35E-02	-0.47	2.30E-01	0.48	3.22E-01	0.45	3.93E-01	-0.14	8.08E-01
Rv3112	molybdenum cofactor biosynthesis protein MoaD(moaD1)	-0.43	2.64E-01	-1.02	6.93E-03	-0.30	4.19E-01	-0.01	9.92E-01	-0.05	9.14E-01	-0.03	9.50E-01
Rv3118	hypothetical protein(Rv3118)	0.66	2.13E-08	-0.10	4.36E-01	1.01	1.27E-18	0.06	6.79E-01	-0.01	9.50E-01	0.46	4.47E-05
Rv3125c	PPE family protein PPE49(PPE49)	-1.08	1.69E-20	-0.67	2.33E-09	-0.95	8.06E-17	0.18	3.03E-01	0.46	4.41E-03	0.24	1.64E-01
Rv3127	hypothetical protein(Rv3127)	-2.11	7.58E-85	-0.72	4.96E-11	-3.26	196	-0.69	4.73E-03	0.81	7.55E-04	-0.46	7.67E-02
Rv3128c	pseudo(Rv3128c)	-2.07	4.66E-34	-0.54	7.54E-04	-1.64	7.95E-24	-0.27	2.22E-01	0.43	4.83E-02	-0.13	5.94E-01
Rv3130c	diacylglycerol O-acyltransferase(tgs1)	-3.14	2.15E-85	-1.04	1.85E-10	-3.79	125	-1.00	2.60E-07	0.80	5.87E-05	-0.61	3.38E-03

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3131	NAD(P)H nitroreductase(Rv3131)	-1.32	3.13E-20	0.65	5.88E-06	-2.42	4.39E-65	-0.64	4.31E-03	1.17	6.54E-08	-0.31	2.21E-01
Rv3132c	two component sensor histidine kinase DevS(devS)	-1.85	3.58E-69	-0.85	8.36E-16	-2.38	2.64E-114	-0.35	8.76E-02	0.96	4.84E-07	-0.42	4.41E-02
Rv3133c	two component transcriptional regulator DevR(devR)	-2.18	5.99E-99	-0.66	2.84E-10	-2.56	2.43E-136	-0.17	4.39E-01	1.28	6.03E-12	-0.18	4.19E-01
Rv3134c	universal stress protein(Rv3134c)	-1.41	6.08E-23	0.00	9.88E-01	-1.63	1.19E-30	-0.52	5.85E-03	1.32	2.26E-13	-0.42	3.55E-02
Rv3135	PPE family protein PPE50(PPE50)	0.49	6.43E-12	1.08	7.00E-58	1.47	4.54E-107	0.63	1.24E-10	0.76	4.99E-15	0.78	7.43E-15
Rv3136	PPE family protein PPE51(PPE51)	0.26	1.24E-04	0.50	6.80E-14	0.38	1.67E-08	0.79	2.12E-19	1.09	4.67E-36	1.60	5.62E-70
Rv3136A	hypothetical protein(Rv3136A)	-1.09	9.38E-11	-1.43	1.42E-17	-0.82	4.77E-07	-0.21	4.92E-01	0.24	4.45E-01	-0.18	5.57E-01
Rv3137	histidinol-phosphatase(Rv3137)	-0.27	7.16E-04	0.07	3.90E-01	-0.37	3.42E-06	0.51	4.27E-04	0.91	1.40E-10	1.01	4.95E-12
Rv3139	acyl-CoA dehydrogenase(fadE24)	-0.53	1.24E-10	-1.45	8.90E-72	-0.99	1.40E-34	-0.39	5.06E-03	-0.22	1.34E-01	-0.62	1.03E-05
Rv3140	acyl-CoA dehydrogenase FadE23(fadE23)	-0.73	1.70E-24	-1.47	4.65E-96	-1.39	2.01E-86	-0.20	1.14E-01	-0.06	6.50E-01	-0.36	3.94E-03
Rv3143	response regulator(Rv3143)	1.21	5.79E-05	0.70	2.58E-02	1.76	4.82E-10	0.34	2.35E-01	0.17	6.06E-01	0.58	3.33E-02
Rv3159c	PPE family protein PPE53(PPE53)	-0.62	2.87E-08	-0.21	6.79E-02	-1.26	1.16E-30	-0.55	2.72E-05	-0.21	1.49E-01	-0.81	1.54E-09
Rv3160c	TetR family transcriptional regulator(Rv3160c)	0.11	1.78E-01	0.17	3.84E-02	-0.68	3.66E-18	-0.83	8.95E-08	-0.63	9.17E-05	-1.02	4.18E-10
Rv3161c	dioxygenase(Rv3161c)	-0.79	1.83E-20	-0.87	4.01E-25	-1.52	3.60E-74	-0.55	8.71E-06	-0.64	1.98E-07	-1.00	6.00E-15
Rv3165c	hypothetical protein(Rv3165c)	-0.94	6.43E-14	-0.29	1.97E-02	-1.18	9.47E-21	-0.08	6.38E-01	0.50	3.33E-04	-0.29	4.77E-02
Rv3166c	hypothetical protein(Rv3166c)	0.14	2.64E-01	0.21	8.47E-02	-0.18	1.58E-01	-0.77	1.98E-06	-0.22	2.27E-01	-1.05	3.55E-10
Rv3168	aminoglycoside phosphotransferase(Rv3168)	-0.44	2.77E-03	-0.25	9.55E-02	-0.25	8.09E-02	-0.59	5.55E-06	-0.08	5.99E-01	-1.02	2.80E-14
Rv3175	amidase(Rv3175)	0.44	4.05E-01	1.18	9.73E-03	0.57	2.39E-01	-0.57	3.03E-01	0.61	2.33E-01	-0.86	1.17E-01
Rv3179	hypothetical protein(Rv3179)	-0.61	1.86E-08	-0.34	1.38E-03	-1.08	7.88E-23	-0.03	8.84E-01	0.59	1.81E-05	-0.73	3.91E-07
Rv3180c	ribonuclease VapC45(Rv3180c)	0.87	3.15E-05	0.88	2.07E-05	0.35	1.14E-01	-0.64	3.47E-03	0.43	5.50E-02	-1.05	2.08E-06
Rv3188	hypothetical protein(Rv3188)	-0.32	4.55E-02	-0.26	1.03E-01	-0.67	1.92E-05	-0.76	4.13E-05	-0.13	5.77E-01	-1.08	1.83E-08
Rv3189	hypothetical protein(Rv3189)	-0.23	1.77E-01	-0.11	5.16E-01	-0.57	3.98E-04	-0.66	2.45E-07	0.12	4.29E-01	-1.01	2.37E-14
Rv3190A	hypothetical protein(Rv3190A)	-0.77	6.90E-34	-0.24	2.54E-04	-1.24	1.59E-85	0.63	1.65E-11	0.19	7.00E-02	0.63	1.02E-10
Rv3191c	transposase(Rv3191c)	0.50	3.25E-02	0.82	1.95E-04	0.95	1.24E-05	0.62	6.02E-02	0.58	9.40E-02	1.47	1.85E-06
Rv3197A	transcriptional regulator WhiB7(whiB7)	-0.56	9.96E-12	-1.01	9.76E-35	-1.56	4.89E-76	-0.56	1.60E-05	-0.14	3.34E-01	-0.72	6.40E-08

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3198A	glutaredoxin protein(Rv3198A)	0.20	1.97E-01	0.45	1.71E-03	1.38	1.05E-25	1.60	5.23E-12	1.05	1.83E-05	2.04	5.60E-18
Rv3202a	hypothetical protein(Rv3202a)	-1.07	1.28E-18	-0.84	1.51E-12	-1.37	3.13E-29	-0.01	9.68E-01	0.13	4.24E-01	-0.22	1.46E-01
Rv3209	hypothetical protein(Rv3209)	-0.43	7.47E-03	-0.19	2.59E-01	-0.91	4.23E-09	-0.21	1.57E-01	-0.01	9.53E-01	-1.19	1.04E-16
Rv3218	hypothetical protein(Rv3218)	0.24	3.42E-01	0.87	4.05E-05	1.51	6.99E-14	1.20	1.05E-04	1.22	1.40E-04	1.78	1.36E-09
Rv3219	transcriptional regulator WhiB1(whiB1)	-0.26	5.59E-05	-1.07	3.17E-63	-0.04	5.80E-01	0.26	1.69E-02	-0.28	1.11E-02	-0.15	2.13E-01
Rv3221c	acetyl-CoA carboxylase biotin carboxyl carrier protein subunit(TB7.3)	-1.10	1.09E-20	-0.59	7.39E-07	-0.97	9.94E-17	0.41	1.14E-02	0.19	2.87E-01	0.28	1.01E-01
Rv3224	iron-regulated short-chain dehydrogenase/reductase(Rv3224)	0.14	1.34E-01	0.65	8.49E-14	0.51	6.17E-09	0.88	3.65E-12	0.76	5.70E-09	1.53	7.92E-32
Rv3231c	hypothetical protein(Rv3231c)	0.60	1.97E-04	0.58	2.50E-04	1.07	8.20E-13	-0.08	6.93E-01	-0.13	5.45E-01	0.11	5.42E-01
Rv3233c	pseudo(Rv3233c)	0.24	4.78E-02	0.48	2.51E-05	1.00	7.90E-20	0.40	2.34E-03	0.17	2.61E-01	1.03	1.47E-16
Rv3246c	two component DNA-binding response regulator MtrA(mtrA)	-0.42	7.60E-11	-0.30	2.94E-06	-1.22	2.89E-84	0.15	1.49E-01	-0.09	4.31E-01	-0.06	5.78E-01
Rv3250c	rubredoxin RubB(rubB)	-0.68	7.53E-12	-0.53	8.62E-08	-0.76	1.63E-14	0.80	6.54E-09	0.55	1.27E-04	1.44	2.36E-24
Rv3251c	rubredoxin RubA(rubA)	-0.56	4.70E-05	-0.47	5.17E-04	-0.47	4.74E-04	0.50	2.95E-02	0.55	1.90E-02	1.22	2.71E-08
Rv3252c	transmembrane alkane 1-monooxygenase AlkB(alkB)	0.12	2.23E-01	-0.41	1.06E-05	0.03	7.93E-01	0.69	7.06E-08	0.43	1.19E-03	1.07	3.71E-16
Rv3268	hypothetical protein(Rv3268)	1.04	6.40E-31	0.74	1.76E-16	0.79	2.08E-18	0.29	1.46E-02	0.43	2.49E-04	-0.17	1.90E-01
Rv3269	hypothetical protein(Rv3269)	-0.57	1.84E-09	-0.66	2.04E-12	-1.65	1.80E-70	-0.86	2.20E-07	-0.29	1.08E-01	-1.34	5.32E-15
Rv3270	manganese/zinc-exporting P-type ATPase(ctpC)	-0.48	4.41E-07	-0.32	1.02E-03	-1.33	1.37E-46	-1.27	6.16E-18	-0.40	1.23E-02	-1.64	4.16E-26
Rv3271c	integral membrane protein(Rv3271c)	-0.80	7.64E-20	-0.36	4.05E-05	-0.32	2.46E-04	1.30	3.31E-10	1.26	1.81E-09	1.73	8.35E-16
Rv3277	transmembrane protein(Rv3277)	0.70	1.14E-13	0.49	2.44E-07	1.37	3.44E-51	0.05	7.57E-01	-0.03	8.66E-01	-0.22	1.35E-01
Rv3278c	transmembrane protein(Rv3278c)	-0.87	7.37E-12	-0.35	3.42E-03	-0.55	7.02E-06	1.23	9.42E-10	0.93	1.44E-05	1.42	1.26E-12
Rv3280	propionyl-CoA carboxylase subunit beta(accD5)	0.37	4.28E-09	0.44	2.14E-12	0.69	3.13E-29	0.66	9.35E-22	0.47	2.72E-11	1.14	4.39E-59
Rv3287c	anti-sigma factor RsbW(rsBW)	-0.48	1.11E-12	-0.84	4.56E-36	-1.11	2.61E-58	-0.44	3.70E-06	-0.61	1.16E-10	-0.69	2.32E-12
Rv3288c	hypothetical protein(usfY)	-0.92	2.35E-24	-1.71	6.77E-81	-2.06	3.50E-116	-0.68	1.13E-06	-1.06	8.98E-15	-1.10	2.59E-14
Rv3289c	transmembrane protein(Rv3289c)	-1.46	9.43E-16	-2.72	5.84E-51	-2.59	8.63E-47	-0.65	1.68E-02	-0.73	7.86E-03	-1.11	5.12E-05

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3290c	L-lysine-epsilon aminotransferase(lat)	-0.82	1.54E-20	-2.63	5.98E-200	-2.01	2.86E-118	-0.86	1.95E-11	-0.94	2.01E-13	-1.32	3.68E-23
Rv3291c	transcriptional regulator LrpA(lrpA)	-1.03	1.95E-05	-0.92	5.96E-05	-0.07	7.50E-01	-0.23	4.41E-01	-0.30	3.19E-01	-0.91	1.17E-03
Rv3308	phosphomannomutase PmmB(pmmB)	0.69	1.99E-15	0.82	4.09E-22	0.97	1.46E-30	0.57	7.16E-07	0.55	3.75E-06	1.14	2.11E-24
Rv3310	acid phosphatase(sapM)	1.14	2.56E-14	1.33	1.58E-19	1.26	1.06E-17	1.37	1.91E-15	0.99	4.81E-08	1.04	7.60E-09
Rv3311	hypothetical protein(Rv3311)	0.69	6.87E-11	1.02	6.36E-24	1.07	4.98E-26	1.13	1.52E-19	0.80	1.87E-09	1.26	2.49E-23
Rv3312A	pilin(Rv3312A)	1.60	1.41E-06	2.12	2.01E-11	3.14	8.17E-25	0.95	3.08E-03	1.28	5.34E-05	1.92	6.86E-11
Rv3312b	hypothetical protein(Rv3312b)	0.33	3.66E-01	0.21	5.47E-01	1.32	9.13E-06	1.66	2.77E-05	1.57	1.42E-04	2.50	1.66E-11
Rv3320c	ribonuclease VapC44(vapC44)	-1.13	2.97E-11	-0.61	2.20E-04	-0.93	2.18E-08	0.89	1.01E-03	0.33	2.88E-01	0.26	3.96E-01
Rv3329	aminotransferase(Rv3329)	0.61	5.95E-11	0.52	1.50E-08	1.12	7.79E-37	0.22	1.97E-01	0.30	7.26E-02	0.34	4.08E-02
Rv3334	MerR family transcriptional regulator(Rv3334)	-0.56	2.24E-08	-0.50	3.69E-07	-1.02	2.30E-23	-0.68	9.41E-09	-0.61	7.98E-07	-0.84	1.07E-11
Rv3351c	hypothetical protein(Rv3351c)	1.02	2.28E-02	1.25	3.48E-03	1.12	9.32E-03	1.36	2.15E-03	1.13	1.66E-02	1.58	2.03E-04
Rv3354	hypothetical protein(Rv3354)	0.61	1.50E-01	-0.06	9.02E-01	0.88	2.65E-02	-0.09	8.78E-01	-0.11	8.47E-01	-1.02	4.28E-02
Rv3358	toxin RelK(relK)	-0.68	7.59E-04	-1.07	1.07E-07	-0.68	6.20E-04	-0.43	2.49E-02	-0.42	3.99E-02	-0.54	5.75E-03
Rv3360	hypothetical protein(Rv3360)	-0.57	7.15E-14	-0.40	1.08E-07	-1.23	7.55E-56	0.56	1.18E-04	0.15	3.86E-01	0.19	2.56E-01
Rv3361c	hypothetical protein(Rv3361c)	0.55	3.42E-05	1.16	5.18E-21	1.29	9.89E-26	1.00	1.18E-09	0.80	4.43E-06	1.38	1.32E-17
Rv3362c	ATP/GTP-binding protein(Rv3362c)	0.86	3.20E-17	1.30	2.11E-41	1.54	4.32E-58	0.78	1.50E-08	0.63	1.21E-05	1.07	2.06E-15
Rv3363c	hypothetical protein(Rv3363c)	0.84	4.84E-07	1.16	5.37E-13	1.53	2.62E-22	0.93	3.60E-06	0.61	5.59E-03	1.22	5.08E-10
Rv3364c	hypothetical protein(Rv3364c)	0.36	4.42E-03	0.64	1.20E-07	0.89	5.22E-14	0.93	3.11E-07	0.82	1.46E-05	1.31	1.74E-13
Rv3365c	hypothetical protein(Rv3365c)	0.88	3.64E-45	1.11	7.63E-74	1.48	1.46E-133	0.53	5.24E-10	0.66	8.41E-15	0.90	1.73E-25
Rv3366	tRNA/rRNA methylase SpoU(spoU)	1.54	2.58E-04	1.96	9.82E-07	2.33	3.13E-09	1.63	8.20E-04	1.57	1.99E-03	2.55	1.54E-08
Rv3371	diacylglycerol O-acyltransferase(Rv3371)	-0.13	2.85E-01	-0.97	5.21E-17	-0.12	2.93E-01	-1.05	3.18E-13	-1.40	4.17E-21	-1.38	9.48E-20
Rv3375	amidase(amiD)	1.00	3.55E-17	0.93	1.62E-15	1.30	6.91E-30	0.41	1.08E-03	0.35	9.63E-03	0.47	1.68E-04
Rv3383c	polyprenyl synthetase IdsB(idsB)	-0.01	9.18E-01	0.87	1.20E-33	0.45	6.48E-10	1.15	3.67E-33	0.46	5.41E-06	0.84	1.96E-16
Rv3384c	ribonuclease VapC46(vapC46)	1.12	1.76E-20	0.70	1.62E-08	0.81	3.37E-11	-0.54	4.22E-03	-0.39	4.64E-02	-1.25	3.35E-11
Rv3388	PE-PGRS family protein PE_PGRS52(PE_PGRS52)	-0.18	5.08E-01	0.33	2.10E-01	-0.16	5.55E-01	-0.14	7.37E-01	1.19	4.61E-04	0.35	3.61E-01
Rv3392c	cyclopropane mycolic acid synthase CmaA(cmaA1)	0.49	3.73E-04	0.41	2.70E-03	0.97	5.28E-14	1.05	1.59E-08	0.71	2.83E-04	0.73	1.77E-04

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3393	nucleoside hydrolase(iunH)	0.61	4.02E-05	0.96	4.30E-12	1.16	3.59E-17	0.93	7.44E-07	0.81	4.44E-05	0.69	3.62E-04
Rv3395A	membrane protein(Rv3395A)	1.29	7.34E-02	0.40	6.13E-01	2.48	1.08E-04	2.95	1.41E-02	3.42	4.29E-03	3.70	1.07E-03
Rv3397c	phytoene synthase(phyA)	0.80	8.10E-05	1.27	6.01E-11	1.25	9.84E-11	-0.01	9.72E-01	-0.17	4.95E-01	-0.07	7.76E-01
	multifunctional dimethylal-lyltransferase/geranyl-transtransferase/farnesyl-transtransferase(idsA1)	0.72	9.36E-05	1.41	8.13E-17	0.94	8.20E-08	0.93	9.98E-05	1.00	4.15E-05	0.61	1.38E-02
Rv3402c	hypothetical protein(Rv3402c)	1.03	4.38E-23	0.86	2.28E-16	1.62	1.13E-56	0.75	4.41E-04	0.21	3.86E-01	1.40	1.07E-10
Rv3403c	hypothetical protein(Rv3403c)	0.86	4.48E-20	1.45	7.08E-59	1.74	8.05E-86	0.56	4.61E-05	0.44	2.45E-03	0.60	3.10E-05
Rv3407	antitoxin VapB47(vapB47)	-0.56	4.87E-12	-0.08	3.58E-01	-0.90	9.85E-30	1.14	9.97E-11	0.72	7.20E-05	1.17	2.47E-10
	ECF RNA polymerase sigma factor SigD(sigD)	-0.55	1.56E-08	-0.50	4.08E-07	-1.21	6.59E-36	0.44	1.29E-02	0.11	6.11E-01	-0.54	2.51E-03
Rv3417c	chaperonin GroEL(groEL1)	-0.57	2.11E-06	-1.76	4.67E-52	-1.07	3.20E-20	-0.69	5.08E-07	-0.78	1.18E-08	-0.80	1.95E-08
Rv3418c	chaperonin GroES(groES)	-1.11	5.88E-13	-2.29	6.96E-52	-1.16	2.54E-14	-0.59	1.67E-06	-0.47	2.25E-04	-0.69	9.64E-08
Rv3424c	hypothetical protein(Rv3424c)	0.85	5.78E-08	0.34	3.70E-02	1.14	6.66E-14	-0.83	3.16E-03	-0.84	3.17E-03	-2.00	9.89E-13
	PPE family protein												
Rv3425	PPE57(PPE57)	0.06	6.86E-01	0.97	9.53E-18	0.44	2.73E-04	1.23	1.10E-15	0.75	7.14E-06	1.83	5.81E-33
	PPE family protein												
Rv3426	PPE58(PPE58)	0.08	5.22E-01	0.78	7.86E-14	0.27	1.66E-02	0.70	1.09E-04	0.33	1.10E-01	1.49	1.37E-17
Rv3427c	transposase(Rv3427c)	0.67	2.18E-03	1.06	2.25E-07	0.68	1.39E-03	-0.05	8.83E-01	0.37	1.93E-01	-0.85	2.56E-03
Rv3438	hypothetical protein(Rv3438)	1.35	5.96E-19	0.94	1.24E-09	1.69	1.40E-30	-0.36	6.39E-02	0.13	5.68E-01	0.08	7.16E-01
Rv3440c	hypothetical protein(Rv3440c)	0.67	9.78E-05	0.60	4.87E-04	1.10	2.20E-11	-0.17	3.57E-01	0.40	2.07E-02	0.10	6.19E-01
Rv3442c	30S ribosomal protein S9(rpsI)	0.64	2.57E-16	0.19	1.76E-02	1.00	9.27E-40	0.14	1.74E-01	0.14	1.67E-01	0.60	6.12E-11
Rv3444c	ESAT-6 like protein EsxT(esxT)	1.37	7.95E-03	1.16	2.50E-02	2.24	2.59E-06	0.69	2.62E-01	0.72	2.57E-01	1.06	5.70E-02
	ESX-4 secretion system protein												
Rv3448	EccD4(eccD4)	1.13	1.63E-03	1.01	4.48E-03	0.93	9.28E-03	-0.32	3.98E-01	0.40	2.79E-01	-0.20	6.06E-01
	membrane-anchored myco-												
Rv3449	sin(mycP4)	0.71	1.50E-02	0.61	3.50E-02	1.02	2.09E-04	0.01	9.80E-01	-0.40	2.39E-01	-0.33	2.98E-01
Rv3453	pseudo(Rv3453)	0.56	5.04E-03	0.51	9.87E-03	1.46	4.59E-16	0.38	4.70E-02	0.03	8.96E-01	0.55	2.57E-03
	50S ribosomal protein												
Rv3461c	L36(rpmJ)	-1.20	7.23E-30	-1.20	3.09E-30	-0.96	7.30E-20	0.11	4.64E-01	0.07	6.93E-01	0.04	8.15E-01
	translation initiation factor IF-												
Rv3462c	1(infA)	-1.34	1.15E-33	-1.27	1.97E-30	-1.18	8.20E-27	0.36	6.73E-02	0.12	6.07E-01	0.08	7.34E-01
Rv3466	hypothetical protein(Rv3466)	-0.88	3.43E-12	-0.56	5.93E-06	-1.21	2.27E-21	0.22	2.51E-01	0.03	9.01E-01	-0.20	3.33E-01
	4-hydroxy-2-oxovalerate al-												
Rv3469c	dolase MhpE(mhpE)	0.39	1.35E-01	0.56	2.17E-02	1.08	2.81E-06	0.56	1.51E-02	0.45	6.51E-02	0.56	1.34E-02
Rv3476c	kgtP	0.41	9.47E-03	0.49	1.16E-03	1.14	1.04E-15	0.43	4.23E-02	0.16	5.29E-01	0.42	4.77E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3477	PE family protein PE31(PE31)	-0.02	8.49E-01	0.06	4.67E-01	0.81	1.16E-30	1.30	1.05E-42	0.80	1.61E-16	2.08	3.20E-98
Rv3478	PE family protein PPE60(PPE60)	-0.16	1.24E-02	0.25	4.39E-05	0.40	4.46E-11	0.75	4.50E-16	0.49	1.85E-07	1.57	3.27E-61
Rv3481c	integral membrane protein(Rv3481c)	0.73	9.27E-07	0.32	4.24E-02	1.25	5.62E-19	0.65	1.82E-04	0.68	1.48E-04	0.82	1.68E-06
Rv3482c	membrane protein(Rv3482c)	1.19	6.02E-36	0.90	3.85E-21	1.24	8.38E-40	0.09	4.85E-01	0.21	9.01E-02	0.41	4.41E-04
Rv3485c	short-chain type dehydrogenase/reductase(Rv3485c)	0.08	3.72E-01	0.71	4.17E-20	0.64	1.06E-16	0.82	5.15E-16	0.56	1.71E-07	1.30	5.22E-38
Rv3487c	carboxylesterase LipF(lipF)	0.23	1.20E-04	0.74	6.04E-38	1.05	1.86E-76	1.45	4.22E-40	1.24	1.00E-28	1.99	1.22E-68
Rv3489	hypothetical protein(Rv3489)	-1.04	8.00E-28	-0.72	3.66E-14	-0.74	2.33E-15	0.58	5.91E-04	0.35	5.28E-02	0.55	1.77E-03
Rv3491	hypothetical protein(Rv3491)	-1.00	2.82E-32	-0.72	4.72E-18	-0.81	4.71E-22	0.05	6.30E-01	-0.33	1.56E-04	-0.43	5.82E-07
Rv3503c	ferredoxin FdxD(fdxD)	-1.55	4.19E-20	-1.27	6.17E-14	-2.08	3.20E-35	0.00	9.99E-01	0.24	2.75E-01	-0.15	5.12E-01
Rv3513c	fatty-acid--CoA ligase FadD18(fadD18)	-0.26	3.23E-02	-1.09	1.36E-19	-0.55	2.75E-06	-0.44	1.86E-05	-0.27	1.39E-02	-0.43	4.09E-05
Rv3516	enoyl-CoA hydratase EchA19(echA19)	0.03	8.48E-01	-0.57	6.91E-06	-0.62	1.37E-06	-0.49	1.64E-03	-0.44	6.92E-03	-1.01	3.22E-10
Rv3517	hypothetical protein(Rv3517)	0.59	1.14E-02	0.50	2.97E-02	0.25	3.08E-01	0.81	1.19E-03	1.08	1.36E-05	0.46	8.23E-02
Rv3524	membrane protein(Rv3524)	0.91	1.75E-25	0.62	1.38E-12	1.23	5.27E-47	0.73	2.52E-09	0.50	1.00E-04	0.40	2.63E-03
Rv3526	3-ketosteroid-9-alpha-monooxygenase oxygenase subunit(kshA)	-0.83	1.83E-36	-1.37	5.95E-96	-1.19	2.72E-73	-0.40	1.05E-04	-0.65	2.43E-10	-0.75	1.02E-12
Rv3528c	hypothetical protein(Rv3528c)	-1.28	4.02E-35	-1.33	3.12E-38	-1.05	1.35E-24	0.37	1.61E-03	-0.30	1.79E-02	0.17	2.02E-01
Rv3529c	hypothetical protein(Rv3529c)	-0.73	2.11E-19	-0.66	1.11E-16	-1.21	2.57E-48	-0.53	2.90E-10	-0.32	2.41E-04	-0.96	3.00E-28
Rv3531c	hypothetical protein(Rv3531c)	-0.18	5.80E-02	-0.25	8.98E-03	-0.49	1.19E-07	-0.86	3.01E-11	-0.46	6.13E-04	-1.14	2.55E-17
Rv3545c	steroid C26-monooxygenase(cyp125)	-0.63	5.59E-18	-0.84	2.28E-31	-1.17	1.44E-57	-0.78	1.91E-32	-0.37	3.24E-08	-1.07	4.28E-55
Rv3548c	short-chain type dehydrogenase/reductase(Rv3548c)	-1.14	2.24E-37	-1.02	1.70E-30	-1.28	5.44E-47	-0.60	1.94E-08	-0.03	8.27E-01	-0.46	4.01E-05
Rv3549c	short-chain type dehydrogenase/reductase(Rv3549c)	-0.99	2.95E-31	-0.73	7.27E-18	-1.08	1.80E-37	-0.51	1.04E-04	-0.01	9.54E-01	-0.52	1.26E-04
Rv3554	electron transfer protein FdxB(fdxB)	1.00	2.38E-11	1.27	1.47E-18	1.15	3.77E-15	-0.30	9.26E-02	0.04	8.74E-01	-0.04	8.28E-01
Rv3563	acyl-CoA dehydrogenase FadE32(fadE32)	0.28	3.43E-02	0.52	2.17E-05	-0.06	6.46E-01	-1.05	3.57E-10	-0.04	8.51E-01	-0.50	4.01E-03
Rv3570c	flavin-dependent monooxygenase oxygenase subunit HsaA(hsaA)	-0.91	2.36E-29	-0.38	3.62E-06	-1.24	2.21E-54	-0.16	2.22E-01	0.04	8.24E-01	-0.41	1.06E-03

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3575c	LacI family transcriptional regulator(Rv3575c)	0.20	3.84E-02	0.32	6.41E-04	0.47	2.18E-07	1.04	2.98E-15	0.81	3.77E-09	1.45	6.00E-28
Rv3577	hypothetical protein(Rv3577)	1.04	1.98E-13	1.21	1.55E-18	1.51	1.00E-28	-0.02	9.17E-01	0.27	1.31E-01	0.10	6.18E-01
Rv3578	arsenic transport integral membrane protein												
Rv3578	ArsB(arsB2)	1.05	4.87E-11	1.26	2.58E-16	1.36	5.81E-19	0.52	2.04E-02	0.81	2.19E-04	1.05	2.20E-07
Rv3579c	23S rRNA (guanosine(2251)-2'-O)-methyltransferase												
Rv3579c	RlmB(Rv3579c)	-0.01	9.22E-01	0.44	2.74E-06	0.36	9.66E-05	0.96	1.35E-17	0.85	1.35E-13	1.12	1.36E-21
Rv3582c	2-C-methyl-D-erythritol 4-phosphate cytidyltransferase(ispD)	-0.80	1.19E-20	-0.88	1.04E-24	-1.20	1.30E-45	-0.02	8.94E-01	-0.12	4.49E-01	-0.45	1.72E-03
Rv3583c	RNA polymerase-binding transcription factor CarD(Rv3583c)	-1.02	5.89E-40	-1.06	5.09E-43	-1.52	2.04E-87	0.04	7.81E-01	-0.24	6.69E-02	-0.32	1.46E-02
Rv3584	lipoprotein LpqE(lpqE)	0.42	3.94E-05	0.29	5.30E-03	1.09	4.49E-29	-0.01	9.74E-01	0.04	8.39E-01	0.33	2.48E-02
Rv3586	DNA integrity scanning protein DisA(Rv3586)	1.21	8.14E-46	1.07	1.79E-36	0.64	2.70E-13	0.34	9.82E-03	0.45	5.26E-04	0.11	4.65E-01
Rv3589	A/G-specific adenine glycosylase(mutY)	1.20	3.88E-15	1.04	6.39E-12	1.67	2.62E-30	0.59	1.43E-02	1.03	9.19E-06	1.06	3.26E-06
Rv3597c	iron-regulated H-NS-like protein(Isr2)	-1.55	5.49E-37	-1.54	1.06E-36	-1.66	6.36E-43	0.35	2.39E-01	-0.03	9.28E-01	-0.08	8.06E-01
Rv3598c	lysine--tRNA ligase(lysS)	0.73	2.15E-14	0.55	7.47E-09	1.76	1.72E-85	0.96	1.37E-17	0.70	2.00E-09	1.28	2.00E-28
Rv3602c	pantothenate synthetase(panC)	0.91	3.70E-18	0.70	2.02E-11	1.32	8.57E-39	-0.41	9.13E-03	0.08	6.85E-01	-0.65	4.69E-05
Rv3613c	hypothetical protein(Rv3613c)	-0.26	9.45E-02	-0.70	1.92E-06	-0.15	3.19E-01	-1.08	2.47E-07	-1.12	9.97E-08	-0.62	5.96E-03
Rv3614c	ESX-1 secretion-associated protein EspD(espD)	-1.16	2.04E-38	-1.31	2.18E-48	-1.27	5.40E-46	-1.16	2.18E-17	-1.17	1.40E-17	-0.67	5.35E-06
Rv3615c	ESX-1 secretion-associated protein EspC(espC)	-0.81	1.32E-21	-1.23	5.15E-48	-1.20	2.44E-46	-1.06	4.42E-09	-1.37	1.53E-14	-0.60	2.26E-03
Rv3616c	ESX-1 secretion-associated protein EspA(espA)	-1.21	7.57E-46	-1.51	3.99E-71	-1.29	6.71E-53	-1.13	1.06E-14	-1.34	3.26E-20	-0.92	3.34E-09
Rv3620c	ESAT-6 like protein EsxW(esxW)	-1.32	1.55E-22	-0.98	1.49E-13	-1.39	3.32E-25	0.22	1.69E-01	-0.07	7.09E-01	0.75	1.14E-07
Rv3622c	PE family protein PE32(PE32)	-0.22	6.02E-01	0.21	5.82E-01	-0.46	2.54E-01	1.44	8.14E-04	1.01	3.19E-02	1.40	1.14E-03
Rv3634c	UDP-glucose 4-epimerase(galE1)	1.03	6.36E-22	0.25	2.85E-02	0.87	3.42E-16	0.11	6.11E-01	-0.01	9.82E-01	0.04	8.70E-01
Rv3635	transmembrane protein(Rv3635)	0.96	2.70E-16	0.90	6.34E-15	1.33	4.24E-32	0.16	2.64E-01	0.43	1.07E-03	0.14	3.19E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3642c	hypothetical protein(Rv3642c)	-1.03	2.12E-11	-0.27	8.69E-02	-0.62	4.23E-05	0.45	1.75E-02	0.31	1.25E-01	-0.05	8.44E-01
Rv3648c	cold shock protein A(cspA)	-1.17	4.01E-34	-0.24	1.46E-02	-0.94	8.59E-23	0.83	1.57E-07	0.56	5.85E-04	1.46	3.19E-19
Rv3650	PE family protein PE33(PE33)	-1.11	8.71E-06	-0.80	8.62E-04	-1.58	6.43E-10	-0.36	3.51E-01	-0.44	2.68E-01	-0.31	4.15E-01
Rv3651	hypothetical protein(Rv3651)	0.42	2.65E-03	1.37	7.35E-27	0.99	2.49E-14	0.50	7.62E-03	0.34	8.96E-02	0.54	4.25E-03
Rv3660c	hypothetical protein(Rv3660c)	0.70	2.16E-11	1.21	5.25E-33	0.89	1.97E-18	0.54	2.70E-04	0.90	3.10E-10	0.87	4.13E-09
	dipeptide ABC transporter substrate-binding lipoprotein												
Rv3666c	DppA(dppA)	0.77	9.44E-09	1.02	3.11E-15	1.02	3.94E-15	0.46	2.79E-03	0.46	4.08E-03	0.55	3.43E-04
Rv3675	membrane protein(Rv3675)	-1.53	2.30E-59	-1.56	2.63E-63	-1.82	1.97E-82	-0.10	6.22E-01	-1.18	6.05E-13	-0.79	2.71E-06
Rv3678A	hypothetical protein(Rv3678A)	-1.24	2.97E-10	-0.24	2.39E-01	-0.69	4.25E-04	1.04	2.15E-05	1.06	2.00E-05	1.40	2.37E-08
	transcriptional regulator												
Rv3681c	WhiB4(whiB4)	-1.04	8.12E-36	-1.11	1.72E-41	-0.88	7.90E-27	0.05	7.56E-01	-0.55	6.43E-06	-0.36	4.83E-03
	hypothetical protein(Rv3686c)						7.40E-						
Rv3686c		0.76	1.17E-19	1.18	1.62E-47	1.83	116	1.12	4.05E-24	1.02	1.83E-19	1.55	1.34E-43
Rv3688c	hypothetical protein(Rv3688c)	-0.99	5.25E-17	-0.25	3.48E-02	-0.58	5.62E-07	0.80	9.61E-07	0.52	2.40E-03	1.05	2.90E-10
Rv3705A	hypothetical protein(Rv3705A)	0.99	9.61E-05	0.48	6.90E-02	1.09	1.18E-05	-0.33	2.96E-01	0.14	6.77E-01	0.49	7.87E-02
Rv3706c	hypothetical protein(Rv3706c)	1.84	1.58E-09	1.45	2.62E-06	2.08	3.83E-12	0.55	9.38E-02	0.73	2.53E-02	1.20	7.95E-05
	2-isopropylmalate synthase(leuA)												
Rv3710		0.69	7.50E-14	1.46	3.33E-58	0.53	5.75E-09	0.30	8.93E-06	0.28	3.19E-05	0.14	5.02E-02
Rv3712	ligase(Rv3712)	0.89	2.96E-11	0.97	1.14E-13	1.33	2.30E-25	0.44	3.53E-03	0.74	6.88E-07	0.96	1.10E-11
	cobryic acid synthase												
Rv3713	CobQ(cobQ2)	0.55	1.28E-06	0.24	3.99E-02	0.67	1.72E-09	0.90	3.45E-07	0.48	1.26E-02	1.16	6.87E-11
Rv3714c	hypothetical protein(Rv3714c)	0.31	1.92E-03	0.67	1.04E-12	0.50	1.81E-07	1.25	5.41E-17	1.21	2.95E-15	1.28	6.93E-17
Rv3726	dehydrogenase(Rv3726)	0.21	6.01E-03	0.17	1.89E-02	0.29	5.03E-05	0.77	5.05E-13	0.48	2.57E-05	1.57	1.57E-49
	transmembrane protein(Rv3737)												
Rv3737		0.87	4.26E-24	0.77	1.50E-19	1.05	2.84E-35	0.20	1.79E-01	0.61	1.01E-05	0.21	1.56E-01
	PPE family protein												
Rv3738c	PPE66(PPE66)	0.11	4.80E-01	0.50	9.88E-05	0.41	1.69E-03	0.67	4.31E-04	0.49	1.70E-02	1.09	1.28E-09
	PPE family protein												
Rv3739c	PPE67(PPE67)	0.03	8.89E-01	0.72	1.78E-05	1.17	3.44E-13	0.62	1.40E-02	0.48	8.14E-02	1.13	1.77E-06
Rv3747	hypothetical protein(Rv3747)	0.17	6.15E-02	-0.09	3.19E-01	1.10	1.86E-43	1.20	2.73E-22	0.67	3.11E-07	1.62	1.08E-37
Rv3748	hypothetical protein(Rv3748)	0.00	9.72E-01	-0.04	6.87E-01	0.64	9.25E-14	0.83	2.24E-12	0.31	2.12E-02	1.15	3.21E-22
	cytidine/deoxycytidylate deaminase(Rv3752c)												
Rv3752c		0.73	2.47E-05	1.06	8.53E-11	1.43	2.04E-19	0.58	2.01E-02	0.61	1.84E-02	1.19	2.53E-07
Rv3763	lipoprotein LpqH(lpqH)	-1.00	1.98E-39	-1.19	7.92E-56	-0.84	1.00E-28	0.10	3.90E-01	-0.42	2.25E-05	0.41	4.99E-05
	two component sensor kinase												
Rv3764c	TcrY(tcrY)	1.34	2.48E-39	1.00	2.33E-22	0.96	2.57E-20	0.70	4.76E-09	0.02	8.84E-01	0.29	3.13E-02

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3765c	two component transcriptional regulator TcrX(tcrX)	0.37	2.02E-06	0.13	1.18E-01	0.18	2.54E-02	1.16	3.31E-21	0.36	8.63E-03	0.75	1.30E-08
Rv3767c	S-adenosylmethionine-dependent methyltransferase(Rv3767c)	0.97	5.84E-36	0.71	5.06E-20	1.31	1.07E-67	0.40	2.49E-05	0.25	1.24E-02	0.38	1.50E-04
Rv3772	histidinol-phosphate aminotransferase(hisC2)	1.08	2.20E-13	0.96	4.97E-11	1.45	2.15E-24	-0.19	3.98E-01	0.16	4.83E-01	0.00	9.87E-01
Rv3774	enoyl-CoA hydratase EchA21(echA21)	-0.03	8.13E-01	0.61	1.69E-11	-0.10	3.16E-01	0.62	7.23E-09	0.63	1.20E-08	1.20	1.00E-28
Rv3782	galactofuranosyl transferase GlfT(glfT1)	0.93	3.69E-46	1.29	8.25E-93	1.33	3.39E-97	0.33	5.68E-04	0.44	4.59E-06	0.39	7.62E-05
Rv3783	O-antigen/lipopolysaccharide ABC transporter permease RfbD(rfbD)	0.71	2.25E-16	1.07	7.96E-38	1.15	1.81E-43	0.30	4.12E-03	0.43	3.12E-05	0.23	3.04E-02
Rv3789	GtrA family protein(Rv3789)	1.18	1.15E-06	1.43	9.76E-10	2.03	2.94E-19	0.38	1.89E-01	0.49	8.83E-02	0.73	5.78E-03
Rv3790	decaprenylphosphoryl-beta-D-ribose oxidase(dprE1)	0.51	3.18E-04	1.13	5.05E-18	1.78	1.11E-44	0.66	6.92E-05	0.87	1.31E-07	1.02	2.15E-10
Rv3791	decaprenylphosphoryl-D-2-keto erythropentose reductase(dprE2)	0.22	1.87E-02	0.45	5.01E-07	0.74	1.53E-17	1.27	4.69E-22	0.96	3.76E-12	1.82	2.09E-43
Rv3792	arabinofuranosyltransferase(aftA)	1.02	9.97E-21	0.97	2.70E-19	1.52	2.96E-47	0.46	4.63E-03	0.83	1.25E-07	0.90	6.61E-09
Rv3793	arabinoxyltransferase C(embC)	0.75	1.38E-16	0.99	2.81E-28	0.87	5.94E-22	0.56	3.49E-05	0.74	3.52E-08	1.13	2.30E-17
Rv3794	arabinoxyltransferase A(embA)	1.14	8.14E-44	1.37	5.21E-64	1.67	3.48E-95	1.33	2.86E-24	1.50	2.88E-30	2.08	2.31E-53
Rv3795	arabinoxyltransferase B(embB)	0.44	4.87E-07	0.99	5.66E-32	0.88	1.85E-25	0.94	1.69E-12	1.13	3.00E-17	1.59	1.18E-32
Rv3796	hypothetical protein(Rv3796)	1.15	3.48E-04	1.67	2.90E-08	1.95	4.15E-11	1.51	4.77E-05	1.30	8.93E-04	1.81	6.40E-07
Rv3797	acyl-CoA dehydrogenase FadE35(fadE35)	1.17	9.15E-14	1.32	8.74E-18	1.60	3.15E-26	0.86	2.56E-07	1.08	9.32E-11	1.09	5.21E-11
Rv3803c	MPT51/MPB51 antigen(fbpD)	-0.59	5.54E-12	-1.05	4.50E-35	-0.42	7.01E-07	-0.34	8.09E-02	-0.35	7.66E-02	-0.24	2.45E-01
Rv3810	cell surface protein(pirG)	-0.07	4.33E-01	-0.41	1.56E-06	-0.51	1.18E-09	-0.51	3.87E-05	-0.11	4.31E-01	-1.31	2.08E-25
Rv3814c	acyltransferase(Rv3814c)	-0.57	6.89E-24	-0.72	2.21E-37	-1.31	4.55E-118	-0.08	2.89E-01	0.07	3.34E-01	0.14	5.21E-02
Rv3826	long-chain-fatty-acid--CoA ligase FadD23(fadD23)	0.77	9.21E-18	0.68	1.91E-14	1.61	5.40E-80	0.97	1.26E-15	0.61	1.89E-06	1.25	4.25E-24
Rv3833	AraC family transcriptional regulator(Rv3833)	-0.35	1.26E-01	0.22	3.31E-01	0.05	8.48E-01	-1.18	5.90E-12	-0.33	8.23E-02	-1.76	3.34E-22
Rv3840	transcriptional regulator(Rv3840)	1.08	1.01E-07	1.10	3.21E-08	1.40	8.44E-13	-0.10	7.23E-01	0.45	5.63E-02	0.20	4.24E-01

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3841	bacterioferritin BfrB(bfrB)	-1.18	8.17E-14	-1.33	3.49E-17	-2.93	1.26E-79	-0.18	3.78E-01	0.73	3.14E-05	-0.10	6.57E-01
Rv3848	transmembrane protein(Rv3848)	-0.43	2.09E-11	-1.21	3.57E-82	-0.68	1.00E-27	-0.17	1.03E-01	-0.82	4.61E-19	-0.62	2.17E-10
Rv3851	membrane protein(Rv3851)	1.92	2.79E-10	1.32	2.38E-05	1.82	1.74E-09	0.32	3.76E-01	0.28	4.65E-01	0.19	6.22E-01
Rv3854c	monooxygenase EthA(ethA)	-0.67	8.62E-13	-1.07	4.70E-31	-1.83	1.14E-88	-0.51	9.87E-03	-0.77	6.46E-05	-0.81	4.42E-05
Rv3861	hypothetical protein(Rv3861)	0.40	3.57E-01	-0.57	2.16E-01	1.31	4.53E-04	0.21	6.83E-01	0.48	3.20E-01	0.41	3.82E-01
Rv3862c	transcriptional regulator WhiB6(whiB6)	0.88	2.00E-05	1.55	4.72E-15	0.70	7.28E-04	0.10	6.79E-01	1.84	4.66E-24	-0.45	3.37E-02
Rv3864	ESX-1 secretion-associated protein EspE(espE)	-0.47	1.90E-15	-0.52	8.51E-19	-1.06	3.21E-74	-0.20	4.14E-02	-0.26	8.99E-03	-0.27	7.25E-03
Rv3865	ESX-1 secretion-associated protein EspF(espF)	-1.24	2.99E-41	-1.03	7.83E-29	-1.63	1.84E-70	0.10	6.35E-01	-0.07	7.32E-01	-0.02	9.43E-01
Rv3872	PE family protein PE35(PE35)	-1.15	1.11E-23	-0.64	2.99E-08	-1.24	1.30E-27	-0.36	5.91E-02	-0.23	2.63E-01	-0.92	9.52E-07
Rv3874	ESAT-6-like protein EsxB(esxB)	-1.37	2.03E-32	-0.96	1.33E-16	-1.50	2.84E-39	-0.03	8.95E-01	0.18	3.16E-01	0.18	3.20E-01
Rv3875	ESAT-6 protein EsxA(esxA)	-0.73	2.18E-39	-0.69	9.10E-35	-1.03	1.13E-77	0.16	8.73E-02	0.00	9.98E-01	0.20	3.50E-02
Rv3877	ESX-1 secretion system protein EccD1(eccD1)	-0.96	2.91E-33	-1.01	7.08E-38	-1.02	8.94E-38	-0.47	3.70E-05	-0.57	7.80E-07	-0.83	7.08E-13
Rv3882c	ESX-1 secretion system protein EccE1(eccE1)	0.98	6.05E-56	1.05	2.51E-65	1.26	5.72E-95	0.76	1.82E-23	0.74	2.21E-21	1.15	8.53E-49
Rv3883c	membrane-anchored mycosin(mycP1)	1.01	4.03E-62	0.79	1.81E-39	1.16	8.00E-84	0.47	1.71E-09	0.57	1.73E-13	0.59	1.65E-13
Rv3890c	ESAT-6 like protein EsxC(esxC)	-0.32	1.07E-03	-0.16	1.00E-01	-0.05	6.06E-01	0.63	5.37E-07	0.37	7.46E-03	1.15	2.49E-20
Rv3891c	ESAT-6 like protein EsxD(esxD)	-0.96	8.43E-17	-0.54	2.08E-06	-0.80	2.19E-12	0.77	1.15E-06	0.74	5.64E-06	1.26	2.06E-15
Rv3892c	PPE family protein PPE69(PPE69)	0.43	1.67E-02	0.57	9.19E-04	0.82	1.37E-06	0.87	5.50E-05	0.83	2.17E-04	1.38	2.30E-11
Rv3899c	hypothetical protein(Rv3899c)	0.87	8.87E-06	1.10	3.09E-09	1.26	8.70E-12	0.51	2.14E-02	0.65	3.17E-03	0.58	8.08E-03
Rv3904c	ESAT-6 like protein EsxE(esxE)	-0.86	8.76E-15	-0.72	5.75E-11	-0.78	1.47E-12	0.75	6.39E-04	0.58	1.13E-02	1.25	1.48E-08
Rv3905c	ESAT-6 like protein EsxF(esxF)	-1.34	4.17E-15	-0.90	1.42E-07	-0.75	1.13E-05	0.94	8.22E-06	0.62	4.93E-03	1.62	4.46E-14
Rv3906c	hypothetical protein(Rv3906c)	-0.01	9.08E-01	-0.54	3.37E-12	0.45	2.46E-09	0.65	4.20E-17	0.12	2.04E-01	1.34	2.89E-64
Rv3908	mutator protein MutT(mutT4)	0.68	1.69E-16	0.67	1.60E-16	1.02	1.27E-37	0.18	8.36E-02	0.53	9.96E-08	0.36	3.68E-04
Rv3909	hypothetical protein(Rv3909)	1.52	1.06E-42	1.43	1.21E-38	1.70	2.21E-54	0.05	8.15E-01	0.36	5.50E-02	0.20	3.25E-01
Rv3918c	chromosome partitioning protein ParA(parA)	0.88	1.69E-24	1.12	6.26E-40	0.97	2.71E-30	0.09	5.84E-01	0.14	3.86E-01	0.13	3.89E-01
Rv3920c	hypothetical protein(Rv3920c)	0.49	1.41E-07	1.11	5.74E-35	1.34	1.53E-50	1.26	1.02E-25	1.09	3.53E-19	1.80	8.18E-47
Rv3921c	membrane protein insertase YidC(Rv3921c)	1.32	1.08E-95	1.48	1.68E-122	2.10	6.39E-248	1.31	1.40E-52	1.05	2.55E-33	1.83	3.19E-93

BDQ+		0.5 h Rv0678 A99V vs. WT		0.5 h Rv0678 inversion vs. WT		0.5 h Rv0678 274insA vs. WT		4 h Rv0678 A99V vs. WT		4 h Rv0678 inversion vs. WT		4 h Rv0678 274insA vs. WT	
Rv3922c	membrane protein insertion efficiency factor(Rv3922c)	1.13	1.65E-38	1.28	3.03E-50	2.00	2.50E-123	1.14	5.58E-33	0.95	2.41E-22	1.61	1.18E-60
Rv3923c	ribonuclease P protein component(rnpA)	1.04	1.03E-30	0.88	2.41E-22	1.84	2.05E-95	1.01	1.59E-25	0.73	2.11E-13	1.36	2.63E-41
Rv3924c	50S ribosomal protein L34(rpmH)	0.45	5.72E-11	0.37	4.40E-08	1.46	7.28E-112	0.81	6.60E-13	0.43	2.83E-04	0.96	2.34E-16
RVnc0001	ncRNA(AS1890)	-2.84	1.51E-09	-2.37	2.44E-09	-2.11	5.92E-08	-1.65	3.66E-07	-1.93	7.34E-08	-1.54	2.15E-06
RVnc0004	ncRNA(B11)	1.95	7.87E-07	2.28	2.15E-09	2.66	1.17E-12	0.09	8.18E-01	0.59	8.51E-02	1.02	9.26E-04
RVnc0006	ncRNA(C8)	0.56	4.04E-01	1.18	5.74E-02	1.30	3.34E-02	-0.74	3.91E-01	1.65	3.33E-02	-1.46	8.76E-02
RVnc0008	ncRNA(F6)	-1.24	1.72E-34	-0.90	1.60E-20	-1.18	1.48E-32	0.46	1.45E-02	0.47	1.64E-02	0.80	1.05E-05
RVnc0013	ncRNA(mcr11)	-2.03	8.33E-05	-1.20	2.27E-02	-1.63	1.44E-03	0.12	8.41E-01	0.58	2.61E-01	-0.19	7.37E-01
RVnc0015	ncRNA(mcr15)	-1.48	7.10E-11	-1.32	2.75E-09	-0.93	2.24E-05	-0.14	5.96E-01	0.17	5.16E-01	-0.45	6.53E-02
RVnc0021	ncRNA(mcr3)	1.26	3.50E-06	1.19	9.97E-06	1.23	4.04E-06	-0.09	7.93E-01	0.54	7.93E-02	0.50	1.09E-01
RVnc0024	ncRNA(mcr7)	1.46	1.19E-17	0.76	1.24E-05	1.55	4.94E-20	-0.46	1.43E-01	-0.57	6.93E-02	-0.39	2.30E-01
RVnc0034	ncRNA(MTS0858)	0.33	5.99E-01	-0.35	5.69E-01	0.15	8.03E-01	-1.32	5.47E-02	-1.21	9.35E-02	-2.44	1.74E-03
RVnc0036	ncRNA(MTS1338)	-0.26	4.68E-01	1.00	1.51E-03	-0.53	1.10E-01	-1.50	3.47E-12	0.13	6.28E-01	-1.44	1.57E-10
RVnc0040	ncRNA(MTS2975)	-0.58	1.28E-02	-0.05	8.24E-01	-0.09	7.10E-01	-0.52	4.58E-02	-1.04	2.13E-04	-0.55	3.25E-02
RVnc0046	miscRNA(ssr)	1.97	1.76E-13	-0.37	1.96E-01	1.38	2.40E-07	-1.66	1.55E-05	0.15	7.63E-01	-1.80	7.02E-06
Rvnt01	tRNA-Ile(ileT)	0.75	5.22E-03	0.19	5.22E-01	1.26	8.79E-07	0.05	8.63E-01	0.22	3.81E-01	0.04	8.72E-01
Rvnt03	tRNA-Leu(leuT)	0.08	9.20E-01	-0.31	6.88E-01	1.47	1.76E-02	-1.37	7.32E-02	0.89	1.93E-01	-2.19	8.95E-03
Rvnt05	tRNA-Tyr(tyrT)	0.40	4.91E-01	1.24	1.31E-02	0.94	6.37E-02	-0.27	6.82E-01	0.34	5.96E-01	-0.20	7.56E-01
Rvnt06	tRNA-Thr(thrT)	-0.41	1.79E-01	-0.14	6.51E-01	0.43	1.20E-01	1.29	5.40E-03	-0.28	6.61E-01	0.60	2.45E-01
Rvnt07	tRNA-Met(metT)	-1.93	4.98E-12	-1.01	2.26E-04	-0.76	5.20E-03	0.88	3.22E-02	0.58	1.93E-01	1.18	4.26E-03
Rvnt11	tRNA-Glu(gluT)	1.09	3.36E-07	-0.30	2.12E-01	0.96	5.24E-06	-0.30	3.72E-01	-0.51	1.26E-01	-0.71	2.75E-02
Rvnt12	tRNA-Asp(aspT)	0.84	1.61E-01	-0.24	7.21E-01	1.97	2.36E-04	-0.30	6.67E-01	0.09	9.03E-01	-0.97	1.41E-01
Rvnt13	tRNA-Phe(pheU)	0.35	5.05E-02	0.40	2.37E-02	1.01	9.82E-10	-0.09	6.85E-01	-0.36	6.49E-02	-0.09	6.81E-01
Rvnt15	tRNA-Ala(alaV)	-1.52	4.52E-07	-0.98	7.32E-04	-1.51	3.25E-07	0.18	7.97E-01	0.13	8.57E-01	-1.17	5.99E-02
Rvnt19	tRNA-Leu(leuW)	0.82	1.03E-01	1.19	1.07E-02	1.62	3.02E-04	-1.06	2.80E-02	-0.26	6.32E-01	0.08	8.82E-01
Rvnt21	tRNA-Pro(proT)	1.09	3.76E-02	0.80	1.32E-01	0.91	7.93E-02	-0.19	8.04E-01	0.15	8.58E-01	0.19	7.91E-01
Rvnt27	tRNA-Gly(glyV)	1.23	1.91E-02	0.86	1.10E-01	2.41	5.83E-07	0.05	9.40E-01	0.66	2.77E-01	-0.15	8.26E-01
Rvnt28	tRNA-Arg(argW)	-1.19	8.35E-03	-0.43	3.16E-01	-1.30	3.50E-03	-0.62	2.17E-01	-0.72	1.74E-01	-0.75	1.38E-01
Rvnt29	tRNA-His(hisT)	-0.23	6.58E-01	1.02	1.24E-02	0.78	5.97E-02	0.15	7.65E-01	-0.25	6.48E-01	-0.07	8.95E-01
Rvnt32	tRNA-Gly(glyT)	-0.15	7.47E-01	-0.43	3.17E-01	0.29	4.84E-01	-0.87	3.60E-02	-1.19	8.54E-03	-1.34	1.86E-03
Rvnt33	tRNA-Cys(cysU)	-2.01	9.61E-05	-1.51	2.61E-03	-0.46	3.68E-01	0.73	1.63E-01	0.18	7.81E-01	-0.45	4.40E-01
Rvnt34	tRNA-Val(valU)	2.18	1.21E-01	1.49	3.14E-01	3.01	1.98E-02	-2.04	4.76E-02	-1.02	3.04E-01	-2.70	1.88E-02
Rvnt35	tRNA-Glu(gluU)	0.62	4.11E-01	-1.34	1.05E-01	1.86	4.56E-03	-0.69	3.83E-01	-1.23	1.39E-01	-2.56	4.45E-03

BDQ+		0.5 h <i>Rv0678</i> A99V vs. WT		0.5 h <i>Rv0678</i> inver- sion vs. WT		0.5 h <i>Rv0678</i> 274insA vs. WT		4 h <i>Rv0678</i> A99V vs. WT		4 h <i>Rv0678</i> inversion vs. WT		4 h <i>Rv0678</i> 274insA vs. WT	
Rvnt36	tRNA-Gln(glnU)	2.14	6.20E-04	2.33	1.31E-04	3.29	1.97E-08	-1.19	5.14E-03	-0.14	7.85E-01	-1.54	4.99E-04
Rvnt38	tRNA-Met(metU)	0.21	1.72E-01	-0.20	2.02E-01	0.00	9.90E-01	1.55	1.31E-09	0.72	9.55E-03	0.98	3.32E-04
Rvnt39	tRNA-Thr(thrU)	1.25	7.31E-02	1.31	5.22E-02	1.55	1.87E-02	-0.40	5.30E-01	-0.40	5.56E-01	0.32	5.91E-01
Rvnt41	tRNA-Ser(serV)	-1.00	5.50E-04	-0.57	5.41E-02	-0.53	7.01E-02	0.87	3.96E-02	0.95	2.64E-02	0.04	9.41E-01
Rvnt43	tRNA-Arg(argU)	-0.09	9.00E-01	-0.40	5.27E-01	1.26	1.43E-02	0.95	1.99E-01	0.32	7.25E-01	0.36	6.60E-01
Rvnt45	tRNA-Ser(serU)	-0.79	1.74E-02	-0.33	2.98E-01	-0.46	1.50E-01	0.87	6.44E-02	1.03	3.12E-02	1.01	2.75E-02

Table 0.9: Supplementary data to Figure 3.11 DEGs up- and downregulated after 4 h BDQ exposure shared between all *Rv0678* variants. The significant DEGs of each mutant compared to the ancestral wild-type under BDQ exposure were assessed, then sorted in up- and downregulated genes, using a Venn diagram the commonly shared up- and downregulated DEGs between all *Rv0678* variants were assessed. These 60 upregulated and 16 downregulated DEGs were combined in the following table, using DAVID (david.ncicrf.gov) for gene name determination and mycobrowser.epfl.ch/, for the functional category. Wt = ancestral wild-type, Log₂FC = Log₂ Fold Change, *padj* = adjusted p-value.

BDQ+ 4h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Locus	DAVID ID	Gene name	Category	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>
Rv0123	886887	hypothetical protein(Rv0123)	conserved hypotheticals	-1.15	1.69E-17	-1.04	2.90E-14	-1.06	8.53E-14
Rv0164	886267	hypothetical protein(TB18.5)	conserved hypotheticals	1.55	2.24E-45	1.32	6.69E-32	2.27	4.40E-92
Rv0236A	3205106	hypothetical protein(Rv0236A)	cell wall and cell processes	1.54	5.90E-05	2.01	9.31E-08	1.95	1.30E-07
Rv0237	886693	lipoprotein LpqI(lpqI)	cell wall and cell processes	1.54	1.21E-20	1.55	2.24E-20	2.08	1.64E-34
Rv0341	886518	isoniazid inducible protein IniB(iniB)	cell wall and cell processes	-1.16	7.57E-30	-2.36	3.58E-124	-1.48	2.61E-43
Rv0359	886482	zinc metalloprotease(Rv0359)	cell wall and cell processes	1.14	5.89E-06	1.43	1.52E-08	1.64	2.11E-11
Rv0364	886473	transmembrane protein(Rv0364)	cell wall and cell processes	1.12	4.50E-16	1.09	9.02E-15	1.34	3.31E-21
Rv0379	886449	protein translocase subunit SecE(secE2)	cell wall and cell processes	2.40	2.01E-08	1.85	4.20E-05	3.30	7.26E-15
Rv0446c	886332	transmembrane protein(Rv0446c)	cell wall and cell processes	1.34	7.02E-24	2.76	5.61E-103	1.55	8.50E-30
Rv0447c	886330	cyclopropane-fatty-acyl-phospholipid synthase UfaA(ufaA1)	lipid metabolism	1.45	1.12E-20	3.19	2.18E-101	1.76	4.27E-28
Rv0448c	886328	hypothetical protein(Rv0448c)	conserved hypotheticals	1.60	8.35E-07	3.91	1.61E-37	2.15	4.18E-11
Rv0449c	886326	hypothetical protein(Rv0449c)	conserved hypotheticals	1.18	3.55E-13	2.62	2.10E-62	1.40	1.68E-16
Rv0516c	887324	anti-anti-sigma factor(Rv0516c)	information pathways	1.30	5.15E-16	1.49	1.47E-20	2.32	1.19E-44
Rv0705	888356	30S ribosomal protein S19(rpsS)	information pathways	1.31	7.34E-28	1.11	1.14E-19	2.06	1.32E-62
Rv0709	888374	50S ribosomal protein L29(rpmC)	information pathways	1.44	1.47E-42	1.04	5.78E-22	2.25	4.15E-94
Rv0846c	885207	oxidase(Rv0846c)	intermediary metabolism and respiration	-1.43	8.31E-24	-1.71	2.17E-32	-2.35	2.90E-54
Rv0847	885051	lipoprotein LpqS(lpqS)	lipid metabolism	-1.28	2.90E-20	-1.39	2.29E-23	-2.10	3.04E-47
Rv0848	885545	cysteine synthase CysK(cysK2)	intermediary metabolism and respiration	-1.18	7.01E-14	-1.42	1.94E-19	-1.75	1.67E-26
Rv0849	885111	MFS-type transporter(Rv0849)	cell wall and cell processes	-1.34	5.63E-13	-1.70	3.26E-20	-1.92	4.22E-23
Rv0867c	885749	resuscitation-promoting factor RpfA(rpfA)	cell wall and cell processes	1.20	5.17E-28	1.27	7.02E-31	1.75	2.47E-55
Rv1099c	885861	fructose 1,6-bisphosphatase(glpX)	intermediary metabolism and respiration	1.00	5.24E-24	1.00	2.81E-23	1.32	1.98E-39

BDQ+ 4h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv1130	885843	2-methylcitrate dehydratase(prpD)	intermediary metabolism and respiration	1.49	1.03E-09	2.81	1.70E-33	2.17	1.53E-19
Rv1131	888949	methylcitrate synthase PrpC(prpC)	intermediary metabolism and respiration	1.97	1.66E-12	2.46	7.63E-19	2.48	6.00E-19
Rv1195	886044	PE family protein PE13(PE13)	PE / PPE	1.87	1.12E-68	1.57	1.10E-48	2.59	9.60E-118
Rv1196	886073	PPE family protein PPE18(PPE18)	PE / PPE	2.22	3.05E-48	1.94	5.50E-37	3.32	1.52E-95
Rv1197	886051	ESAT-6 like protein EsxK(esxK)	cell wall and cell processes	2.05	7.32E-60	1.50	5.01E-32	3.39	7.34E-145
Rv1198	886090	ESAT-6 like protein EsxL(esxL)	cell wall and cell processes	1.94	5.69E-94	1.40	3.99E-49	3.35	2.69E-250
Rv1242	887095	ribonuclease VapC33(vapC33)	virulence, detoxification, adaptation	1.24	4.15E-06	1.17	3.19E-05	1.42	7.42E-08
Rv1344	886848	acyl carrier protein MbtL(mbtL)	lipid metabolism	1.56	8.37E-13	1.36	1.90E-09	2.40	8.40E-30
Rv1473A	3205054	transcriptional regulator(Rv1473A)	regulatory proteins	2.01	1.56E-03	2.15	8.82E-04	1.92	2.79E-03
Rv1534	886420	transcriptional regulator(Rv1534)	regulatory proteins	1.17	1.06E-16	1.00	1.04E-11	1.60	8.93E-31
Rv1613	885291	tryptophan synthase subunit alpha(trpA)	intermediary metabolism and respiration	1.27	8.88E-20	1.13	8.70E-16	2.00	1.42E-43
Rv1685c	885682	hypothetical protein(Rv1685c)	conserved hypotheticals	-2.46	7.80E-42	-2.51	3.02E-43	-2.64	1.22E-42
Rv1686c	885672	ABC transporter permease(Rv1686c)	cell wall and cell processes	-2.48	8.86E-125	-2.56	1.34E-131	-2.67	1.68E-126
Rv1687c	885678	ABC transporter ATP-binding protein(Rv1687c)	cell wall and cell processes	-2.40	1.24E-72	-2.72	1.73E-93	-2.65	1.38E-78
Rv1688	885679	3-methyladenine DNA glycosylase(mpg)	information pathways	-1.11	1.44E-09	-1.24	1.37E-10	-1.06	2.07E-08
Rv1955	885966	toxin HigB(higB)	virulence, detoxification, adaptation	-1.08	1.08E-12	-1.20	3.12E-15	-1.32	1.46E-16
Rv1956	885964	antitoxin HigA(higA)	virulence, detoxification, adaptation	-1.31	1.46E-12	-1.05	2.31E-08	-1.25	1.21E-10
Rv2257c	888069	hypothetical protein(Rv2257c)	conserved hypotheticals	1.28	4.01E-14	1.26	5.30E-13	2.14	5.64E-38
Rv2258c	888755	transcriptional regulator(Rv2258c)	regulatory proteins	1.55	3.41E-31	1.41	1.45E-25	2.30	9.18E-63
Rv2275	888355	cyclo(L-tyrosyl-L-tyrosyl) synthase(Rv2275)	conserved hypotheticals	1.41	3.31E-15	1.21	7.92E-11	1.81	3.06E-24
Rv2276	888373	cytochrome P450 Cyp121(cyp121)	intermediary metabolism and respiration	1.94	4.36E-25	1.80	6.95E-21	2.69	7.44E-48
Rv2311	885168	hypothetical protein(Rv2311)	conserved hypotheticals	1.12	2.20E-02	1.14	2.42E-02	1.17	1.35E-02
Rv2329c	888116	nitrate / nitrite transporter(narK1)	cell wall and cell processes	1.40	1.60E-39	1.08	6.29E-23	1.90	1.17E-65
Rv2662	888589	hypothetical protein(Rv2662)	conserved hypotheticals	-1.19	4.72E-08	-1.27	1.99E-08	-1.33	3.57E-09
Rv2806	888905	membrane protein(Rv2806)	cell wall and cell processes	1.57	1.42E-02	1.64	1.21E-02	1.62	9.44E-03

BDQ+ 4h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated			A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv2874	888162	integral membrane C-type cytochrome biogenesis protein DipZ(dipZ)	1.39	1.81E-11	3.01	7.88E-52	1.49	3.65E-12
Rv2875	887724	major secreted immunogenic protein Mpt70(mpt70)	1.86	2.91E-21	3.70	2.42E-83	2.06	2.06E-23
Rv2913c	887809	D-amino acid aminohydrolase(Rv2913c)	-1.01	2.21E-25	-1.06	3.79E-27	-1.22	8.57E-33
Rv2949c	887206	chorismate pyruvate-lyase(Rv2949c)	1.80	5.22E-50	1.18	2.29E-21	2.73	9.47E-104
Rv2950c	887202	long-chain-fatty-acid--AMP ligase FadD29(fadD29)	1.50	2.62E-76	1.05	9.11E-37	2.23	4.84E-150
Rv2952	887311	phthiotriol / phenolphthiotriol dimycoserates methyltransferase(Rv2952)	1.66	5.17E-28	1.47	2.25E-21	2.46	1.99E-58
Rv2953	887772	trans-acting enoyl reductase(Rv2953)	1.52	3.84E-41	1.31	1.43E-29	2.11	1.95E-76
Rv2987c	888225	3-isopropylmalate dehydratase small subunit(leuD)	1.35	4.07E-16	1.41	6.56E-17	2.54	2.34E-53
Rv2988c	887875	3-isopropylmalate dehydratase large subunit(leuC)	1.12	1.48E-22	1.33	3.06E-31	2.05	5.64E-69
Rv2989	887723	transcriptional regulator(Rv2989)	1.27	5.55E-15	1.22	3.08E-13	1.79	1.97E-28
Rv2993c	887867	2-hydroxyhepta-2,4-diene-1,7-dioate isomerase(Rv2993c)	1.19	5.44E-20	1.29	9.76E-23	1.54	5.59E-33
Rv3012c	887335	glutamyl-tRNA(GLN) amidotransferase subunit C(gatC)	1.20	1.78E-03	1.56	4.27E-05	1.98	6.16E-08
Rv3198A	3205084	glutaredoxin protein(Rv3198A)	1.60	5.23E-12	1.05	1.83E-05	2.04	5.60E-18
Rv3218	888906	hypothetical protein(Rv3218)	1.20	1.05E-04	1.22	1.40E-04	1.78	1.36E-09
Rv3271c	888716	integral membrane protein(Rv3271c)	1.30	3.31E-10	1.26	1.81E-09	1.73	8.35E-16
Rv3312b	19394686	hypothetical protein(Rv3312b)	1.66	2.77E-05	1.57	1.42E-04	2.50	1.66E-11
Rv3351c	888109	hypothetical protein(Rv3351c)	1.36	2.15E-03	1.13	1.66E-02	1.58	2.03E-04
Rv3366	887648	tRNA / rRNA methylase SpoU(spoU)	1.63	8.20E-04	1.57	1.99E-03	2.55	1.54E-08
Rv3371	888053	diacylglycerol O-acyltransferase(Rv3371)	-1.05	3.18E-13	-1.40	4.17E-21	-1.38	9.48E-20
Rv3395A	3205044	membrane protein(Rv3395A)	2.95	1.41E-02	3.42	4.29E-03	3.70	1.07E-03
Rv3487c	888430	carboxylesterase LipF(lipF)	1.45	4.22E-40	1.24	1.07E-28	1.99	1.22E-68
Rv3622c	885712	PE family protein PE32(PE32)	1.44	8.14E-04	1.01	3.19E-02	1.40	1.14E-03
Rv3678A	3205049	hypothetical protein(Rv3678A)	1.04	2.15E-05	1.06	2.00E-05	1.40	2.37E-08
Rv3686c	885306	hypothetical protein(Rv3686c)	1.12	4.05E-24	1.02	1.83E-19	1.55	1.34E-43

BDQ+ 4h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv3714c	885456	hypothetical protein(Rv3714c)	conserved hypotheticals	1.25	5.41E-17	1.21	2.95E-15	1.28	6.93E-17
Rv3794	886123	arabinosyltransferase A(embA)	cell wall and cell processes	1.33	2.86E-24	1.50	2.88E-30	2.08	2.31E-53
Rv3796	886128	hypothetical protein(Rv3796)	conserved hypotheticals	1.51	4.77E-05	1.30	8.93E-04	1.81	6.40E-07
Rv3920c	886255	hypothetical protein(Rv3920c)	conserved hypotheticals	1.26	1.02E-25	1.09	3.53E-19	1.80	8.18E-47
Rv3921c	886238	membrane protein insertase YidC(Rv3921c)	cell wall and cell processes	1.31	1.40E-52	1.05	2.55E-33	1.83	3.19E-93
RVnc0001	14515861	ncRNA(AS1890)	conserved hypotheticals	-1.65	3.66E-07	-1.93	7.34E-08	-1.54	2.15E-06

Table 0.10: Supplementary data to Figure 3.12 DEGs up- and downregulated after 0.5 h BDQ exposure shared between all *Rv0678* variants. The significant DEGs of each mutant compared to the ancestral wild-type under BDQ exposure were assessed, then sorted in up- and downregulated genes, using a Venn diagram the commonly shared up- and downregulated DEGs between all *Rv0678* variants were assessed. These 77 upregulated and 86 downregulated DEGs were combined in the following table, using DAVID (david.ncicrf.gov) for gene name determination and mycobrowser.epfl.ch/, for the functional category. Wt = ancestral wild-type, Log₂FC = Log₂ Fold Change, *padj* = adjusted p-value.

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Locus	DAVID ID	Gene name	Category	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>	Log ₂ FC	<i>padj</i>
Rv0078B	14515845	hypothetical protein(Rv0078B)	conserved hypotheticals	-2.53	1.70E-27	-1.37	4.81E-09	-2.34	3.85E-24
Rv0108c	886918	hypothetical protein(Rv0108c)	conserved hypotheticals	-1.26	1.00E-28	-1.35	3.78E-33	-1.20	1.10E-26
Rv0186A	14515880	metallothionein(mymT)	virulence, detoxification, adaptation	-2.33	3.91E-50	-2.51	4.89E-58	-2.96	9.97E-81
Rv0236c	886707	alpha-(1->3)-arabinofuranosyltransferase(aftD)	cell wall and cell processes	1.31	6.10E-50	1.71	1.33E-86	1.68	7.78E-84
Rv0313	886572	hypothetical protein(Rv0313)	conserved hypotheticals	-1.40	1.71E-19	-1.04	2.28E-11	-1.38	2.23E-19
Rv0316	886560	muconolactone isomerase(Rv0316)	intermediary metabolism and respiration	1.14	2.77E-09	1.67	1.79E-20	1.97	1.00E-28
Rv0340	886514	hypothetical protein(Rv0340)	conserved hypotheticals	-1.20	2.26E-24	-1.05	3.44E-19	-1.65	7.37E-45
Rv0362	886476	Mg ²⁺ transport transmembrane protein MgtE(mgtE)	cell wall and cell processes	1.20	4.12E-12	1.27	4.08E-14	1.75	1.29E-26
Rv0397A	14515846	hypothetical protein(Rv0397A)	conserved hypotheticals	-1.18	4.64E-32	-1.27	7.54E-38	-1.43	5.17E-47
Rv0429c	886366	polypeptide deformylase(def)	information pathways	1.33	7.89E-21	1.40	1.03E-23	1.88	5.62E-43
Rv0447c	886330	cyclopropane-fatty-acyl-phospholipid synthase UfaA(ufaA1)	lipid metabolism	1.80	6.93E-43	2.98	1.84E-121	2.11	5.37E-60
Rv0448c	886328	hypothetical protein(Rv0448c)	cell wall and cell processes	2.31	1.41E-16	3.53	1.57E-39	2.13	2.11E-14
Rv0449c	886326	hypothetical protein(Rv0449c)	conserved hypotheticals	1.56	1.51E-54	2.40	5.57E-134	1.62	1.74E-60
Rv0516c	887324	anti-anti-sigma factor(Rv0516c)	information pathways	1.44	3.65E-21	1.26	1.57E-16	2.49	3.51E-62
Rv0542c	887507	2-succinylbenzoic acid--CoA ligase(menE)	intermediary metabolism and respiration	1.22	2.15E-46	1.27	1.74E-51	1.56	8.27E-79
Rv0544c	887511	transmembrane protein(Rv0544c)	cell wall and cell processes	1.24	3.01E-10	1.09	2.38E-08	1.64	2.98E-18
Rv0572c	887696	hypothetical protein(Rv0572c)	conserved hypotheticals	-2.13	1.02E-81	-1.74	1.69E-61	-2.32	3.61E-95
Rv0575c	887720	oxidoreductase(Rv0575c)	intermediary metabolism and respiration	-1.37	9.52E-10	-1.30	4.37E-09	-2.19	4.31E-21
Rv0608	887895	antitoxin VapB28(vapB28)	virulence, detoxification, adaptation	-1.23	4.22E-44	-1.17	9.59E-41	-1.46	2.28E-61

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv0614	887914	hypothetical protein(Rv0614)	conserved hypotheticals	1.25	3.29E-02	1.62	3.50E-03	1.85	6.34E-04
Rv0647c	888070	hypothetical protein(Rv0647c)	conserved hypotheticals	1.01	2.00E-28	1.44	4.18E-58	1.58	1.68E-70
Rv0662c	888117	antitoxin VapB7(vapB7)	virulence, detoxification, adaptation	1.11	1.75E-02	1.72	7.38E-05	1.22	6.78E-03
Rv0771	888872	4-carboxymuconolactone decarboxylase(Rv0771)	intermediary metabolism and respiration	-1.21	2.95E-07	-1.01	6.71E-06	-1.26	5.45E-08
Rv0787	885468	hypothetical protein(Rv0787)	conserved hypotheticals	1.12	1.07E-05	1.40	1.22E-08	1.84	1.49E-14
Rv0827c	885375	HTH-type transcriptional regulator KmtR(kmtR)	regulatory proteins	-1.80	8.74E-37	-1.03	4.06E-13	-2.66	2.16E-76
Rv0841	3205068	transmembrane protein(Rv0841)	cell wall and cell processes	2.47	1.83E-02	2.33	2.49E-02	2.10	4.47E-02
Rv0846c	885207	oxidase(Rv0846c)	intermediary metabolism and respiration	-1.12	7.55E-22	-1.45	1.04E-35	-1.86	7.51E-57
Rv0847	885051	lipoprotein LpqS(lpqS)	lipid metabolism	-1.03	4.62E-19	-1.37	1.14E-32	-1.77	6.63E-54
Rv0848	885545	cysteine synthase CysK(cysK2)	intermediary metabolism and respiration	-1.38	5.45E-37	-1.59	7.74E-49	-2.34	4.07E-104
Rv0849	885111	MFS-type transporter(Rv0849)	cell wall and cell processes	-1.16	3.83E-20	-1.59	1.66E-36	-2.14	5.02E-65
Rv0856	885783	hypothetical protein(Rv0856)	conserved hypotheticals	-1.40	3.25E-39	-1.27	2.05E-33	-1.29	3.41E-34
Rv0867c	885749	resuscitation-promoting factor RpfA(rpfA)	cell wall and cell processes	1.46	6.57E-53	1.40	1.31E-49	2.75	7.12E-197
Rv0882	885248	transmembrane protein(Rv0882)	cell wall and cell processes	1.11	2.81E-04	1.08	3.17E-04	1.90	1.94E-11
Rv0941c	885914	hypothetical protein(Rv0941c)	conserved hypotheticals	-1.06	4.15E-39	-1.08	1.96E-41	-1.25	2.38E-54
Rv0967	885312	copper-sensing transcriptional repressor CsoR(csoR)	regulatory proteins	-1.99	1.95E-86	-1.44	2.61E-46	-2.70	2.91E-157
Rv0968	885052	hypothetical protein(Rv0968)	conserved hypotheticals	-2.14	8.28E-50	-1.74	1.47E-33	-2.80	2.91E-85
Rv0969	885254	copper-exporting ATPase(ctpV)	cell wall and cell processes	-1.35	1.84E-45	-1.14	5.04E-33	-2.07	1.52E-106
Rv0979A	3205057	50S ribosomal protein L32(rpmF)	information pathways	1.21	2.50E-27	1.00	2.14E-19	2.16	2.00E-90
Rv0979c	885063	hypothetical protein(Rv0979c)	conserved hypotheticals	-1.34	7.71E-04	-1.43	2.31E-04	-1.10	3.94E-03
Rv1033c	887957	two component transcriptional regulator TrcR(trcR)	regulatory proteins	-1.39	1.44E-23	-1.96	7.94E-44	-1.54	1.00E-28
Rv1038c	888372	ESAT-6 like protein EsxJ(esxJ)	cell wall and cell processes	-1.44	2.98E-15	-1.09	1.32E-09	-1.29	6.37E-13
Rv1081c	887102	membrane protein(Rv1081c)	conserved hypotheticals	1.65	6.30E-12	1.67	1.59E-12	2.12	4.00E-20
Rv1097c	885962	hypothetical protein(Rv1097c)	cell wall and cell processes	1.19	3.17E-25	1.42	3.28E-37	1.77	2.79E-58
Rv1130	885843	2-methylcitrate dehydratase(prpD)	intermediary metabolism and respiration	1.22	3.93E-10	3.72	1.10E-96	1.92	5.12E-25

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv1131	888949	methylocitrate synthase PrpC(prpC)	intermediary metabolism and respiration	1.72	1.28E-10	4.17	1.22E-62	2.71	2.26E-26
Rv1155a	19394672	hypothetical protein(Rv1155a)		-1.67	2.98E-25	-1.09	9.00E-12	-1.74	1.50E-27
Rv1159A	3205109	4a-hydroxytetrahydrobiopterin dehydratase(Rv1159A)		-1.78	4.30E-48	-1.40	1.86E-30	-2.55	6.48E-98
Rv1176c	886080	hypothetical protein(Rv1176c)	conserved hypotheticals	1.25	8.19E-06	1.36	6.31E-07	2.18	5.63E-17
Rv1184c	886063	hypothetical protein(Rv1184c)	cell wall and cell processes	1.36	4.65E-63	1.33	7.89E-62	2.18	1.22E-170
Rv1244	887093	lipoprotein LpqZ(lpqZ)	cell wall and cell processes	1.21	1.46E-19	1.10	1.05E-16	1.82	8.77E-46
Rv1284	886987	beta-carbonic anhydrase(canA)	intermediary metabolism and respiration	-1.32	7.40E-25	-1.85	6.15E-47	-1.79	9.78E-44
Rv1347c	886846	lysine N-acetyltransferase MbtK(mbtK)	lipid metabolism	1.32	1.35E-11	1.25	1.11E-10	2.49	8.16E-43
Rv1348	886853	iron ABC transporter ATP-binding protein / permease IrtA(irtA)	cell wall and cell processes	1.26	5.27E-46	1.64	1.85E-79	2.21	2.64E-146
Rv1353c	886827	HTH-type transcriptional regulator(Rv1353c)	regulatory proteins	1.03	3.73E-04	1.49	4.68E-08	1.29	3.38E-06
Rv1517	886467	hypothetical protein(Rv1517)	cell wall and cell processes	-1.41	2.92E-26	-1.23	1.52E-21	-1.36	1.43E-25
Rv1519	886453	hypothetical protein(Rv1519)	conserved hypotheticals	1.33	1.63E-14	1.33	6.69E-15	2.50	6.00E-53
Rv1552	886376	fumarate reductase flavoprotein subunit(frdA)	intermediary metabolism and respiration	-1.53	1.94E-21	-1.06	2.63E-12	-1.47	1.17E-20
Rv1632c	885649	hypothetical protein(Rv1632c)	conserved hypotheticals	1.39	1.78E-06	1.69	1.31E-09	2.02	1.75E-13
Rv1686c	885672	ABC transporter permease(Rv1686c)	cell wall and cell processes	-1.09	1.00E-28	-2.63	3.89E-157	-1.65	3.73E-64
Rv1687c	885678	ABC transporter ATP-binding protein(Rv1687c)	cell wall and cell processes	-1.59	2.07E-71	-2.85	1.51E-224	-2.02	6.02E-115
Rv1733c	885214	transmembrane protein(Rv1733c)	cell wall and cell processes	-2.71	1.00E-100	-1.02	7.25E-18	-3.43	4.58E-145
Rv1735a	19394674	hypothetical protein(Rv1735a)		-2.03	5.42E-13	-1.10	3.02E-05	-2.40	7.04E-17
Rv1738	885215	hypothetical protein(Rv1738)	conserved hypotheticals	-3.63	2.42E-66	-1.66	6.20E-15	-5.14	3.73E-131
Rv1788	885895	PE family protein PE18(PE18)	PE / PPE	1.11	1.58E-02	1.63	1.29E-04	1.92	3.87E-06
Rv1813c	885546	hypothetical protein(Rv1813c)	conserved hypotheticals	-3.18	3.83E-223	-1.54	5.59E-54	-3.99	0.00E+00
Rv1851	885532	urease accessory protein UreF(ureF)	intermediary metabolism and respiration	1.08	4.06E-07	1.60	1.14E-15	1.58	4.11E-15
Rv1894c	885081	hypothetical protein(Rv1894c)	conserved hypotheticals	-1.01	5.05E-40	-1.45	4.21E-81	-1.81	1.36E-125
Rv1936	885882	monooxygenase(Rv1936)	intermediary metabolism and respiration	-1.02	1.39E-22	-1.22	1.93E-32	-1.18	5.82E-30
Rv1982A	11117810	antitoxin VapB36(vapB36)	virulence, detoxification, adaptation	-1.32	7.94E-32	-1.43	1.23E-37	-2.08	2.68E-77

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv1990c	885422	transcriptional regulator(Rv1990c)	regulatory proteins virulence, detoxification, adapta- tio	-1.10	9.72E-36	-1.35	6.50E-53	-1.41	9.05E-57
Rv1991A	14515890	antitoxin MazE6(mazE6)		-1.36	6.06E-43	-1.19	3.33E-35	-2.08	5.74E-89
Rv1993c	885631	hypothetical protein(Rv1993c) HTH-type transcriptional regulator	conserved hypotheticals	-1.22	8.81E-20	-1.12	2.70E-17	-1.80	5.26E-41
Rv1994c	888889	CmtR(cmtR)	regulatory protein virulence, detoxification, adapta- tio	-1.31	4.28E-45	-1.33	8.70E-47	-1.95	4.16E-97
Rv2005c	888831	universal stress protein(Rv2005c)	intermediary metabolism and respiration	-1.44	2.49E-36	-1.19	1.49E-25	-1.68	4.57E-50
Rv2007c	888887	ferredoxin(fdxA)		-3.43	6.04E-99	-1.84	5.94E-30	-4.10	2.10E-140
Rv2030c	887536	hypothetical protein(Rv2030c)	conserved hypotheticals virulence, detoxification, adapta- tio	-2.01	7.92E-49	-1.16	3.69E-17	-2.30	7.94E-65
Rv2031c	887579	alpha-crystallin(hspX)		-3.42	4.10E-99	-1.62	1.47E-23	-3.85	4.76E-126
Rv2046	888308	lipoprotein LppI(lppI)	cell wall and cell processes	1.11	3.31E-04	1.24	3.34E-05	2.34	5.92E-17
Rv2058c	887801	50S ribosomal protein L28(rpmB2)	information pathways	2.47	7.10E-04	2.23	2.24E-03	3.56	2.43E-07
Rv2185c	887239	hypothetical protein(TB16.3)	conserved hypotheticals	-1.15	7.71E-24	-1.53	2.06E-41	-1.23	1.70E-27
Rv2237A	14515864	hypothetical protein(Rv2237A)	conserved hypotheticals	-1.95	3.25E-32	-1.41	1.32E-17	-1.55	4.06E-21
Rv2307B	3205074	hypothetical protein(Rv2307B) molybdopterin biosynthesis protein	conserved hypotheticals intermediary metabolism and respiration	-1.08	4.30E-12	-1.07	1.48E-12	-1.26	8.63E-16
Rv2338c	886018	MoeW(moeW)		-1.30	1.64E-69	-1.50	4.71E-93	-1.53	3.23E-95
Rv2348c	886006	hypothetical protein(Rv2348c)	conserved hypotheticals	-1.02	2.71E-26	-1.53	1.76E-57	-1.09	1.77E-30
Rv2352c	888959	PPE family protein PPE38(PPE38)	RPE / PPE	-1.56	1.41E-09	-1.00	1.19E-04	-1.23	1.55E-06
Rv2365c	885259	hypothetical protein(Rv2365c)	conserved hypotheticals	1.27	3.95E-15	1.29	9.12E-16	1.07	4.65E-11
Rv2377c	885968	hypothetical protein(Rv2377c)	lipid metabolism	-1.22	3.81E-19	-1.20	4.12E-19	-1.17	2.99E-18
Rv2383c	885838	phenyloxazoline synthase(mbtB)	lipid metabolism	1.11	3.35E-41	1.13	4.81E-43	1.29	1.63E-55
Rv2386c	885823	salicylate synthase(mbtI)	lipid metabolism	1.01	1.63E-23	1.27	2.57E-37	1.63	9.04E-62
Rv2450c	885760	resuscitation-promoting factor RpfE(rpfE)	cell wall and cell processes	1.62	7.27E-30	1.17	6.65E-16	2.85	1.28E-97
Rv2557	887865	hypothetical protein(Rv2557) glutamine ABC transporter per- mease(Rv2563)	conserved hypotheticals	-1.36	3.39E-68	-1.72	2.36E-108	-1.80	5.43E-118
Rv2563	887516	glutamine ABC transporter ATP-binding protein(glnQ)	cell wall and cell processes	1.24	5.41E-23	1.13	1.89E-19	2.11	2.32E-68
Rv2564	887898		cell wall and cell processes virulence, detoxification, adapta- tio	1.12	6.11E-16	1.44	8.40E-27	2.15	4.91E-61
Rv2624c	888939	universal stress protein(Rv2624c)		-2.77	1.61E-58	-1.07	3.93E-10	-3.30	4.33E-82

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv2625c	887692	zinc metalloprotease Rip3(Rv2625c)	cell wall and cell processes	-2.33	7.22E-37	-1.09	4.61E-09	-2.89	1.61E-56
Rv2632c	887231	hypothetical protein(Rv2632c)	conserved hypotheticals	-1.56	1.39E-68	-1.68	3.88E-80	-1.83	1.89E-93
Rv2641	888629	cadmium inducible protein CadI(cadI)	conserved hypotheticals	-2.44	1.77E-42	-2.46	1.30E-43	-3.73	1.84E-97
Rv2643	887674	arsenic-transport integral membrane protein ArsC(arsC)	cell wall and cell processes	-1.10	9.95E-24	-1.27	1.20E-31	-1.85	1.76E-63
Rv2646	887706	integrase(Rv2646)	insertion seqs and phages	1.62	1.45E-08	1.34	3.10E-06	1.85	2.25E-11
Rv2647	885870	hypothetical protein(Rv2647)	conserved hypotheticals	1.57	7.32E-04	1.07	2.53E-02	2.04	3.97E-06
Rv2658c	888562	prophage protein(Rv2658c)	insertion seqs and phages	-1.45	7.00E-28	-2.75	4.53E-90	-2.17	6.15E-59
Rv2659c	885098	prophage integrase(Rv2659c)	insertion seqs and phages	-1.17	2.00E-20	-1.90	1.23E-50	-1.69	5.92E-41
Rv2663	888561	hypothetical protein(Rv2663)	conserved hypotheticals	-1.55	8.97E-36	-1.66	1.36E-41	-1.88	2.29E-51
Rv2730	888323	hypothetical protein(Rv2730)	conserved hypotheticals	-2.35	9.51E-16	-1.18	3.16E-05	-1.94	8.80E-12
Rv2781c	888488	oxidoreductase(Rv2781c)	intermediary metabolism and respiration	1.10	3.00E-48	1.52	3.50E-96	1.40	2.00E-80
Rv2782c	888470	zinc protease(pepR)	intermediary metabolism and respiration	1.14	1.04E-41	1.18	1.29E-44	1.08	3.51E-38
Rv2816c	888520	CRISPR-associated endoribonuclease Cas2(Rv2816c)	conserved hypotheticals	-1.69	1.76E-14	-1.89	6.60E-18	-1.94	7.84E-19
Rv2830c	888537	antitoxin VapB22(vapB22)	virulence, detoxification, adaptation	-1.18	2.90E-27	-1.08	2.08E-23	-1.46	1.03E-40
Rv2873	887155	cell surface lipoprotein(mpt83)	cell wall and cell processes	1.88	1.27E-65	2.64	7.13E-131	1.54	9.67E-45
Rv2874	888162	integral membrane C-type cytochrome biogenesis protein DipZ(dipZ)	intermediary metabolism and respiration	2.09	7.42E-45	2.95	1.29E-92	2.08	4.25E-45
Rv2875	887724	major secreted immunogenic protein Mpt70(mpt70)	cell wall and cell processes	2.33	4.67E-53	3.48	9.42E-119	2.32	1.74E-53
Rv2882c	887464	ribosome recycling factor(frr)	information pathways	-1.31	1.71E-31	-1.17	2.23E-25	-1.22	9.00E-28
Rv2911	887189	penicillin-binding protein DacB2(dacB2)	cell wall and cell processes	1.18	3.73E-18	1.16	4.47E-18	1.74	1.30E-40
Rv2912c	888017	TetR family HTH-type transcriptional regulator(Rv2912c)	regulatory proteins	-1.76	3.24E-20	-1.37	4.54E-13	-1.81	1.05E-21
Rv2913c	887809	D-amino acid aminohydrolase(Rv2913c)	intermediary metabolism and respiration	-1.71	6.05E-49	-1.52	1.57E-39	-2.07	1.90E-71
Rv2952	887311	phthiotriol / phenolphthiotriol dimycoserosates methyltransferase(Rv2952)	intermediary metabolism and respiration	1.07	1.72E-22	1.38	5.51E-39	2.23	3.03E-105
Rv2955c	887318	hypothetical protein(Rv2955c)	conserved hypotheticals	1.07	1.87E-10	1.34	8.16E-17	2.01	8.93E-38
Rv2961	887316	transposase(Rv2961)	insertion seqs and phage	-1.17	2.09E-04	-1.85	9.99E-09	-1.69	1.39E-07

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv2962c	887892	PGL / p-HBAD biosynthesis rhamnosyl-transferase(Rv2962c)	intermediary metabolism and respiration	-1.77	5.90E-58	-1.66	4.87E-52	-1.87	1.66E-65
Rv2963	887263	integral membrane protein(Rv2963)	cell wall and cell processes	-1.62	2.60E-75	-1.76	7.83E-90	-2.52	1.62E-179
Rv2983	888161	2-phospho-L-lactate guanylyltransferase(Rv2983)	conserved hypotheticals	1.04	1.98E-04	1.13	3.00E-05	1.37	2.16E-07
Rv2987c	888225	3-isopropylmalate dehydratase small subunit(leuD)	intermediary metabolism and respiration	1.19	4.19E-18	1.62	1.13E-35	2.31	8.33E-75
Rv2989	887723	transcriptional regulator(Rv2989)	regulatory proteins	1.01	4.08E-12	1.36	2.62E-22	1.62	1.12E-31
Rv3012c	887335	glutamyl-tRNA(GLN) amidotransferase subunit C(gatC)	information pathways	1.65	5.93E-07	1.80	2.41E-08	2.54	3.09E-16
Rv3031	888543	1,4-alpha-glucan-branching protein(Rv3031)	conserved hypotheticals	1.01	3.28E-22	1.16	2.12E-29	1.11	3.10E-27
Rv3036c	887320	hypothetical protein(TB22.2)	cell wall and cell processes	1.52	8.88E-21	1.01	1.19E-09	1.97	6.93E-36
Rv3130c	888841	diacylglycerol O-acyltransferase(tgs1)	lipid metabolism	-3.14	2.15E-85	-1.04	1.85E-10	-3.79	5.53E-125
Rv3289c	888736	transmembrane protein(Rv3289c)	cell wall and cell processes	-1.46	9.43E-16	-2.72	5.84E-51	-2.59	8.63E-47
Rv3310	887988	acid phosphatase(sapM)	cell wall and cell processes	1.14	2.56E-14	1.33	1.58E-19	1.26	1.06E-17
Rv3312A	3205113	pilin(Rv3312A)	cell wall and cell processes	1.60	1.41E-06	2.12	2.01E-11	3.14	8.17E-25
Rv3351c	888109	hypothetical protein(Rv3351c)	conserved hypotheticals	1.02	2.28E-02	1.25	3.48E-03	1.12	9.32E-03
Rv3366	887648	tRNA / rRNA methylase SpoU(spoU)	information pathways	1.54	2.58E-04	1.96	9.82E-07	2.33	3.13E-09
Rv3418c	887583	chaperonin GroES(groES)	virulence, detoxification, adaptation	-1.11	5.88E-13	-2.29	6.96E-52	-1.16	2.54E-14
Rv3444c	887587	ESAT-6 like protein EsxT(esxT)	cell wall and cell processes	1.37	7.95E-03	1.16	2.50E-02	2.24	2.59E-06
Rv3462c	887325	translation initiation factor IF-1(InfA)	information pathways	-1.34	1.15E-33	-1.27	1.97E-30	-1.18	8.20E-27
Rv3503c	888311	ferredoxin FdxD(fdxD)	intermediary metabolism and respiration	-1.55	4.19E-20	-1.27	6.17E-14	-2.08	3.20E-35
Rv3528c	888297	hypothetical protein(Rv3528c)	conserved hypotheticals	-1.28	4.02E-35	-1.33	3.12E-38	-1.05	1.35E-24
Rv3548c	887452	short-chain type dehydrogenase / reductase(Rv3548c)	intermediary metabolism and respiration	-1.14	2.24E-37	-1.02	1.70E-30	-1.28	5.44E-47
Rv3577	888442	hypothetical protein(Rv3577)	conserved hypotheticals	1.04	1.98E-13	1.21	1.55E-18	1.51	1.00E-28
Rv3578	888329	arsenic transport integral membrane protein ArsB(arsB2)	cell wall and cell processes	1.05	4.87E-11	1.26	2.58E-16	1.36	5.81E-19
Rv3583c	887854	RNA polymerase-binding transcription factor CarD(Rv3583c)	regulatory proteins	-1.02	5.89E-40	-1.06	5.09E-43	-1.52	2.04E-87
Rv3589	886639	A / G-specific adenine glycosylase(mutY)	information pathways	1.20	3.88E-15	1.04	6.39E-12	1.67	2.62E-30

BDQ+ 0.5h shared significant DEGs of <i>Rv0678</i> mutants up and downregulated				A99V vs. Wt		Inversion vs. Wt		274insA vs. Wt	
Rv3597c	885580	iron-regulated H-NS-like protein(lsr2)	information pathways	-1.55	5.49E-37	-1.54	1.06E-36	-1.66	6.36E-43
Rv3614c	885777	ESX-1 secretion-associated protein EspD(espD)	cell wall and cell processes	-1.16	2.04E-38	-1.31	2.18E-48	-1.27	5.40E-46
Rv3616c	885377	ESX-1 secretion-associated protein EspA(espA)	cell wall and cell processes	-1.21	7.57E-46	-1.51	3.99E-71	-1.29	6.71E-53
Rv3675	885155	membrane protein(Rv3675)	cell wall and cell processes	-1.53	2.30E-59	-1.56	2.63E-63	-1.82	1.97E-82
Rv3706c	885222	hypothetical protein(Rv3706c)	conserved hypotheticals	1.84	1.58E-09	1.45	2.62E-06	2.08	3.83E-12
Rv3789	886109	GtrA family protein(Rv3789)	cell wall and cell processes	1.18	1.15E-06	1.43	9.76E-10	2.03	2.94E-19
Rv3794	886123	arabinosyltransferase A(embA)	cell wall and cell processes	1.14	8.14E-44	1.37	5.21E-64	1.67	3.48E-95
Rv3796	886128	hypothetical protein(Rv3796)	conserved hypotheticals	1.15	3.48E-04	1.67	2.90E-08	1.95	4.15E-11
Rv3797	886122	acyl-CoA dehydrogenase FadE35(fadE35)	lipid metabolism	1.17	9.15E-14	1.32	8.74E-18	1.60	3.15E-26
Rv3840	886167	transcriptional regulator(Rv3840)	regulatory proteins intermediary metabolism and respiration	1.08	1.01E-07	1.10	3.21E-08	1.40	8.44E-13
Rv3841	886176	bacterioferritin BfrB(bfrB)	cell wall and cell processes	-1.18	8.17E-14	-1.33	3.49E-17	-2.93	1.26E-79
Rv3851	886186	membrane protein(Rv3851)	cell wall and cell processes	1.92	2.79E-10	1.32	2.38E-05	1.82	1.74E-09
Rv3865	886172	ESX-1 secretion-associated protein EspF(espF)	cell wall and cell processes	-1.24	2.99E-41	-1.03	1.00E-28	-1.63	1.84E-70
Rv3909	886245	hypothetical protein(Rv3909)	conserved hypotheticals	1.52	1.06E-42	1.43	1.21E-38	1.70	2.21E-54
Rv3921c	886238	membrane protein insertase YidC(Rv3921c)	cell wall and cell processes	1.32	1.08E-95	1.48	1.68E-122	2.10	6.39E-248
Rv3922c	886256	membrane protein insertion efficiency factor(Rv3922c)	virulence, detoxification, adapta- tion	1.13	1.65E-38	1.28	3.03E-50	2.00	2.50E-123
RVnc0001	14515861	ncRNA(AS1890)	conserved hypotheticals	-2.84	1.51E-09	-2.37	2.44E-09	-2.11	5.92E-08
RVnc0004	14515875	ncRNA(B11)	conserved hypotheticals	1.95	7.87E-07	2.28	2.15E-09	2.66	1.17E-12
RVnc0013	14515855	ncRNA(mcr11)	conserved hypotheticals	-2.03	8.33E-05	-1.20	2.27E-02	-1.63	1.44E-03
RVnc0021	14515894	ncRNA(mcr3)	conserved hypotheticals	1.26	3.50E-06	1.19	9.97E-06	1.23	4.04E-06
Rvnt36	2700442	tRNA-Gln(glnU)	conserved hypotheticals	2.14	6.20E-04	2.33	1.31E-04	3.29	1.97E-08

Appendix B: Supplementary Results of Chapter 3.2

Table 0.1: Parameters tested to establish sequential treatment and time kill assay for *M. tuberculosis*. The same parameters were tested for the sequence RIF->EMB, which is not shown here.

Concentration Pre-exposure [% of MIC]	Concentration Main-exposure [% of MIC]	Pre-exposure time [h]	Set up	Sampling timepoint [days]	Readout
75	75	1.5	96- well plate	0-4, 7, 10, 14, 21, 28	OD ₆₀₀
50	50				
25	25				
75	75	2	96- well plate	0-4, 7, 10, 14, 21, 28	OD ₆₀₀
50	50				
25	25				
75	75	8	96- well plate	0-4, 7, 10, 14, 21, 28	OD ₆₀₀
50	50				
25	25				
75	75	24	96- well plate	0-4, 7, 10, 14, 21, 28	OD ₆₀₀
50	50				
25	25				
75	75	2	96- well plate	0-6, 9-18	OD ₆₀₀
50	50				
25	25				
75	75	5	96- well plate	0-6, 9-18	OD ₆₀₀
50	50				
25	25				
75	75	8	96- well plate	0-6, 9-18	OD ₆₀₀
50	50				
25	25				
25	12	8	96- well plate	0, 2, 5, 6, 10, 14	CFU / mL
25	25				
25	50				
50	12	8	96- well plate	0, 2, 5, 6, 10, 14	CFU / mL
50	25				
50	50				
25	12	13	96- well plate	0, 2, 5, 6, 10, 14	CFU / mL
25	25				
25	50				
50	12	13	96- well plate	0, 2, 5, 6, 10, 14	CFU / mL
50	25				
50	50				
25	12	8	96- well plate	0, 3, 6, 11, 14	CFU / mL
25	25				
25	50				

Concentration Pre-exposure [% of MIC]	Concentration Main-exposure [% of MIC]	Pre-exposure time [h]	Set up	Sampling timepoint [days]	Readout
50	12	8	96- well plate	0, 3, 6, 11, 14	CFU / mL
50	25				
50	50				
25	12	10	96- well plate	0, 3, 6, 11, 14	CFU / mL
25	25				
25	50				
50	12	10	96- well plate	0, 3, 6, 11, 14	CFU / mL
50	25				
50	50				
25	12	8	10 µL conical tubes 45° angle	0, 2, 5, 10, 14	CFU / mL
25	25				
50	12	8	10 µL conical tubes 45° angle	0, 2, 5, 10, 14	CFU / mL
50	25				
25	25	8	10 µL conical tubes 45° angle	0, 1, 2, 5, 10, 14	CFU / mL
25	25	8	10 µL conical tubes 45° angle	0, 1, 2, 5, 10, 14	CFU / mL

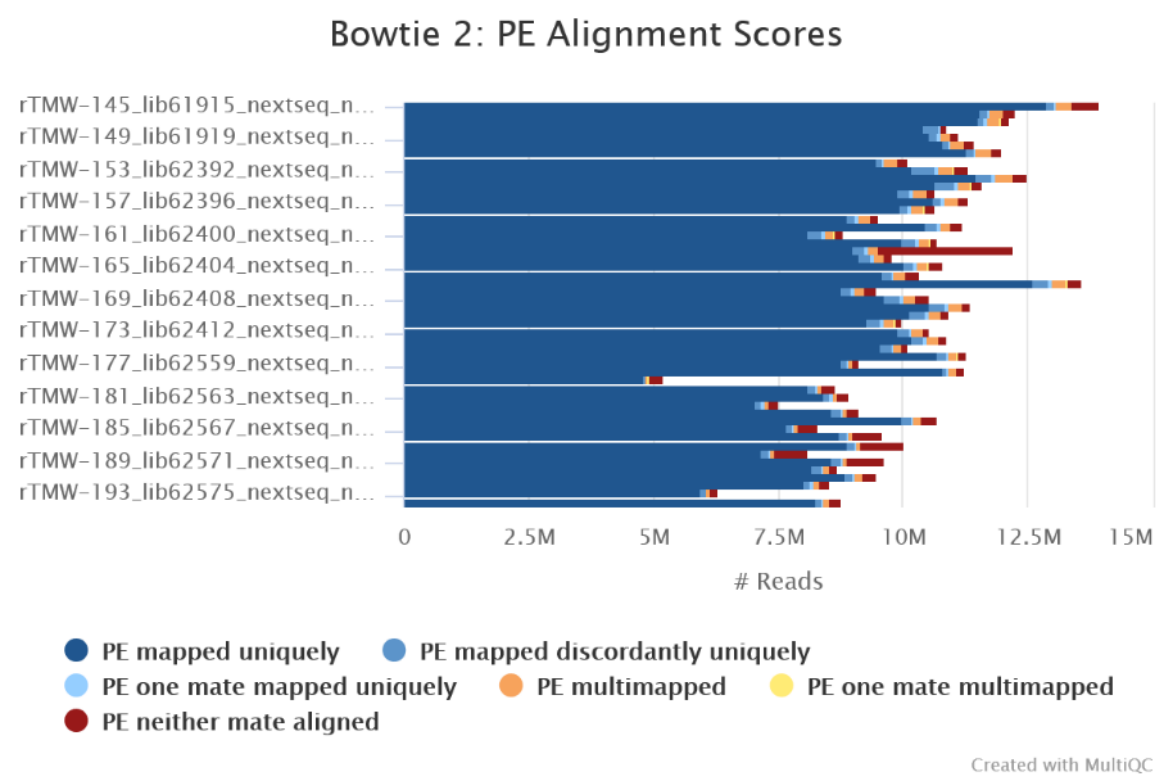


Figure 0.1: Bowtie2 PE alignment scores as quality control Hysteresis Sequencing. Bowtie2 Paired-end (PE) plot of number of reads aligning to reference in different ways as read counts.

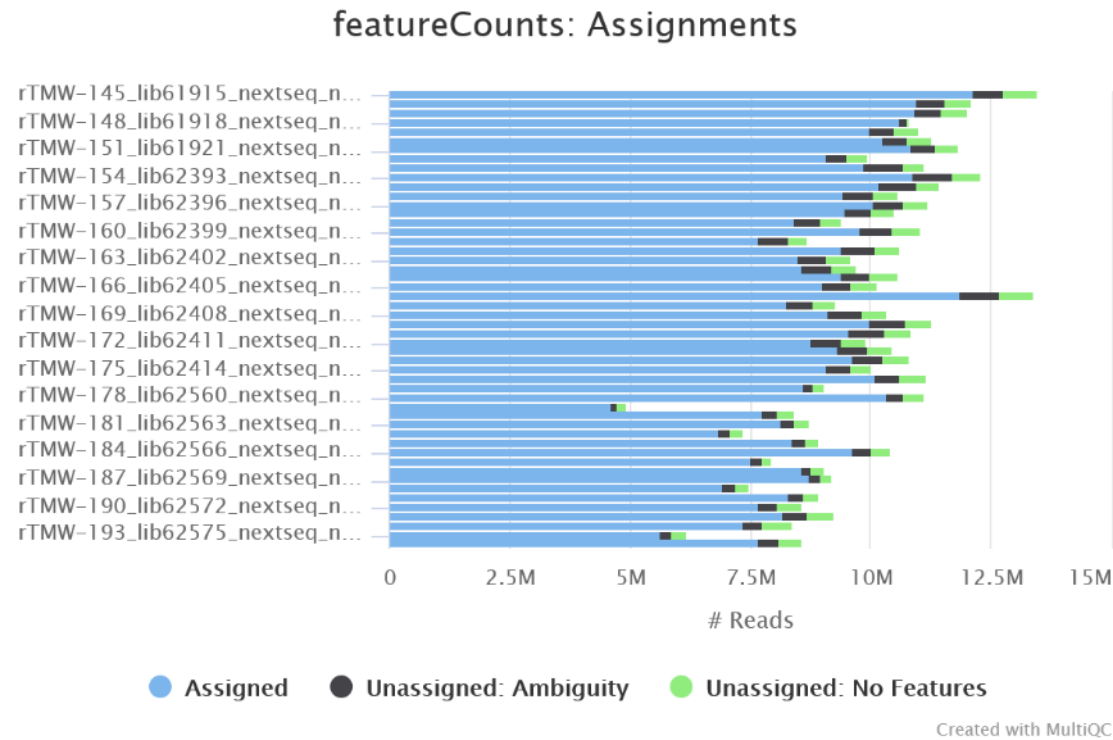


Figure 0.2: Figure 0.3: Quality control of assignment scores of Hysteresis Sequencing. FeatureCounts was used to assign mapped reads with features.

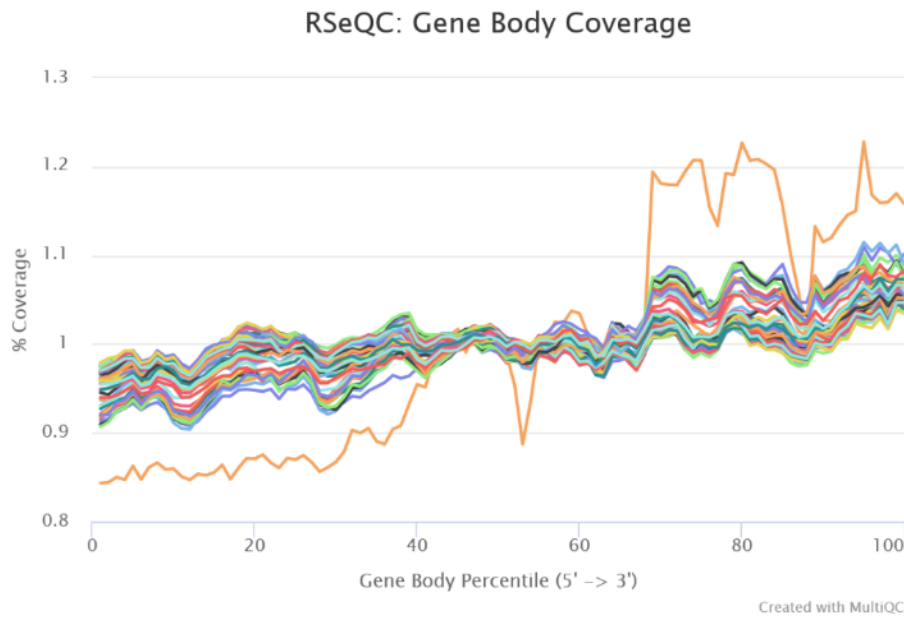


Figure 0.4: Gene body coverage as quality control of Hysteresis Sequencing. Gene Body Coverage assessed with RSeQC, uses read coverage over gene bodies to control uniformity of read coverage and check for 5' or 3' bias.

Table 0.2: RNA sequencing of Hysteresis experiment. Quality measurements of Qubit and Fragment analyser

Sample ID	Sample name	RQN	Qubit [ng / μ L]	Non-rRNA % (total)
rTMW-145_lib61915_nextseq_nz0012_101bp.bam	Med TP1 1	10	18.5	86.99
rTMW-146_lib61916_nextseq_nz0012_101bp.bam	Med TP1 2	10	16.4	85.03
rTMW-147_lib61917_nextseq_nz0012_101bp.bam	Med TP1 3	10	24	80.16
rTMW-148 --> inhogeneous gene body cover, excluded	Med TP1 4	9.8	20.8	11.28
rTMW-149_lib61919_nextseq_nz0012_101bp.bam	Med TP1 5	10	21.2	83.93
rTMW-150_lib61920_nextseq_nz0012_101bp.bam	RIF TP1 1	9.9	21	80.57
rTMW-151_lib61921_nextseq_nz0012_101bp.bam	RIF TP1 2	9.9	21.8	76.58
rTMW-152_lib61922_nextseq_nz0012_101bp.bam	RIF TP1 3	9.8	22.4	77.94
rTMW-153_lib62392_nextseq_nz0012_101bp.bam	RIF TP1 4	10	23.4	84.08
rTMW-154_lib62393_nextseq_nz0012_101bp.bam	RIF TP1 5	10	22.6	93.34
rTMW-155_lib62394_nextseq_nz0012_101bp.bam	EMB TP1 1	9.8	18.5	92.18
rTMW-156_lib62395_nextseq_nz0012_101bp.bam	EMB TP1 2	10	21	93.13
rTMW-157_lib62396_nextseq_nz0012_101bp.bam	EMB TP1 3	10	19.9	91.93
rTMW-158_lib62397_nextseq_nz0012_101bp.bam	EMB TP1 4	10	26.6	88.91
rTMW-159_lib62398_nextseq_nz0012_101bp.bam	EMB TP1 5	10	22.2	92.15
rTMW-160_lib62399_nextseq_nz0012_101bp.bam	Med TP2 1	10	29.2	91.87
rTMW-161_lib62400_nextseq_nz0012_101bp.bam	Med TP2 2	10	27.4	91.22
rTMW-162_lib62401_nextseq_nz0012_101bp.bam	Med TP2 3	10	29.6	93.09
rTMW-163_lib62402_nextseq_nz0012_101bp.bam	Med TP2 4	10	32	91.99
rTMW-164_lib62403_nextseq_nz0012_101bp.bam	Med TP2 5	10	28.4	94.70
rTMW-165_lib62404_nextseq_nz0012_101bp.bam	EMB RIF TP2 1	9.8	26.4	91.19

Sample ID	Sample name	RQN	Qubit [ng / μ L]	Non-rRNA % (total)
rTMW-166_lib62405_nextseq_nz0012_101bp.bam	EMB RIF TP2 2	9.5	31.4	89.49
rTMW-167_lib62406_nextseq_nz0012_101bp.bam	EMB RIF TP2 3	9.5	29.6	93.12
rTMW-168_lib62407_nextseq_nz0012_101bp.bam	EMB RIF TP2 4	9.3	24.8	88.92
rTMW-169_lib62408_nextseq_nz0012_101bp.bam	EMB RIF TP2 5	9.9	33.6	93.43
rTMW-170_lib62409_nextseq_nz0012_101bp.bam	RIF EMB TP2 1	10	28.8	94.81
rTMW-171_lib62410_nextseq_nz0012_101bp.bam	RIF EMB TP2 2	10	31.4	96.16
rTMW-172_lib62411_nextseq_nz0012_101bp.bam	RIF EMB TP2 3	10	35	94.49
rTMW-173_lib62412_nextseq_nz0012_101bp.bam	RIF EMB TP2 4	10	27.8	93.45
rTMW-174_lib62413_nextseq_nz0012_101bp.bam	RIF EMB TP2 5	10	26.2	92.64
rTMW-175_lib62414_nextseq_nz0012_101bp.bam	Med TP3 1	10	20.2	87.31
rTMW-176_lib62415_nextseq_nz0012_101bp.bam	Med TP3 2	10	12.2	89.41
rTMW-177_lib62559_nextseq_nz0012_101bp.bam	Med TP3 3	10	12.8	44.24
->exclude non-rRNA <60 %				
rTMW-178_lib62560_nextseq_nz0012_101bp.bam	Med TP3 4	10	20.6	71.70
rTMW-179 --> Reads mapped < 10M, excluded	Med TP3 5	10	25	63.34
rTMW-180_lib62562_nextseq_nz0012_101bp.bam	EMB RIF TP3 1	10	24	68.68
rTMW-181_lib62563_nextseq_nz0012_101bp.bam	EMB RIF TP3 2	10	22.2	63.06
rTMW-182_lib62564_nextseq_nz0012_101bp.bam	EMB RIF TP3 3	10	20.2	68.02
rTMW-183_lib62565_nextseq_nz0012_101bp.bam	EMB RIF TP3 4	10	19.6	56.48
-> exclude non-rRNA <60 %				
rTMW-184_lib62566_nextseq_nz0012_101bp.bam	EMB RIF TP3 5	10	22.4	63.31
rTMW-185_lib62567_nextseq_nz0012_101bp.bam	RIF EMB TP3 1	9.9	22.6	45.17
-> exclude non-rRNA <60 %				
rTMW-186_lib62568_nextseq_nz0012_101bp.bam	RIF EMB TP3 2	9.7	25	47.10
-> exclude non-rRNA <60 %				
rTMW-187_lib62569_nextseq_nz0012_101bp.bam	RIF EMB TP3 3	10	20.2	44.48
-> exclude non-rRNA <60 %				
rTMW-188_lib62570_nextseq_nz0012_101bp.bam	RIF EMB TP3 4	10	26.8	60.26
rTMW-189_lib62571_nextseq_nz0012_101bp.bam	RIF EMB TP3 5	10	24.2	60.51
rTMW-190_lib62572_nextseq_nz0012_101bp.bam	Med TP0 1	9.8	8.46	81.51
rTMW-191_lib62573_nextseq_nz0012_101bp.bam	Med TP0 2	9.8	9.88	87.63
rTMW-192_lib62574_nextseq_nz0012_101bp.bam	Med TP0 3	10	7.52	86.07
rTMW-193_lib62575_nextseq_nz0012_101bp.bam	Med TP0 4	9.8	6.04	75.97
rTMW-194_lib62576_nextseq_nz0012_101bp.bam	Med TP0 5	10	10.6	84.26

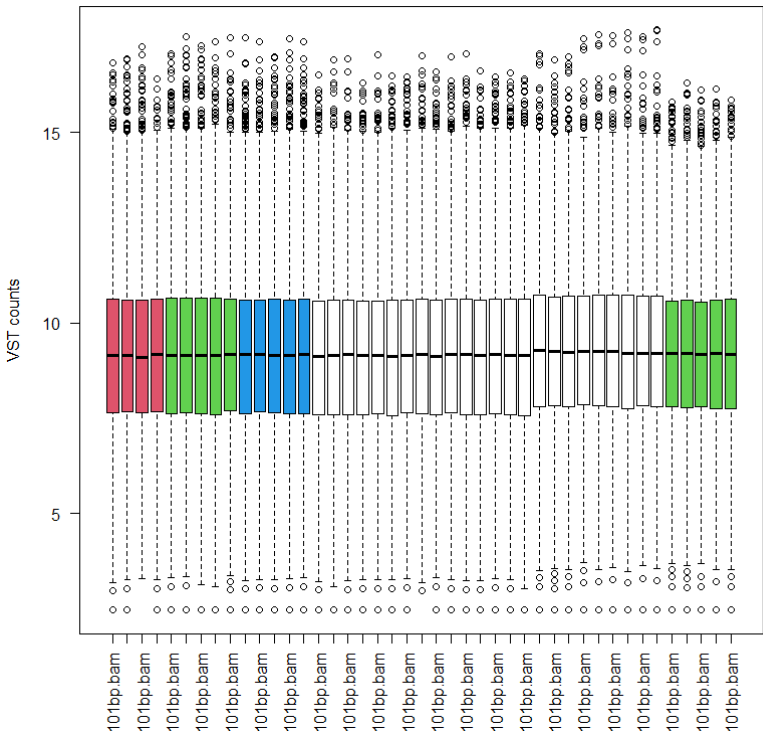


Figure 0.5: Biostatistics quality control using VST counts comparing means of samples of hysteresis sequencing analysis. The variance stabilizing transformation (vst) was used to normalize counts. The means of theses VST counts were compared using DESeq. This method was used to verify outliers after DESeq analysis before downstream differentially gene expression (DEG) analysis.

Table 0.3: Significant DEGs of EMB vs Medium 8 h (TP1), EMB->RIF 0.5h (TP2) and EMB->RIF 48 h (TP3): The significant differentially expressed genes (DEG) of each timepoint of the EMB pre-exposure sequence with the respective gene name and category according to mycobrowser.epfl.ch. Significant values of the respective timepoint are highlighted yellow ($\log_2FC > 1$, $padj < 0.05$). $\log_2FC$ = Log2 Fold change, $padj$ = adjusted p-value, EMB = ethambutol, RIF = rifampicin.

Locus	Gene name	Category	EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
			$\log_2FC$	$padj$	$\log_2FC$	$padj$	$\log_2FC$	$padj$
Rv0065	Possible toxin VapC1	virulence, detoxification, adaptation	-1.24	2.94E-10	0.41	5.02E-02	0.68	1.88E-03
Rv0076c	Probable membrane protein	cell wall and cell processes	-1.09	4.67E-06	-0.61	1.09E-02	-0.26	4.71E-01
Rv0081	Probable transcriptional regulatory protein	regulatory proteins	-1.00	9.31E-08	1.39	1.01E-14	-0.04	9.04E-01
Rv0082	Probable oxidoreductase	intermediary metabolism and respiration	0.32	2.21E-01	1.51	6.11E-10	0.27	4.76E-01
Rv0083	Probable oxidoreductase	intermediary metabolism and respiration	-0.31	6.91E-02	1.32	6.22E-18	0.05	8.61E-01
Rv0085	Possible hycP, hydrogenase, integral membrane protein	intermediary metabolism and respiration	-0.29	5.72E-02	1.26	3.54E-19	-0.05	8.37E-01
Rv0087	Possible hycE (alternate gene name: hevE), formate hydrogenlyase	intermediary metabolism and respiration	-0.50	1.13E-04	1.22	4.05E-23	0.06	8.01E-01
Rv0105c	50S ribosomal protein L28-1 RpmB1	information pathways	-1.47	1.05E-02	0.25	7.10E-01	-0.15	9.10E-01
Rv0122	Hypothetical protein	conserved hypothetical	1.00	1.40E-03	-0.55	6.61E-02	0.33	5.19E-01
Rv0124	PE-PGRS family protein PE_PGRS2	PE / PPE	1.18	1.04E-05	-0.05	8.89E-01	0.64	5.83E-02
Rv0186A	Metallothionein, MymT	virulence, detoxification, adaptation	1.17	2.67E-06	-0.86	4.95E-04	-0.41	2.35E-01
Rv0195	Possible two component transcriptional regulatory protein (probably LuxR-family)	regulatory proteins	-1.08	4.61E-04	0.06	8.87E-01	1.04	4.73E-05
Rv0251c	Heat shock protein Hsp	virulence, detoxification, adaptation	1.23	1.29E-05	1.44	7.92E-08	-0.54	1.64E-01
Rv0260c	Possible transcriptional regulatory protein	regulatory proteins	-1.21	1.39E-02	-0.30	6.07E-01	-0.05	9.54E-01
Rv0263c	Conserved hypothetical protein	conserved hypothetical	1.21	5.34E-22	-0.59	2.33E-06	-0.13	5.31E-01
Rv0278c	PE-PGRS family protein PE_PGRS3	PE / PPE	1.30	1.54E-05	-0.18	6.09E-01	0.32	4.97E-01
Rv0279c	PE-PGRS family protein PE_PGRS4	PE / PPE	1.48	6.03E-06	-0.05	9.11E-01	0.25	6.29E-01
Rv0285	PE family protein PE5	PE / PPE	1.91	3.28E-13	-0.35	2.26E-01	0.34	3.83E-01
Rv0287	ESAT-6 like protein EsxG	cell wall and cell processes	1.45	7.66E-17	-0.55	1.55E-03	0.47	4.02E-02
Rv0290	ESX conserved component EccD3. ESX-3 type VII secretion system protein	cell wall and cell processes	1.22	7.23E-07	-0.35	1.82E-01	0.51	1.26E-01
Rv0291	Probable mycP3, membrane-anchored serine protease	intermediary metabolism and respiration	1.21	6.15E-07	-0.28	2.91E-01	0.42	2.14E-01
Rv0297	PE-PGRS family protein PE_PGRS5	PE / PPE	1.09	7.01E-07	0.12	6.62E-01	0.39	2.07E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv0304c	PPE family protein PPE5	PE / PPE	-1.06	5.19E-16	0.12	4.36E-01	0.24	1.89E-01
Rv0341	IniB, isoniazid inducible gene protein	cell wall and cell processes	1.56	8.15E-14	-0.48	2.54E-02	3.46	9.48E-52
Rv0342	IniA, isoniazid-inducible gene	cell wall and cell processes	0.20	6.38E-02	0.42	2.30E-05	2.36	3.02E-104
Rv0343	IniC, isoniazid-inducible gene	cell wall and cell processes	0.35	1.31E-03	0.37	3.90E-04	1.61	2.13E-46
Rv0350	Probable chaperone protein DnaK	virulence, detoxification, adaptation	0.04	8.80E-01	1.53	4.94E-16	-0.36	2.03E-01
Rv0351	Probable grpE protein (HSP-70 cofactor)	virulence, detoxification, adaptation	0.12	6.60E-01	1.50	1.78E-11	-0.02	9.60E-01
Rv0352	Probable dnaJ1, chaperone protein	virulence, detoxification, adaptation	-0.06	8.01E-01	1.33	4.68E-13	0.21	4.77E-01
Rv0353	Probable hspR, heat shock regulatory protein	virulence, detoxification, adaptation	0.03	8.93E-01	1.07	1.91E-09	0.26	3.41E-01
Rv0384c	ClpB, probable endopeptidase ATP binding protein (chain B) (heat shock protein F84.1)	virulence, detoxification, adaptation	0.63	2.04E-03	1.28	2.40E-11	-0.77	2.22E-03
Rv0400c	Acyl-CoA dehydrogenase FadE7	lipid metabolism	1.32	7.97E-10	-0.18	4.78E-01	-0.04	9.16E-01
Rv0441c	Hypothetical protein	conserved hypothetical	1.09	5.07E-08	0.04	8.72E-01	0.35	2.12E-01
Rv0488	Probable conserved integral membrane protein	cell wall and cell processes	-1.22	1.10E-12	0.45	9.66E-03	0.69	5.88E-04
Rv0516c	Possible anti-anti-sigma factor	information pathways	1.36	2.30E-13	-0.56	2.83E-03	0.47	5.49E-02
Rv0563	Probable protease transmembrane protein heat shock protein HtpX	virulence, detoxification, adaptation	-0.21	2.12E-01	1.16	7.96E-15	0.04	8.90E-01
Rv0578c	PE-PGRS family protein PE_PGRS7	PE / PPE	1.41	1.25E-05	0.19	6.27E-01	0.21	6.83E-01
Rv0600c	Two component sensor kinase [second part]	regulatory proteins	-1.08	1.93E-02	-0.46	3.11E-01	-0.18	8.44E-01
Rv0650	Possible sugar kinase	intermediary metabolism and respiration	2.60	3.86E-05	-0.39	4.94E-01	0.28	7.77E-01
Rv0746	PE-PGRS family protein PE_PGRS9	PE / PPE	2.24	1.86E-06	-0.50	3.45E-01	0.20	7.98E-01
Rv0763c	Possible ferredoxin	intermediary metabolism and respiration	0.13	8.63E-01	-0.14	8.56E-01	1.22	9.25E-03
Rv0791c	Conserved protein	conserved hypothetical	-0.39	2.46E-02	0.16	4.14E-01	-1.11	7.71E-09
Rv0793	Possible monooxygenase	intermediary metabolism and respiration	-0.39	2.86E-01	-0.49	2.17E-01	-1.39	1.92E-09
Rv0805	Class III cyclic nucleotide phosphodiesterase (cNMP PDE)	intermediary metabolism and respiration	-1.06	7.89E-23	0.01	9.67E-01	-0.18	2.47E-01
Rv0833	PE-PGRS family protein PE_PGRS13	PE / PPE	2.25	4.32E-06	-0.06	9.22E-01	0.09	9.17E-01
Rv0834c	PE-PGRS family protein PE_PGRS14	PE / PPE	1.18	5.62E-06	-0.12	7.21E-01	0.51	1.53E-01
Rv0836c	Hypothetical protein	conserved hypothetical	-1.06	8.51E-05	0.19	5.37E-01	0.06	8.41E-01
Rv0841	Probable conserved transmembrane protein	cell wall and cell processes	0.45	4.98E-01	2.30	3.52E-04	-2.75	8.42E-02
Rv0872c	PE-PGRS family protein PE_PGRS15	PE / PPE	1.33	4.17E-06	0.32	3.30E-01	0.49	2.32E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv0874c	Conserved hypothetical protein	conserved hypothetical	0.18	2.78E-01	0.53	3.63E-04	1.03	3.66E-11
Rv0887c	Conserved hypothetical protein	conserved hypothetical	0.19	3.15E-01	1.09	2.29E-10	0.26	3.78E-01
Rv0962c	Possible lipoprotein LprP	cell wall and cell processes	-1.05	1.35E-03	0.33	3.33E-01	0.20	7.04E-01
Rv0972c	Acyl-CoA dehydrogenase FadE12	lipid metabolism	-1.31	1.52E-21	0.72	1.22E-06	0.55	4.92E-04
Rv0980c	PE-PGRS family protein PE_PGRS18	PE / PPE	1.55	5.59E-06	-0.31	3.80E-01	-0.15	7.90E-01
Rv0991c	Conserved serine rich protein	conserved hypothetical	-1.35	1.57E-12	0.46	2.21E-02	-0.80	7.14E-04
Rv1004c	Probable membrane protein	cell wall and cell processes	1.30	1.74E-08	-0.14	6.19E-01	0.35	2.90E-01
Rv1048c	Hypothetical protein	cell wall and cell processes	-0.62	1.10E-04	-0.12	5.12E-01	-1.02	3.61E-10
Rv1057	Conserved hypothetical protein	cell wall and cell processes	0.34	1.22E-03	0.05	7.24E-01	1.02	2.16E-17
Rv1066	Conserved hypothetical protein	cell wall and cell processes	1.31	2.81E-05	-0.44	1.78E-01	0.11	8.45E-01
Rv1067c	PE-PGRS family protein PE_PGRS19	PE / PPE	1.76	1.42E-06	-0.12	7.83E-01	0.26	6.64E-01
Rv1072	Probable conserved transmembrane protein	cell wall and cell processes	-0.09	6.76E-01	1.06	1.82E-09	-0.15	6.06E-01
Rv1073	Conserved hypothetical protein	conserved hypothetical	-0.46	1.31E-02	1.69	1.20E-23	-0.05	8.63E-01
Rv1087	PE-PGRS family protein PE_PGRS21	PE / PPE	1.38	3.79E-05	-0.02	9.66E-01	0.27	6.07E-01
Rv1091	PE-PGRS family protein PE_PGRS22	PE / PPE	1.50	4.36E-07	0.03	9.50E-01	0.07	8.94E-01
Rv1128c	Conserved hypothetical protein	conserved hypothetical	-0.37	2.11E-02	1.15	3.82E-16	0.22	3.49E-01
Rv1129c	Probable transcriptional regulator protein	regulatory proteins	0.65	4.17E-04	3.24	6.00E-90	-0.39	1.75E-01
Rv1130	Possible methylcitrate dehydratase PrpD	intermediary metabolism and respiration	0.85	1.88E-08	3.42	1.85E-146	0.09	7.53E-01
Rv1131	Probable prpC, methylcitrate synthase	intermediary metabolism and respiration	0.65	2.93E-04	3.35	3.07E-105	0.19	5.97E-01
Rv1169c	PE family protein. Possible lipase LipX.	PE / PPE	-1.21	2.84E-06	0.35	2.09E-01	-0.65	5.47E-02
Rv1199c	Possible transposase	insertion seqs and phages	-1.03	1.74E-08	0.73	7.55E-05	-0.14	6.12E-01
Rv1259	Probable uracil DNA glycosylase, UdgB	information pathways	1.24	9.93E-05	-0.48	1.58E-01	-0.11	8.44E-01
Rv1325c	PE-PGRS family protein PE_PGRS24	PE / PPE	1.89	1.70E-05	-0.15	7.93E-01	0.36	6.03E-01
Rv1375	Conserved hypothetical protein	conserved hypothetical	-0.22	1.85E-02	0.34	1.75E-04	-1.26	1.03E-39
Rv1385	Probable orotidine 5'-phosphate decarboxylase PyrF	intermediary metabolism and respiration	1.07	6.98E-05	-0.93	2.42E-04	-0.06	9.01E-01
Rv1389	Probable guanylate kinase Gmk	intermediary metabolism and respiration	1.29	1.14E-13	-0.41	2.21E-02	0.14	6.20E-01
Rv1441c	PE-PGRS family protein PE_PGRS26	PE / PPE	1.03	7.09E-04	-0.28	4.19E-01	-0.04	9.36E-01
Rv1450c	PE-PGRS family protein PE_PGRS27	PE / PPE	1.30	5.23E-05	-0.04	9.28E-01	0.24	6.36E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv1463	Probable conserved ATP-binding protein ABC transporter	cell wall and cell processes	1.03	1.91E-17	-0.25	4.63E-02	0.10	6.05E-01
Rv1468c	PE-PGRS family protein PE_PGRS29	PE / PPE	1.05	8.92E-07	-0.10	7.19E-01	0.07	8.37E-01
Rv1471	Probable thioredoxin TrxB1	intermediary metabolism and respiration	0.75	2.93E-07	2.12	2.20E-56	-0.88	2.46E-07
Rv1507A	Hypothetical protein	conserved hypothetical	-0.93	1.59E-02	0.39	3.47E-01	1.27	3.53E-02
Rv1542c	Hemoglobin GlbN	intermediary metabolism and respiration	-1.97	2.93E-07	0.14	7.54E-01	0.81	6.68E-02
Rv1569	Probable 8-amino-7-oxononanoate synthase BioF1	intermediary metabolism and respiration	1.51	4.20E-06	-0.39	2.87E-01	-0.07	9.02E-01
Rv1574	Probable PhiRv1 phage related protein	insertion seqs and phages	-0.13	7.16E-01	1.07	3.03E-04	0.83	2.59E-02
Rv1575	Probable phiRV1 phage protein	insertion seqs and phages	-0.23	6.60E-01	0.95	5.30E-02	1.37	2.95E-02
Rv1581c	Probable PhiRv1 phage protein	insertion seqs and phages	-1.30	6.06E-06	0.85	7.98E-03	0.67	5.83E-02
Rv1582c	Probable phiRv1 phage protein	insertion seqs and phages	-1.29	1.43E-11	1.00	1.87E-07	0.86	1.22E-04
Rv1592c	Conserved hypothetical protein	conserved hypothetical	-1.17	3.99E-37	0.12	2.38E-01	-1.18	1.02E-29
Rv1651c	PE-PGRS family protein PE_PGRS30	PE / PPE	1.21	1.74E-08	-0.14	5.84E-01	0.00	9.99E-01
Rv1654	Probable acetylglutamate kinase ArgB	intermediary metabolism and respiration	1.08	4.83E-05	-0.45	1.10E-01	-0.26	5.36E-01
Rv1674c	Probable transcriptional regulatory protein	regulatory proteins	0.29	1.14E-01	0.59	1.40E-03	-1.16	2.16E-09
Rv1682	Probable coiled-coil structural protein	conserved hypothetical	-1.10	1.75E-11	0.35	3.59E-02	0.08	7.69E-01
Rv1733c	Probable conserved transmembrane protein	conserved hypothetical	-0.05	8.77E-01	1.35	1.95E-06	0.52	1.98E-01
Rv1734c	Conserved hypothetical protein	conserved hypothetical	-1.79	4.06E-04	0.91	1.25E-01	0.15	8.53E-01
Rv1735a	NA		-1.68	3.46E-05	3.32	2.33E-05	0.09	8.61E-01
Rv1735c	Hypothetical membrane protein	cell wall and cell processes	-1.89	3.45E-11	0.80	9.83E-03	0.30	4.28E-01
Rv1736c	Probable nitrate reductase NarX	intermediary metabolism and respiration	-0.43	4.80E-02	1.51	5.35E-14	0.38	1.86E-01
Rv1754c	Conserved protein	conserved hypothetical	-0.61	1.07E-05	-0.55	5.16E-05	-1.04	5.76E-10
Rv1759c	PE-PGRS family protein Wag22	PE / PPE	1.39	1.14E-06	0.00	9.98E-01	0.30	4.97E-01
Rv1813c	Conserved hypothetical protein	conserved hypothetical	-0.52	3.08E-02	1.12	5.00E-07	0.09	8.27E-01
Rv1815	Conserved protein	conserved hypothetical	-0.28	7.34E-04	1.06	3.50E-45	-0.36	5.87E-04
Rv1816	Possible transcriptional regulatory protein	conserved hypothetical	-0.69	4.40E-09	1.15	3.61E-28	-0.18	3.20E-01
Rv1818c	PE-PGRS family protein PE_PGRS33	PE / PPE	2.19	7.09E-08	-0.28	5.54E-01	0.37	5.60E-01
Rv1870c	Conserved hypothetical protein	conserved hypothetical	-1.11	2.17E-19	0.03	8.39E-01	-0.14	4.60E-01
Rv1888A	Conserved hypothetical protein	conserved hypothetical	-1.10	1.17E-08	0.27	1.94E-01	-0.10	7.57E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv1930c	Conserved hypothetical protein	conserved hypothetical	1.39	3.21E-03	-0.37	4.78E-01	0.40	5.65E-01
Rv1958c	Hypothetical protein	conserved hypothetical	-1.19	4.58E-05	0.05	9.02E-01	0.22	5.94E-01
Rv1982c	Possible toxin VapC36	virulence, detoxification, adaptation	-1.02	2.16E-15	0.45	5.27E-04	0.25	1.52E-01
Rv1995	NA		-0.81	1.82E-03	0.38	2.26E-01	-1.15	2.12E-05
Rv1997	Probable metal cation transporter P-type ATPase A CtpF	cell wall and cell processes	-1.39	2.52E-13	1.03	2.55E-08	0.55	2.58E-02
Rv2003c	Conserved hypothetical protein	conserved hypothetical	-1.00	1.55E-08	1.10	2.03E-09	0.43	5.33E-02
Rv2004c	Conserved protein	conserved hypothetical	-1.17	1.46E-14	1.46	8.10E-25	0.73	7.37E-05
Rv2005c	Universal stress protein family protein	virulence, detoxification, adaptation	-1.05	4.45E-08	0.84	6.40E-06	0.72	2.71E-03
Rv2012	Conserved hypothetical protein	conserved hypothetical	-0.91	7.48E-06	0.67	2.56E-03	-1.61	6.84E-14
Rv2028c	Universal stress protein family protein	virulence, detoxification, adaptation	-0.85	1.95E-06	2.18	7.87E-36	0.16	5.57E-01
Rv2029c	6-phosphofructokinase PfkB	intermediary metabolism and respiration	-1.21	1.40E-14	2.46	5.32E-63	0.35	9.57E-02
Rv2030c	Conserved protein	conserved hypothetical	-0.57	6.92E-03	1.74	5.63E-20	0.26	3.88E-01
Rv2031c	Heat shock protein HspX	virulence, detoxification, adaptation	-0.29	3.87E-01	1.49	4.44E-07	0.17	7.36E-01
Rv2032	Conserved protein Acg	conserved hypothetical	0.01	9.71E-01	1.08	6.88E-04	0.41	3.88E-01
Rv2033c	Conserved hypothetical protein	conserved hypothetical	0.67	5.24E-02	-1.09	1.01E-03	-0.20	7.03E-01
Rv2050	Conserved protein	conserved hypothetical	-0.15	3.02E-01	1.08	2.67E-18	-0.49	3.31E-03
Rv2052c	Conserved protein	conserved hypothetical	0.92	8.18E-06	1.63	8.95E-18	0.53	4.88E-02
Rv2053c	Probable transmembrane protein FxsA	cell wall and cell processes	-0.33	4.49E-02	1.72	1.85E-31	0.00	9.97E-01
Rv2073c	Probable shortchain dehydrogenase	intermediary metabolism and respiration	1.05	3.60E-05	-0.42	9.95E-02	0.11	8.05E-01
Rv2123	PPE family protein PPE37	PE / PPE	1.08	6.60E-07	-0.72	2.91E-04	0.14	7.83E-01
Rv2126c	PE-PGRS family protein PE_PGRS37	PE / PPE	1.26	3.82E-06	0.07	8.44E-01	0.19	6.83E-01
Rv2142c	Possible toxin ParE2	virulence, detoxification, adaptation	-1.15	1.84E-08	-0.26	2.29E-01	0.21	4.50E-01
Rv2153c	MurG peptidoglycan biosynthesis	cell wall and cell processes	1.18	4.24E-04	-0.11	7.97E-01	0.23	6.64E-01
Rv2158c	MurE peptidoglycan biosynthesis	cell wall and cell processes	1.21	1.72E-05	-0.18	5.87E-01	0.24	5.77E-01
Rv2161c	Conserved protein	conserved hypothetical	1.07	4.83E-08	0.19	3.87E-01	0.08	8.04E-01
Rv2162c	PE-PGRS family protein PE_PGRS38	PE / PPE	1.34	1.12E-06	0.13	6.98E-01	0.06	9.04E-01
Rv2206	Probable conserved transmembrane protein	cell wall and cell processes	1.11	2.91E-08	-0.22	3.21E-01	0.04	9.04E-01
Rv2207	Probable phosphoribosyltransferase CobT	intermediary metabolism and respiration	1.64	1.70E-07	-0.41	2.24E-01	0.05	9.24E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv2208	Probable cobS, cobalamin 5'-phosphate synthase	intermediary metabolism and respiration	1.13	6.39E-08	-0.31	1.65E-01	-0.01	9.78E-01
Rv2225	3-methyl-2-oxobutanoate hydroxymethyltransferase PanB	intermediary metabolism and respiration	0.94	3.15E-13	0.09	5.18E-01	-1.18	4.38E-16
Rv2277c	Possible glycerolphosphodiesterase	intermediary metabolism and respiration	-1.17	1.67E-04	-0.27	3.98E-01	0.14	8.11E-01
Rv2281	Putative phosphate-transport permease PitB	cell wall and cell processes	-1.54	5.43E-32	0.66	1.59E-06	-0.21	2.80E-01
Rv2353c	PPE family protein PPE39	PE / PPE	-1.76	6.08E-04	0.20	7.70E-01	0.78	2.75E-01
Rv2365c	Conserved hypothetical protein	conserved hypothetical	1.07	1.42E-04	0.50	8.18E-02	0.52	1.81E-01
Rv2372c	Conserved hypothetical protein	conserved hypothetical	0.99	1.48E-10	1.19	8.00E-17	0.39	5.47E-02
Rv2373c	Probable chaperone protein DnaJ2	virulence, detoxification, adaptation	0.15	1.58E-01	1.18	3.73E-36	0.13	3.82E-01
Rv2378c	Lysine-N-oxygenase MbtG	lipid metabolism	0.00	9.85E-01	-1.05	2.19E-22	0.33	6.24E-02
Rv2398c	Probable sulfate-transport ABC transporter CysW	cell wall and cell processes	-1.02	4.43E-11	0.32	4.69E-02	-0.24	2.90E-01
Rv2428	Alkyl hydroperoxide reductase C protein AhpC	virulence, detoxification, adaptation	-1.00	6.66E-09	0.06	7.66E-01	-0.45	4.61E-02
Rv2432c	Hypothetical protein	conserved hypothetical	-1.26	1.57E-06	-0.01	9.66E-01	1.04	1.36E-03
Rv2450c	Probable resuscitation-promoting factor RpfE	cell wall and cell processes	1.04	1.70E-08	-0.70	7.44E-05	0.00	9.97E-01
Rv2451	Hypothetical proline and serine rich protein	conserved hypothetical	-1.07	3.91E-02	-0.50	3.59E-01	0.17	8.54E-01
Rv2452c	Hypothetical	conserved hypothetical	-1.36	1.07E-10	0.09	7.16E-01	0.60	2.13E-02
Rv2466c	Conserved protein	conserved hypothetical	-0.67	9.54E-04	1.45	2.12E-14	-1.03	1.63E-05
Rv2490c	PE-PGRS family protein PE_PGRS43	PE / PPE	1.24	4.03E-09	0.34	1.24E-01	0.43	1.35E-01
Rv2491	Conserved hypothetical protein	conserved hypothetical	-1.21	8.03E-03	0.60	2.29E-01	0.76	2.40E-01
Rv2512c	Transposase for insertion sequence element IS1081	insertion seqs and phages	-1.12	3.48E-11	1.22	1.78E-11	0.06	8.40E-01
Rv2516c	Hypothetical protein	conserved hypothetical	-1.25	1.35E-12	0.50	4.95E-03	-0.42	7.08E-02
Rv2540c	Probable chorismate synthase AroF	intermediary metabolism and respiration	1.26	4.20E-07	0.13	6.74E-01	0.26	4.87E-01
Rv2541	Hypothetical alanine rich protein	conserved hypothetical	1.92	3.98E-03	-0.85	1.97E-01	0.74	5.07E-01
Rv2546	Possible toxin VapC18	virulence, detoxification, adaptation	-0.29	5.84E-01	1.09	2.32E-02	0.48	6.30E-01
Rv2570	Conserved hypothetical protein	conserved hypothetical	0.57	7.24E-03	0.23	3.31E-01	1.18	2.53E-06
Rv2615c	PE-PGRS family protein PE_PGRS45	PE / PPE	1.42	1.30E-06	-0.41	1.88E-01	0.10	8.47E-01
Rv2622	Possible methyltransferase (methylase)	intermediary metabolism and respiration	-0.62	1.05E-03	0.31	1.24E-01	-1.41	4.94E-11
Rv2623	TB31.7, universal stress protein family protein	virulence, detoxification, adaptation	-0.27	9.93E-02	1.22	1.65E-16	0.72	2.22E-04

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv2624c	Universal stress protein family protein	virulence, detoxification, adaptation	-1.20	1.62E-13	1.79	1.18E-26	0.37	7.90E-02
Rv2625c	Probable conserved transmembrane alanine and leucine rich protein	cell wall and cell processes	-0.99	1.58E-08	1.20	6.53E-13	0.66	2.33E-03
Rv2626c	Hypoxic response protein 1 Hrp1	conserved hypothetical	-0.43	1.52E-01	1.23	6.82E-06	0.58	1.31E-01
Rv2630	Hypothetical protein	conserved hypothetical	-1.41	7.90E-22	1.13	2.27E-15	0.61	7.39E-04
Rv2631	Conserved hypothetical protein	conserved hypothetical	-1.26	3.77E-25	1.04	1.59E-18	0.71	5.63E-07
Rv2635	Hypothetical protein	conserved hypothetical	-3.18	4.07E-02	-1.31	4.69E-01	0.44	8.90E-01
Rv2658c	Possible prophage protein	insertion seqs and phages	-1.27	6.85E-06	-0.55	1.46E-01	0.00	9.96E-01
Rv2659c	Probable PhiRv2 prophage integrase	insertion seqs and phages	-1.19	2.39E-11	0.62	1.67E-03	0.13	6.15E-01
Rv2662	Hypothetical protein	conserved hypothetical	-2.26	3.08E-06	0.29	6.51E-01	0.70	5.01E-02
Rv2663	Hypothetical protein	conserved hypothetical	-1.11	4.94E-13	0.49	1.89E-03	0.08	7.48E-01
Rv2677c	Probable protoporphyrinogen oxidase HemY	intermediary metabolism and respiration	1.59	1.55E-06	-0.10	8.09E-01	0.43	3.77E-01
Rv2693c	Probable conserved integral membrane	cell wall and cell processes	-0.27	7.22E-02	1.15	2.37E-17	0.67	1.67E-04
Rv2707	Probable conserved transmembrane	cell wall and cell processes	-0.69	9.28E-06	1.12	8.61E-15	-0.21	3.58E-01
Rv2710	RNA polymerase sigma factor SigB	information pathways	-0.12	5.28E-01	1.09	7.17E-11	-0.34	1.68E-01
Rv2719c	Possible conserved membrane protein	conserved hypothetical	1.40	9.39E-09	0.07	8.13E-01	-0.21	5.84E-01
Rv2728c	Conserved alanine rich protein	intermediary metabolism and respiration	1.01	1.14E-04	0.22	4.37E-01	0.39	3.01E-01
Rv2739c	Possible alanine rich transferase	intermediary metabolism and respiration	1.04	5.44E-07	-0.32	1.53E-01	0.02	9.54E-01
Rv2782c	Probable zinc protease PepR	virulence, detoxification, adaptation	1.05	2.62E-08	0.12	5.94E-01	0.14	6.48E-01
Rv2791c	Probable transposase	insertion seqs and phages	1.17	1.65E-20	-0.02	9.08E-01	0.27	1.18E-01
Rv2793c	Probable tRNA pseudouridine synthase B TruB	regulatory proteins	1.03	2.79E-06	-0.01	9.67E-01	0.22	5.15E-01
Rv2804c	Hypothetical protein	conserved hypothetical	-3.16	1.64E-02	1.45	2.47E-01	1.02	6.02E-01
Rv2812	Probable transposase	insertion seqs and phages	-0.33	1.57E-01	1.49	5.65E-12	0.29	4.46E-01
Rv2813	Conserved hypothetical protein	conserved hypothetical	-0.81	4.26E-03	1.63	2.40E-08	0.66	1.07E-01
Rv2817c	Conserved hypothetical protein	conserved hypothetical	-1.13	3.99E-08	0.38	9.34E-02	0.44	1.36E-01
Rv2823c	Conserved protein	conserved hypothetical	-1.05	7.54E-18	0.10	4.95E-01	-0.16	3.83E-01
Rv2863	Possible toxin VapC23	virulence, detoxification, adaptation	-1.84	1.05E-02	-1.75	2.71E-02	-0.28	8.67E-01
Rv2875	Major secreted immunogenic protein Mpt70	cell wall and cell processes	-0.59	1.30E-03	1.20	1.69E-12	0.26	3.71E-01
Rv2897c	Conserved hypothetical protein	conserved hypothetical	0.47	1.48E-05	1.10	5.02E-28	0.52	1.38E-03

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv2913c	Possible D-amino acid aminohydrolase	intermediary metabolism and respiration	-0.47	3.76E-03	0.27	1.26E-01	-1.44	5.94E-16
Rv2961	Probable transposase	insertion seqs and phages	-0.57	2.98E-01	-0.40	5.11E-01	1.55	3.18E-02
Rv2980	Possible conserved secreted protein	conserved hypothetical	1.16	6.34E-07	-0.32	1.47E-01	-0.28	4.25E-01
Rv3018c	PPE family protein PPE46	conserved hypothetical	0.18	1.24E-01	0.19	1.12E-01	-1.02	2.26E-16
Rv3020c	ESAT-6 like protein EsxS	cell wall and cell processes	-0.05	8.16E-01	0.71	2.21E-05	-1.26	6.53E-11
Rv3021c	PPE family protein PPE47	PE / PPE	-0.43	1.05E-02	0.24	1.65E-01	-1.73	1.80E-25
Rv3022A	PE family protein PE29	PE / PPE	-0.05	8.72E-01	0.36	1.27E-01	-1.20	2.51E-08
Rv3022c	PPE family protein PPE48	PE / PPE	-0.27	4.50E-01	0.47	1.56E-01	-2.11	3.91E-16
Rv3023c	Probable transposase	insertion seqs and phages	-1.14	1.87E-06	1.17	1.31E-06	0.21	5.95E-01
Rv3054c	Conserved hypothetical protein	conserved hypothetical	-0.23	3.68E-01	1.22	5.89E-07	-1.01	1.99E-04
Rv3083	Probable monooxygenase (hydroxylase)	intermediary metabolism and respiration	0.32	1.42E-03	1.26	1.61E-43	-0.59	1.12E-06
Rv3084	Probable acetyl-hydrolase / esterase LipR	intermediary metabolism and respiration	0.48	1.01E-02	1.33	2.14E-14	-0.45	1.10E-01
Rv3085	Probable short-chain type dehydrogenase / reductase	intermediary metabolism and respiration	0.06	6.07E-01	1.30	1.05E-36	-0.38	1.92E-02
Rv3087	Possible triacylglycerol synthase	lipid metabolism	0.26	8.56E-02	1.15	2.90E-17	0.50	5.62E-03
Rv3088	Putative triacylglycerol synthase Tgs4	lipid metabolism	-0.06	5.47E-01	1.06	9.58E-38	0.34	8.29E-04
Rv3089	Probable chain -fatty-acid-CoA ligase FadD13	lipid metabolism	-0.26	8.44E-03	1.04	9.97E-30	0.19	1.35E-01
Rv3093c	Hypothetical oxidoreductase	conserved hypothetical	1.16	1.23E-05	0.10	7.68E-01	-0.13	7.60E-01
Rv3108	Hypothetical protein	conserved hypothetical	-1.21	4.20E-02	1.02	1.32E-01	-0.88	3.54E-01
Rv3109	Probable molybdenum cofactor biosynthesis protein A MoaA1	intermediary metabolism and respiration	-1.04	3.46E-05	-0.29	2.80E-01	0.19	5.84E-01
Rv3114	Conserved hypothetical protein	conserved hypothetical	-1.22	8.95E-03	0.13	8.28E-01	-0.34	6.02E-01
Rv3115	Probable transposase	insertion seqs and phages	-1.16	3.37E-08	1.13	4.80E-07	-0.06	8.80E-01
Rv3118	Conserved hypothetical protein SseC1	Conserved hypothetical protein SseC1	1.03	2.10E-07	-0.09	7.23E-01	-0.20	4.97E-01
Rv3124	Transcriptional regulatory protein MoaR1	regulatory proteins	-1.02	1.49E-09	-0.05	7.99E-01	0.31	1.80E-01
Rv3126c	Hypothetical protein	conserved hypothetical	-1.27	3.44E-03	0.59	2.19E-01	0.45	4.62E-01
Rv3128c	Conserved hypothetical protein	conserved hypothetical	-1.92	8.11E-23	1.58	1.69E-16	0.81	3.75E-04
Rv3129	Conserved hypothetical protein	conserved hypothetical	-1.18	9.06E-06	0.14	6.57E-01	0.33	3.92E-01
Rv3130c	Triacylglycerol synthase (diacylglycerol acyltransferase) Tgs1	intermediary metabolism and respiration	-0.01	9.74E-01	1.18	1.25E-10	1.07	3.59E-06

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv3131	Conserved protein	conserved hypothetical	0.26	1.90E-01	1.12	2.32E-10	0.94	2.68E-05
Rv3168	Putative aminoglycoside phosphotransferase	intermediary metabolism and respiration	-0.52	4.71E-03	0.12	6.06E-01	-1.55	6.68E-15
Rv3169	Conserved protein	conserved hypothetical	0.27	9.05E-02	-0.22	2.03E-01	-1.04	1.73E-09
Rv3177	Possible peroxidase	intermediary metabolism and respiration	-1.54	3.58E-03	-0.22	7.24E-01	-0.32	7.28E-01
Rv3178a	NA		-2.58	3.73E-02	1.13	3.75E-01	-0.16	9.42E-01
Rv3202a	NA		1.05	2.77E-05	0.97	5.36E-05	0.41	1.65E-01
Rv3229c	Possible linoleoyl-CoA desaturase	lipid metabolism	-0.67	1.65E-05	1.37	5.12E-21	0.04	8.93E-01
Rv3230c	Hypothetical oxidoreductase	intermediary metabolism and respiration	0.22	1.60E-01	1.31	1.46E-21	0.06	7.98E-01
Rv3251c	Probable rubredoxin RubA	intermediary metabolism and respiration	1.25	2.44E-10	0.79	3.16E-05	-0.32	2.39E-01
Rv3275c	Probable phosphoribosylaminoimidazole carboxylase PurE	intermediary metabolism and respiration	1.05	1.45E-06	-0.34	1.38E-01	-0.17	6.34E-01
Rv3288c	Putative protein UsfY	conserved hypothetical	-1.43	5.03E-26	0.83	1.48E-09	0.40	2.04E-02
Rv3289c	Possible transmembrane protein	cell wall and cell processes	-1.27	1.52E-06	1.02	6.82E-04	0.59	7.85E-02
Rv3290c	Probable L-lysine-epsilon aminotransferase Lat	intermediary metabolism and respiration	-1.14	1.93E-14	0.53	4.08E-04	0.21	3.27E-01
Rv3307	Probable purine nucleoside phosphorylase DeoD	intermediary metabolism and respiration	1.11	3.25E-05	-0.42	1.40E-01	-0.09	8.51E-01
Rv3344c	PE-PGRS family protein PE_PGRS49	PE / PPE	1.72	1.96E-07	0.22	5.67E-01	0.46	3.48E-01
Rv3345c	PE-PGRS family protein PE_PGRS50	PE / PPE	1.85	1.06E-07	0.16	7.11E-01	0.51	3.05E-01
Rv3388	PE-PGRS family protein PE_PGRS52	PE / PPE	1.41	4.74E-05	-0.43	2.61E-01	0.01	9.90E-01
Rv3395A	Probable membrane protein	conserved hypothetical	-0.22	3.02E-01	-0.26	2.25E-01	-1.21	2.94E-04
Rv3424c	Hypothetical protein	conserved hypothetical	-1.31	2.54E-06	-0.28	3.58E-01	0.65	7.71E-02
Rv3428c	Possible transposase	insertion seqs and phages	-1.01	4.93E-05	1.01	1.03E-04	0.49	1.47E-01
Rv3433c	Conserved protein	conserved hypothetical	1.09	7.74E-04	-0.16	6.88E-01	0.35	4.82E-01
Rv3438	Conserved protein	conserved hypothetical	1.03	8.31E-04	-0.52	1.04E-01	0.08	8.76E-01
Rv3463	Conserved protein	conserved hypothetical	-0.17	3.82E-01	0.96	7.25E-08	-1.32	1.81E-10
Rv3507	PE-PGRS family protein PE_PGRS53	PE / PPE	1.45	5.49E-06	-0.05	9.00E-01	0.52	2.51E-01
Rv3508	PE-PGRS family protein PE_PGRS54	PE / PPE	1.80	3.66E-05	-0.18	7.45E-01	-0.01	9.88E-01
Rv3511	PE-PGRS family protein PE_PGRS55	PE / PPE	1.56	1.54E-07	-0.13	7.32E-01	-0.01	9.78E-01
Rv3512	PE-PGRS family protein PE_PGRS56	PE / PPE	1.62	8.41E-06	-0.21	6.22E-01	0.17	7.81E-01
Rv3514	PE-PGRS family protein PE_PGRS57	PE / PPE	1.50	1.06E-05	0.01	9.75E-01	0.22	6.84E-01

			EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rv3533c	PPE family protein PPE62	PE / PPE	-1.13	9.47E-11	0.42	2.13E-02	0.85	4.92E-05
Rv3548c	Probable short-chain type dehydrogenase / reductase	intermediary metabolism and respiration	-1.03	8.11E-23	0.23	6.28E-02	-0.45	2.40E-04
Rv3552	Possible CoA-transferase	intermediary metabolism and respiration	-1.04	3.26E-13	0.57	3.54E-04	-0.72	3.59E-05
Rv3589	Probable adenine glycosylase MutY	intermediary metabolism and respiration	1.15	6.82E-05	-0.44	1.45E-01	0.01	9.85E-01
Rv3590c	PE-PGRS family protein PE_PGRS58	PE / PPE	1.09	2.54E-07	-0.14	5.65E-01	0.07	8.36E-01
Rv3595c	PE-PGRS family protein PE_PGRS59	PE / PPE	1.24	9.17E-09	0.63	3.29E-03	0.54	5.87E-02
Rv3612c	Conserved hypothetical protein	conserved hypothetical	-1.17	6.51E-14	0.45	8.38E-03	-0.26	2.57E-01
Rv3613c	Hypothetical protein	conserved hypothetical	-1.12	1.21E-10	0.17	4.27E-01	-0.59	8.33E-03
Rv3616c	ESX-1 secretion-associated protein A, EspA	cell wall and cell processes	-0.62	6.08E-06	0.43	1.34E-03	-1.14	2.69E-13
Rv3643	Hypothetical protein	conserved hypothetical	-3.41	1.83E-02	-0.25	8.56E-01	-1.50	5.00E-01
Rv3647c	Conserved hypothetical protein	conserved hypothetical	1.57	2.62E-05	-0.38	3.60E-01	-0.24	6.82E-01
Rv3652	PE-PGRS family-related protein PE_PGRS60	PE / PPE	1.03	6.79E-08	-0.20	3.26E-01	-0.17	5.88E-01
Rv3653	PE-PGRS family-related protein PE_PGRS61	PE / PPE	1.24	1.10E-06	0.08	8.09E-01	0.25	5.34E-01
Rv3657c	Possible conserved alanine rich membrane protein	cell wall and cell processes	-0.86	1.75E-04	0.89	2.77E-04	1.05	1.57E-03
Rv3659c	Conserved hypothetical protein	conserved hypothetical	-0.57	8.31E-06	1.67	1.93E-44	0.24	2.08E-01
Rv3662c	Conserved hypothetical protein	conserved hypothetical	1.31	5.65E-06	-0.47	1.03E-01	0.10	8.45E-01
Rv3673c	Possible membrane-anchored thioredoxin-like protein	cell wall and cell processes	1.06	2.30E-13	-0.14	3.86E-01	0.13	5.71E-01
Rv3706c	Conserved hypothetical proline rich protein	conserved hypothetical	1.00	2.61E-08	-0.06	8.04E-01	0.46	6.32E-02
Rv3830c	Transcriptional regulatory protein (probably TetR-family)	regulatory proteins	-0.08	6.90E-01	1.07	3.64E-11	0.22	4.34E-01
Rv3837c	Probable phosphoglycerate mutase	intermediary metabolism and respiration	0.29	3.85E-02	1.32	9.33E-27	0.35	4.57E-02
Rv3841	Bacterioferritin BfrB	intermediary metabolism and respiration	0.56	4.71E-02	1.21	4.19E-06	0.17	6.99E-01
Rv3861	Hypothetical protein	conserved hypothetical	1.07	1.77E-02	-0.42	3.76E-01	0.44	6.20E-01
Rv3862c	Possible transcriptional regulatory protein WhiB-like WhiB6	regulatory proteins	-0.62	4.17E-03	0.05	8.58E-01	1.07	1.91E-05
RVnc0021	NA		1.50	2.29E-06	0.31	3.85E-01	1.29	9.84E-04
RVnc0036	NA		-0.24	3.17E-01	-0.35	1.91E-01	1.03	8.86E-06
Rvnt02	NA		-1.18	6.15E-07	-0.13	6.45E-01	0.14	7.33E-01

		EMB vs. Medium 8 h (TP1)		EMB-RIF vs Med 0.5 h (TP2)		EMB-RIF vs Med 48 h (TP3)	
Rvnt03	NA	0.78	1.92E-01	-0.71	2.04E-01	1.80	3.55E-02
Rvnt20	NA	-1.23	1.77E-01	-1.93	3.40E-02	0.24	8.83E-01
Rvnt22	NA	-3.49	1.54E-03	-0.22	8.39E-01	0.02	9.88E-01
Rvnt33	NA	-0.02	9.65E-01	-0.40	2.79E-01	1.65	1.33E-02

Table 0.4: Significant DEGs of RIF vs Medium 8 h (TP1), RIF-> EMB 0.5 h (TP2) and RIF->EMB 48 h (TP3): The significant differentially expressed genes (DEG) of each *M. tuberculosis* at timepoint of the EMB pre-exposure sequence with the respective gene name and category according to mycobrowser.epfl.ch. Significant values of the respective timepoint are highlighted yellow ($\log_2FC > 1$, $padj < 0.05$). $\log_2FC$ = Log2 Fold change, $padj$ = adjusted p-value, EMB = ethambutol, RIF= rifampicin.

Locus	Gene and function	Category	RIF vs Med 8 h (TP1)		RIF-EMB 0.5 h (TP2)		RIF-EMB 48 h (TP3)	
			$\log_2FC$	$padj$	$\log_2FC$	$padj$	$\log_2FC$	$padj$
Rv0076c	Probable membrane protein	cell wall and cell processes	-1.05	5.86E-05	-0.22	4.21E-01	-0.63	1.32E-01
Rv0081	Probable transcriptional regulatory protein	regulatory proteins	-0.46	3.15E-02	1.05	2.41E-08	-0.17	6.21E-01
Rv0082	Probable oxidoreductase	intermediary metabolism and respiration	0.11	7.49E-01	1.14	9.11E-06	0.53	1.78E-01
Rv0124	PE-PGRS family protein PE_PGRS2	PE / PPE	0.89	3.14E-03	-0.03	9.30E-01	1.17	1.66E-03
Rv0183	Possible lysophospholipase	intermediary metabolism and respiration	-0.12	2.76E-01	-0.13	1.73E-01	-1.01	7.35E-14
Rv0251c	Heat shock protein Hsp	virulence, detoxification	0.52	1.15E-01	1.65	1.00E-09	-1.13	6.98E-03
Rv0278c	PE-PGRS family protein PE_PGRS3	PE / PPE	1.27	1.21E-04	-0.10	8.08E-01	0.56	2.56E-01
Rv0279c	PE-PGRS family protein PE_PGRS4	PE / PPE	1.19	9.41E-04	0.06	9.00E-01	1.00	4.43E-02
Rv0285	PE family protein PE5	PE / PPE	1.43	4.10E-07	-0.14	6.89E-01	0.62	1.38E-01
Rv0287	ESAT-6 like protein EsxG	cell wall and cell processes	1.07	5.58E-09	-0.24	2.34E-01	0.41	1.38E-01
Rv0290	ESX conserved component EccD3. ESX-3 type VII secretion system protein	cell wall and cell processes	1.08	5.69E-05	-0.27	3.52E-01	0.60	1.23E-01
Rv0291	Probable membrane-anchored mycosin MycP3 (serine protease)	intermediary metabolism and respiration	1.02	1.41E-04	-0.18	5.64E-01	0.64	9.16E-02
Rv0342	Isoniazid inducible gene protein IniA	cell wall and cell processes	0.13	3.14E-01	0.80	6.19E-17	1.64	5.74E-35
Rv0343	Isoniazid inducible gene protein IniC	cell wall and cell processes	0.19	1.42E-01	0.46	8.80E-06	1.33	9.04E-22
Rv0350	Probable chaperone protein DnaK (heat shock protein 70)	virulence, detoxification	0.18	4.96E-01	1.20	7.78E-10	-0.63	4.16E-02
Rv0351	Probable grpE protein (HSP-70 cofactor)	virulence, detoxification	0.18	5.53E-01	1.17	5.45E-07	-0.09	8.53E-01
Rv0578c	PE-PGRS family protein PE_PGRS7	PE / PPE	1.12	1.81E-03	0.33	4.00E-01	0.56	2.94E-01
Rv0628c	Conserved hypothetical protein	conserved hypothetical	0.62	9.68E-04	0.10	6.39E-01	1.03	1.85E-05
Rv0736	Anti-sigma factor RslA	information pathways	0.51	6.00E-03	-0.12	5.65E-01	1.03	2.03E-05
Rv0746	PE-PGRS family protein PE_PGRS9	PE / PPE	2.10	4.21E-05	-0.28	6.38E-01	0.58	4.72E-01
Rv0833	PE-PGRS family protein PE_PGRS13	PE / PPE	2.30	1.51E-05	-0.11	8.80E-01	0.59	4.86E-01
Rv0834c	PE-PGRS family protein PE_PGRS14	PE / PPE	1.06	2.17E-04	-0.12	7.21E-01	0.83	3.48E-02
Rv0872c	PE-PGRS family protein PE_PGRS15	PE / PPE	1.13	4.54E-04	0.38	2.66E-01	0.87	4.93E-02
Rv0874c	Conserved hypothetical protein	conserved hypothetical	0.06	8.00E-01	0.16	4.05E-01	1.02	1.03E-07
Rv0887c	Conserved hypothetical protein	conserved hypothetical	0.65	6.58E-04	0.31	1.52E-01	1.37	5.12E-08
Rv0970	Probable conserved integral membrane protein	conserved hypothetical	0.20	1.94E-01	0.04	8.29E-01	1.10	2.81E-10
Rv0972c	Acyl-CoA dehydrogenase FadE12	lipid metabolism	-1.01	1.26E-12	0.67	1.12E-05	0.38	5.25E-02
Rv0980c	PE-PGRS family protein PE_PGRS18	PE / PPE	1.24	1.25E-03	-0.16	6.97E-01	0.33	5.76E-01
Rv1066	Conserved hypothetical protein	conserved hypothetical	1.13	1.27E-03	-0.62	5.52E-02	0.53	3.17E-01
Rv1067c	PE-PGRS family protein PE_PGRS19	PE / PPE	1.62	4.96E-05	-0.04	9.40E-01	0.46	4.71E-01
Rv1087	PE-PGRS family protein PE_PGRS21	PE / PPE	1.13	2.47E-03	0.01	9.79E-01	0.74	1.67E-01

			RIF vs Med 8 h (TP1)		RIF-EMB 0.5 h (TP2)		RIF-EMB 48 h (TP3)	
Rv1091	PE-PGRS family protein PE_PGRS22	PE / PPE	1.19	3.00E-04	0.21	5.75E-01	0.54	2.66E-01
Rv1129c	Probable transcriptional regulator protein	regulatory proteins	0.14	5.54E-01	3.07	1.91E-80	-0.11	7.98E-01
Rv1130	Possible methylcitrate dehydratase PrpD	intermediary metabolism and respiration	0.38	2.91E-02	3.00	3.81E-112	1.10	3.33E-06
Rv1131	Probable prpC, methylcitrate synthase	intermediary metabolism and respiration	0.38	6.35E-02	2.92	1.55E-79	1.10	3.82E-04
Rv1176c	Conserved hypothetical protein	conserved hypothetical	-0.27	1.29E-01	-0.09	6.35E-01	-1.15	1.70E-07
Rv1243c	PE-PGRS family protein PE_PGRS23	PE / PPE	0.66	5.27E-03	0.53	1.84E-02	1.00	1.55E-03
Rv1325c	PE-PGRS family protein PE_PGRS24	PE / PPE	1.75	3.44E-04	-0.10	8.72E-01	0.67	3.59E-01
Rv1357c	Conserved hypothetical protein	conserved hypothetical	-1.29	9.43E-03	0.21	NA	0.45	6.07E-01
Rv1397c	Possible toxin VapC10	virulence, detoxification	-1.04	7.55E-17	-0.41	9.89E-04	-0.79	4.21E-06
Rv1450c	PE-PGRS family protein PE_PGRS27	PE / PPE	1.15	1.24E-03	0.10	8.25E-01	0.92	6.39E-02
Rv1471	Probable thioredoxin TrxB1	intermediary metabolism and respiration	1.09	3.12E-13	1.12	1.66E-15	-0.46	3.87E-02
Rv1569	Probable 8-amino-7-oxononanoate synthase BioF1	intermediary metabolism and respiration	1.02	5.54E-03	-0.17	7.02E-01	0.09	8.93E-01
Rv1575	Probable PhiRv1 phage protein	insertion seqs and phages	0.35	5.38E-01	0.58	NA	1.44	4.87E-02
Rv1662	Probable polyketide synthase Pks8	lipid metabolism	0.00	9.97E-01	0.40	2.65E-04	1.03	2.60E-12
Rv1663	Probable polyketide synthase Pks17	lipid metabolism	-0.07	7.08E-01	0.13	4.95E-01	1.10	7.40E-08
Rv1733c	Probable conserved transmembrane protein	conserved hypothetical	0.06	8.87E-01	1.32	6.23E-06	0.32	5.24E-01
Rv1735c	Hypothetical membrane protein	conserved hypothetical	-1.14	1.01E-04	0.31	4.21E-01	0.11	8.34E-01
Rv1736c	Probable nitrate reductase NarX	intermediary metabolism and respiration	-0.36	1.40E-01	1.30	3.80E-10	0.43	2.05E-01
Rv1738	Conserved protein	conserved hypothetical	-0.07	8.84E-01	1.02	6.47E-03	0.58	3.25E-01
Rv1754c	Conserved protein	conserved hypothetical	-0.36	2.24E-02	-0.40	6.12E-03	-1.27	2.39E-09
Rv1759c	PE-PGRS family protein Wag22	PE / PPE	1.11	4.64E-04	-0.04	9.10E-01	0.64	1.65E-01
Rv1806	PE family protein PE20	PE / PPE	0.24	4.33E-01	-0.17	5.95E-01	-1.09	3.39E-03
Rv1818c	PE-PGRS family protein PE_PGRS33	PE / PPE	1.84	3.53E-05	-0.23	6.61E-01	0.70	2.97E-01
Rv1840c	PE-PGRS family protein PE_PGRS34	PE / PPE	0.58	2.44E-03	0.49	5.96E-03	1.04	2.36E-05
Rv1930c	Conserved hypothetical protein	conserved hypothetical	1.11	3.99E-02	0.20	NA	0.48	5.42E-01
Rv1944c	Conserved protein	conserved hypothetical	-0.03	8.96E-01	-0.15	3.94E-01	-1.12	9.59E-06
Rv1973	Possible conserved Mce associated membrane protein	cell wall and cell processes	0.10	8.62E-01	0.31	NA	1.33	5.71E-03
Rv1996	Universal stress protein family protein	virulence, detoxification	0.48	2.55E-02	1.19	4.89E-11	0.36	2.40E-01
Rv2004c	Conserved protein	conserved hypothetical	-0.74	6.77E-06	1.22	5.91E-17	0.50	3.03E-02
Rv2007c	Ferredoxin FdxA	intermediary metabolism and respiration	-0.33	4.24E-01	1.06	1.80E-03	0.28	6.36E-01
Rv2012	Conserved hypothetical protein	conserved hypothetical	-0.39	9.18E-02	0.48	4.93E-02	-1.13	2.64E-05
Rv2028c	Universal stress protein family protein	virulence, detoxification	-0.55	5.94E-03	1.41	1.62E-14	0.13	6.87E-01
Rv2029c	6-phosphofructokinase PfkB	intermediary metabolism and respiration	-0.80	1.97E-06	1.75	3.28E-31	0.20	4.50E-01
Rv2030c	Conserved protein	conserved hypothetical	-0.36	1.41E-01	1.37	2.30E-12	0.11	7.79E-01
Rv2031c	Heat shock protein HspX	virulence, detoxification	0.01	9.75E-01	1.46	1.33E-06	-0.06	9.21E-01
Rv2032	Conserved protein Acg	conserved hypothetical	0.06	8.87E-01	1.40	1.12E-05	0.31	5.87E-01
Rv2052c	Conserved protein	conserved hypotheticals	0.76	7.86E-04	1.01	5.45E-07	0.92	2.36E-03
Rv2086	Conserved hypothetical protein	conserved hypotheticals	-0.22	5.88E-01	-0.33	NA	1.20	9.87E-03

			RIF vs Med 8 h (TP1)		RIF-EMB 0.5 h (TP2)		RIF-EMB 48 h (TP3)	
Rv2092c	ATP-dependent DNA helicase HelY	information pathways	0.35	5.70E-04	0.51	5.73E-09	1.02	2.18E-16
Rv2113	Probable integral membrane protein	cell wall and cell processes	-0.12	3.98E-01	-0.51	6.32E-06	-1.08	1.07E-08
Rv2207	Probable nicotinate-nucleotide-dimethylbenzimidazol phosphoribosyltransferase CobT	intermediary metabolism and respiration	1.11	1.41E-03	-0.25	5.19E-01	0.21	7.18E-01
Rv2281	Putative phosphate-transport permease PitB	cell wall and cell processes	-1.14	2.32E-17	0.39	9.50E-03	0.08	7.56E-01
Rv2353c	PPE family protein PPE39	PE / PPE	-1.29	2.27E-02	-0.03	NA	0.95	2.45E-01
Rv2386c	Isochorismate synthase MbtI	lipid metabolism	0.16	4.22E-01	-0.27	1.30E-01	-1.34	4.51E-08
Rv2490c	PE-PGRS family protein PE_PGRS43	PE / PPE	1.08	1.66E-06	0.44	5.15E-02	0.91	3.36E-03
Rv2570	Conserved hypothetical protein	conserved hypotheticals	0.40	1.08E-01	-0.03	9.10E-01	1.15	1.60E-04
Rv2578c	Conserved hypothetical protein	conserved hypotheticals	-0.11	7.24E-01	0.93	1.99E-06	1.05	3.61E-04
Rv2615c	PE-PGRS family protein PE_PGRS45	PE / PPE	1.12	5.83E-04	-0.17	6.48E-01	0.35	4.99E-01
Rv2622	Possible methyltransferase (methylase)	intermediary metabolism and respiration	-0.53	1.33E-02	0.27	2.11E-01	-1.51	7.27E-09
Rv2623	Universal stress protein family protein TB31.7	virulence, detoxification	0.02	9.18E-01	1.16	1.91E-14	0.57	1.73E-02
Rv2624c	Universal stress protein family protein	virulence, detoxification	-0.84	1.58E-06	1.42	3.24E-16	0.48	4.44E-02
Rv2626c	Hypoxic response protein 1 Hrp1	conserved hypotheticals	-0.28	4.32E-01	1.36	1.13E-06	0.39	4.17E-01
Rv2662	Hypothetical protein	conserved hypotheticals	-2.40	1.62E-05	0.39	5.53E-01	0.32	5.14E-01
Rv2677c	Probable protoporphyrinogen oxidase HemY	intermediary metabolism and respiration	1.15	1.81E-03	0.03	9.57E-01	0.67	2.11E-01
Rv2710	RNA polymerase sigma factor SigB	information pathways	-0.12	6.16E-01	1.26	8.66E-14	-0.82	1.58E-03
Rv2745c	Transcriptional regulatory protein ClgR	regulatory proteins	-0.13	6.23E-01	0.83	5.34E-05	-1.07	3.47E-04
Rv2811	Conserved hypothetical protein	conserved hypotheticals	0.50	5.70E-02	0.61	6.09E-03	1.13	1.34E-03
Rv2813	Conserved hypothetical protein	conserved hypotheticals	-0.11	7.53E-01	1.15	2.97E-04	1.35	1.23E-03
Rv2875	Major secreted immunogenic protein Mpt70	cell wall and cell processes	0.79	2.46E-05	0.14	5.68E-01	1.17	9.64E-06
Rv2896c	Conserved hypothetical protein	conserved hypotheticals	0.36	9.77E-03	-0.10	5.10E-01	1.01	5.81E-09
Rv2897c	Conserved hypothetical protein	conserved hypotheticals	0.76	9.29E-12	0.70	4.47E-11	1.11	2.51E-10
Rv2913c	Possible D-amino acid aminohydrolase	intermediary metabolism and respiration	-0.21	2.89E-01	0.21	2.62E-01	-1.27	6.61E-09
Rv2932	Phenolphthiocerol synthesis type-I polyketide synthase PpsB	lipid metabolism	0.40	1.66E-06	0.28	3.74E-04	1.54	1.52E-51
Rv2933	Phenolphthiocerol synthesis type-I polyketide synthase PpsC	lipid metabolism	0.79	1.26E-37	0.31	7.18E-07	1.45	7.76E-69
Rv2934	Phenolphthiocerol synthesis type-I polyketide synthase PpsD	lipid metabolism	0.75	1.54E-16	0.14	1.72E-01	1.68	2.51E-46
Rv2935	Phenolphthiocerol synthesis type-I polyketide synthase PpsE	lipid metabolism	0.42	5.44E-08	0.03	7.65E-01	1.04	5.95E-26
Rv2936	Daunorubicin-dim-transport ATP-binding protein ABC transporter DrrA	cell wall and cell processes	0.49	4.22E-16	-0.05	4.65E-01	1.08	1.86E-41
Rv2939	Possible conserved polyketide synthase associated protein PapA5	lipid metabolism	0.10	4.36E-01	-0.15	2.14E-01	1.01	3.06E-12
Rv3074	Conserved hypothetical protein	conserved hypotheticals	0.43	1.37E-04	0.67	1.30E-12	1.18	6.00E-18
Rv3083	Probable monooxygenase (hydroxylase)	intermediary metabolism and respiration	0.21	6.87E-02	1.11	1.31E-33	-0.49	1.21E-03
Rv3084	Probable acetyl-hydrolase / esterase LipR	intermediary metabolism and respiration	0.14	5.43E-01	1.08	2.15E-09	-0.02	9.70E-01

			RIF vs Med 8 h (TP1)		RIF-EMB 0.5 h (TP2)		RIF-EMB 48 h (TP3)	
Rv3085	Probable short-chain type dehydrogenase / reductase	intermediary metabolism and respiration	0.10	4.70E-01	1.08	1.28E-24	-0.23	2.88E-01
Rv3113	Possible phosphatase	intermediary metabolism and respiration	-1.15	3.19E-03	0.00	NA	0.47	3.94E-01
Rv3127	Conserved protein	conserved hypotheticals	-0.04	9.05E-01	1.02	2.73E-05	0.62	1.04E-01
Rv3130c	Triacylglycerol synthase (diacylglycerol acyl-transferase) Tgs1	lipid metabolism	0.45	4.27E-02	1.13	1.68E-09	0.92	1.21E-03
Rv3168	Putative aminoglycoside phosphotransferase	virulence, detoxification	-0.27	2.03E-01	0.03	8.98E-01	-1.11	7.22E-06
Rv3174	Probable short-chain dehydrogenase / reductase	lipid metabolism	-0.07	8.41E-01	-0.05	8.76E-01	-1.37	1.06E-02
Rv3289c	Possible transmembrane protein	cell wall and cell processes	-1.18	4.21E-05	1.10	3.28E-04	0.29	5.06E-01
Rv3343c	PPE family protein PPE54	PE / PPE	-0.06	7.07E-01	0.60	5.98E-07	1.03	1.76E-09
Rv3344c	PE-PGRS family protein PE_PGRS49	PE / PPE	1.56	1.33E-05	0.14	7.56E-01	1.31	7.54E-03
Rv3345c	PE-PGRS family protein PE_PGRS50	PE / PPE	1.68	8.20E-06	0.12	7.97E-01	1.34	9.16E-03
Rv3388	PE-PGRS family protein PE_PGRS52	PE / PPE	1.13	3.38E-03	-0.19	6.72E-01	0.39	5.19E-01
Rv3445c	ESAT-6 like protein EsxU	cell wall and cell processes	0.68	2.27E-03	-0.22	4.01E-01	1.27	2.19E-06
Rv3467	Conserved hypothetical protein	conserved hypotheticals	-0.56	6.60E-02	0.21	4.65E-01	1.10	4.58E-04
Rv3507	PE-PGRS family protein PE_PGRS53	PE / PPE	1.30	2.25E-04	-0.07	8.69E-01	0.81	1.05E-01
Rv3508	PE-PGRS family protein PE_PGRS54	PE / PPE	1.67	5.46E-04	-0.27	6.35E-01	0.54	4.67E-01
Rv3511	PE-PGRS family protein PE_PGRS55	PE / PPE	1.33	4.23E-05	0.01	9.77E-01	0.37	4.67E-01
Rv3512	PE-PGRS family protein PE_PGRS56	PE / PPE	1.37	6.72E-04	-0.09	8.66E-01	0.95	9.89E-02
Rv3514	PE-PGRS family protein PE_PGRS57	PE / PPE	1.28	6.49E-04	0.02	9.65E-01	0.90	8.61E-02
Rv3552	Possible CoA-transferase	intermediary metabolism and respiration	-1.13	1.34E-14	0.42	1.56E-02	-0.63	4.18E-03
Rv3565	Possible aspartate aminotransferase AspB	intermediary metabolism and respiration	-1.02	5.36E-14	-0.05	8.21E-01	-0.07	8.20E-01
Rv3595c	PE-PGRS family protein PE_PGRS59	PE / PPE	1.09	2.46E-06	0.57	1.21E-02	1.09	4.91E-04
Rv3647c	Conserved hypothetical protein	conserved hypotheticals	1.18	4.95E-03	-0.23	6.34E-01	-0.12	8.71E-01
Rv3654c	Conserved hypothetical protein	conserved hypotheticals	0.37	4.32E-01	0.57	NA	1.61	9.58E-03
Rv3659c	Conserved hypothetical protein	conserved hypotheticals	-0.37	1.05E-02	1.57	9.49E-39	0.18	4.40E-01
Rv3743c	Probable cation transporter P-type ATPase CtpJ	PE / PPE	-0.02	9.51E-01	-0.44	7.59E-02	-1.13	8.77E-03
Rv3823c	Conserved integral membrane transport protein MmpL8	cell wall and cell processes	0.20	3.98E-02	-0.17	6.55E-02	1.06	1.87E-19
Rv3832c	Conserved protein	conserved hypotheticals	-0.19	3.16E-01	-0.05	8.34E-01	-1.52	4.69E-06
Rv3839	Conserved hypothetical protein	conserved hypotheticals	0.38	1.16E-01	-0.26	2.90E-01	-1.11	1.27E-03
Rv3841	Bacterioferritin BfrB	intermediary metabolism and respiration	0.03	9.39E-01	1.92	8.79E-14	0.01	9.80E-01
RVnc0021	NA	NA	1.08	2.24E-03	-0.39	2.98E-01	2.03	6.07E-06
RVnc0046	NA	NA	0.82	8.69E-03	-0.13	7.21E-01	1.01	1.51E-02
Rvnt34	NA	NA	0.26	NA	-0.22	NA	2.29	3.15E-02
Rvnt35	NA	NA	-0.61	1.74E-01	-1.06	5.98E-03	0.56	4.42E-01

Table 0.5: GO analysis of EMB vs Medium 8 h (TP1), upregulated DEGs, Chapter 4.2.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Ben- ja- mini	FDR
Annotation Cluster 1,	Enrichment Score 2.99													
GOTERM_CC_DIRECT	GO:0009986~cell surface	7	7.61	3.29E-05	RV1818C	885551	PE-PGRS family protein PE_PGRS33(PE_PGRS33)	40	43	2574	1.05E+01	4.60E-04	0.00	0.00
					RV1651C	885174	PE-PGRS family protein PE_PGRS30(PE_PGRS30)							
					RV0285	886608	PE family protein PE5(PE5)							
					RV1441C	886626	PE-PGRS family protein PE_PGRS26(PE_PGRS26)							
					RV0278C	886623	PE-PGRS family protein PE_PGRS3(Rv0278c)							
					RV1468C	886556	PE-PGRS family protein PE_PGRS29(PE_PGRS29)							
					RV3653	886259	PE-PGRS family-related protein PE_PGRS61(PE_PGRS61)							
UP_KW_CELLULAR_COMPONENT	KW-0134~Cell wall	7	7.61	3.07E-04	RV2615C	888215	PE-PGRS family protein PE_PGRS45(PE_PGRS45)	22	64	1326	6.59E+00	2.76E-03	0.00	0.00
					RV1818C	885551	PE-PGRS family protein PE_PGRS33(PE_PGRS33)							
					RV1651C	885174	PE-PGRS family protein PE_PGRS30(PE_PGRS30)							
					RV0278C	886623	PE-PGRS family protein PE_PGRS3(Rv0278c)							
					RV1468C	886556	PE-PGRS family protein PE_PGRS29(PE_PGRS29)							
					RV3653	886259	PE-PGRS family-related protein PE_PGRS61(PE_PGRS61)							
					RV0980C	885327	PE-PGRS family protein PE_PGRS18(PE_PGRS18)							
UP_KW_CELLULAR_COMPONENT	KW-0964~Secreted	9	9.78	1.70E-03	RV2615C	888215	PE-PGRS family protein PE_PGRS45(PE_PGRS45)	22	157	1326	3.46E+00	1.52E-02	0.01	0.01
					RV1818C	885551	PE-PGRS family protein PE_PGRS33(PE_PGRS33)							

[illegible]

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Ben- ja- mini	FDR
Annotation Cluster 1,	Enrichment Score 2.99													
					RV2450C	885760	resuscitation-promoting factor RpfE(rpfE)							
					RV1651C	885174	PE-PGRS family protein							
					RV0285	886608	PE_PGRS30(PE_PGRS30)							
					RV0287	886604	PE family protein PE5(PE5)							
					RV0291	886615	ESAT-6 like protein EsxG(esxG)							
					RV1759C	885325	membrane-anchored mycosin							
					RV0278C	886623	MycP(mycP3)							
					RV1468C	886556	PE-PGRS family protein							
					RV1004C	886039	Wag22(wag22)							
					RV3653	886259	PE-PGRS family protein							
					RV0980C	885327	PE_PGRS29(PE_PGRS29)							
							PE_PGRS61(PE_PGRS61)							
							PE-PGRS family protein							
							PE_PGRS18(PE_PGRS18)							
Annotation Cluster 2	Enrichment Score: 0.49													
Category	Term	Count	%	P-Value	Genes			List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Ben- ja- mini	FDR
GOTERM_BP_DIRECT	GO:0071555~cell wall organization	3	3.26	2.39E-01	RV2153C	888036	UDP-N-acetylglucosamine--N-acetylmuramyl-(pentapeptide) pyrophosphoryl-undecaprenol-N-acetylglucosamine transferase(murG)	33	65	2226	3.11E+00	1.00E+00	1	1
					RV2158C	887252	UDP-N-acetylmuramoylalanyl-D-glutamate--2,6-diaminopimelate ligase(murE)							
					RV2719C	887959	membrane protein(Rv2719c)							

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Ben- ja- mini	FDR
Annotation Cluster 1,	Enrichment Score 2.99													
UP_KW_BIO- LOGICAL_PRO- CESS	KW-0961~Cell wall biogenesis / degradation	3	3.26	3.70E-01	RV2153C	888036	UDP-N-acetylglucosamine--N-acetylmuramyl-(pentapeptide) pyrophosphoryl-undecaprenol-N-acetylglucosamine transferase(murG)	30	64	1446	2.26E+00	1.00E+00	1	1
					RV2158C	887252	UDP-N-acetylmuramoylalanyl-D-glutamate--2,6-diaminopimelate ligase(murE)							
					RV2719C	887959	membrane protein(Rv2719c)							
UP_KW_BIO- LOGICAL_PRO- CESS	KW-9998~Cell wall biogenesis / degradation	3	3.26	3.70E-01	RV2153C	888036	UDP-N-acetylglucosamine--N-acetylmuramyl-(pentapeptide) pyrophosphoryl-undecaprenol-N-acetylglucosamine transferase(murG)	30	64	1446	2.26E+00	1.00E+00	1	1
					RV2158C	887252	UDP-N-acetylmuramoylalanyl-D-glutamate--2,6-diaminopimelate ligase(murE)							
					RV2719C	887959	membrane protein(Rv2719c)							

Table 0.6: GO analysis of EMB vs Medium 8 h (TP1), downregulated DEGs, Chapter 4.2.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
Annotation cluster 1	Enrichment Score: 1.88.													
GOTERM_MF_DIRECT	GO:0004803~transposase activity	4	5	5.85E-03	RV3115 RV3023 C RV1199 C RV2512 C	888790 888525 886092 888515	transposase(Rv3115) transposase(Rv3023c) IS1081 transposase(Rv1199c) IS1081 transposase(Rv2512c)	44	23	2612	10.32	3.25E-01	0.40	0.40
GOTERM_BP_DIRECT	GO:0006313~DNA transposition	4	5	7.80E-03	RV3115 RV3023 C RV1199 C RV2512 C RV3124	888790 888525 886092 888515 888825	transposase(Rv3115) transposase(Rv3023c) IS1081 transposase(Rv1199c) IS1081 transposase(Rv2512c) regulator MoaR(moaR1)	40	24	2226	9.28	3.19E-01	0.37	0.37
GOTERM_MF_DIRECT	GO:0003677~DNA binding	9	11.2 5	4.86E-02	RV3115 RV3023 C RV2512 C RV2817 C RV0195 RV0260 C RV1199 C	888790 888525 888515 888506 886762 886651 886092	transposase(Rv3115) transposase(Rv3023c) IS1081(Rv2512c) CRISPR-associated endonuclease Cas1(Rv2817c) two component transcriptional regulator(Rv0195) transcriptional regulator(Rv0260c) (Rv1199c)	44	251	2612	2.13	9.64E-01	1	1

			RV2659 C	885098	(Rv2659c)	
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Table 0.7: KEGG pathway enrichment analysis of EMB vs Medium 8 h (TP1) upregulated DEGs, Chapter 4.4.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List	Pop	Pop	Fold En-	Bonfer-	Benja-	FDR
								To- tal	Hits	To- tal	richment	roni	mini	
KEGG_PATHWAY	mtu01100: Metabolic pathways	14	15.05	7.29E-02	RV1569	886345	8-amino-7-oxononanoate synthase(bioF1)	17	675	1109	1.35E+00	0.90	1	1
					RV0400C	886427	acyl-CoA dehydrogenase FadE7(fadE7)							
					RV1385	886763	orotidine 5'-phosphate decarboxylase(pyrF)							
					RV1389	886787	guanylate kinase(gmk)							
					RV2158C	887252	UDP-N-acetylmuramoylalanyl-D-glutamate--2,6-diaminopimelate ligase(murE)							
					RV2540C	887379	chorismate synthase(aroF)							
					RV3307	887542	purine nucleoside phosphorylase(deoD)							
					RV2208	887572	adenosylcobinamide-GDP ribazoletransferase(cobS)							
					RV2677C	887711	protoporphyrinogen oxidase(hemY)							
					RV2207	887781	nicotinate-nucleotide-dimethylbenzimidazol phosphoribosyltransferase(cobT)							
					RV2153C	888036	(murG)							
					RV1654	888076	acetylglutamate kinase(argB)							
					RV0650	888082	sugar kinase(Rv0650)							
					RV3275C	888721	5-(carboxyamino)imidazole ribonucleotide mutase(purE)							
KEGG_PATHWAY	mtu00860: Porphyrin metabolism	3	3.23	9.25E-02	RV2207	887781	nicotinate-nucleotide-dimethylbenzimidazol phosphoribosyltransferase(cobT)	17	36	1109	5.44E+00	0.95	1	1
					RV2208	887572	adenosylcobinamide-GDP ribazoletransferase(cobS)							
					RV2677C	887711	protoporphyrinogen oxidase(hemY)							

Table 0.8: GO analysis of EMB->RIF vs Medium TP2 upregulated DEGs, Chapter 4.2.2.

[illegible]

Category	Term	Count	%	P Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
GOTERM_BP_DIRECT	GO:0071731~response to nitric oxide	3	3.896	4.13E-02	RV3087	888653	diacylglycerol O-acyltransferase(Rv3087)	44	17	2226	8.93	9.50E-01	3.11E-01	2.931E-01
					RV3130C	888841	diacylglycerol O-acyltransferase(tgs1)							
					RV3088	888669	diacylglycerol O-acyltransferase(tgs4)							
GOTERM_BP_DIRECT	GO:0019432~triglyceride biosynthetic process	3	3.896	4.59E-02	RV3087	888653	diacylglycerol O-acyltransferase(Rv3087)	44	18	2226	8.43	9.64E-01	3.11E-01	2.931E-01
					RV3130C	888841	diacylglycerol O-acyltransferase(tgs1)							
					RV3088	888669	diacylglycerol O-acyltransferase(tgs4)							
GOTERM_BP_DIRECT	GO:0006071~glycerol metabolic process	3	3.896	6.60E-02	RV3087	888653	diacylglycerol O-acyltransferase(Rv3087)	44	22	2226	6.90	9.92E-01	3.35E-01	3.157E-01
					RV3130C	888841	(tgs1)							
					RV3088	888669	(tgs4)							

Table 0.9: KEGG Pathway enrichment analysis of EMB->RIF vs Medium TP2 upregulated DEGs, Chapter 4.2.2

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Ben- ja- mini	FDR
KEGG_PATH-WAY	mtu00561: Glyc- erolipid metabo- lism	3	3.896	4.59E-02	RV3087	888653	diacylglycerol O-acyltransfer- ase(Rv3087)	14	30	1109	7.92E+00	6.76E- 01	8.21E- 01	8.21E- 01
					RV3130C	888841	diacylglycerol O-acyltransfer- ase(tgs1)							
					RV3088	888669	diacylglycerol O-acyltransfer- ase(tgs4)							
KEGG_PATH-WAY	mtu01040: Bio- synthesis of un- saturated fatty acids	2	2.597	6.85E-02	RV3229C	888821	stearoyl-CoA 9-desatu- rase(desA3)	14	6	1109	2.64E+01	8.18E- 01	8.21E- 01	8.21E- 01
					RV3230C	888748	stearoyl-CoA 9-desaturase electron transfer (Rv3230c)							

Table 0.10: GO analysis of RIF vs Medium 8 h (TP1) upregulated DEGs, Chapter 4.2.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
Annotation Cluster 1	Enrichment Score 2.45													
UP_KW_CELLULAR_COMPOSITION	KW-0964~Secreted	6	15.79	9.11E-04	RV2615C RV1818C RV0285 RV0287 RV0278C RV0980C	888215 885551 886608 886604 886623 885327	PE_PGRS45(PE_PGRS45) PE_PGRS33(PE_PGRS33) PE family protein PE5(PE5) ESAT-6 like protein EsxG(esxG) PE_PGRS3(Rv0278c) PE_PGRS18(PE_PGRS18)	9	157	1326	5.63E+00	7.26E-03	7.28E-03	7.28E-03
GOTERM_CC_DIRECT	GO:0005576~extracellular region	8	21.05	2.30E-03	RV2615C RV0278C RV0291 RV0285 RV0287 RV1818C RV0980C RV1759C	888215 886623 886615 886608 886604 885551 885327 885325	PE_PGRS45(PE_PGRS45) PE_PGRS3(Rv0278c) membrane-anchored mycosin MycP(mycP3) PE family protein PE5(PE5) ESAT-6 like protein EsxG(esxG) PE_PGRS33(PE_PGRS33) PE_PGRS18(PE_PGRS18) Wag22(wag22)	14	428	2574	3.44E+00	2.95E-02	2.99E-02	2.99E-02
UP_KW_BIOLOGICAL_PROCESS	KW-0843~Virulence	6	15.79	2.77E-03	RV2615C RV1818C RV0285 RV0287	888215 885551 886608 886604	PE_PGRS45(PE_PGRS45) PE_PGRS33(PE_PGRS33) PE family protein PE5(PE5) ESAT-6 like protein EsxG(esxG)	11	164	1446	4.81E+00	1.65E-02	1.66E-02	1.66E-02

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bonfer- roni	Benja- mini	FDR
UP_KW_CELLU- LAR_COMPO- NENT	KW-0134~Cell wall	4	10.53	5.05E-03	RV0278C	886623	PE_PGRS3(Rv0278c)	9	64	1326	9.21E+00	3.97E-02	2.02E-02	2.02E-02
					RV0980C	885327	PE_PGRS18(PE_PGRS18)							
					RV2615C	888215	PE_PGRS45(PE_PGRS45)							
					RV1818C	885551	PE_PGRS33(PE_PGRS33)							
					RV0278C	886623	PE_PGRS3(Rv0278c)							
GOTERM_CC_DI- RECT	GO:0009986~cell sur- face	3	7.89	1.89E-02	RV0980C	885327	PE_PGRS18(PE_PGRS18)	14	43	2574	1.28E+01	2.20E-01	1.23E-01	1.23E-01
					RV1818C	885551	PE_PGRS33(PE_PGRS33)							
					RV0285	886608	PE family protein PE5(PE5)							
					RV0278C	886623	PE_PGRS3(Rv0278c)							
Annotation Cluster 2	Enrichment Score: 0.11402223169208481													
GOTERM_CC_DI- RECT	GO:0009274~pepti- doglycan-based cell wall	5	13.16	4.57E-01	RV1818C	885551	PE_PGRS33(PE_PGRS33)	14	675	2574	1.36E+00	1.00E+00	1.00E+00	1.00E+00
					RV0290	886599	ESX-3 secretion system protein EccD(eccD3)							
					RV1569	886345	8-amino-7-oxononano- ate synthase(bioF1)							
					RV1450C	886605	PE_PGRS27(PE_PGRS27)							
					RV0291	886615	membrane-anchored mycosin MycP(mycP3)							
UP_KW_CELLU- LAR_COMPO- NENT	KW-1003~Cell mem- brane	3	7.89	8.61E-01	RV2615C	888215	PE_PGRS45(PE_PGRS45)	9	492	1326	8.98E-01	1.00E+00	1.00E+00	1.00E+00
					RV0290	886599	ESX-3 secretion system protein EccD(eccD3)							
					RV0291	886615	membrane-anchored mycosin MycP(mycP3)							

[illegible]

Table 0.11: KEGG Pathway enrichment analysis of RIF vs Medium 8 h (TP1) upregulated DEGs, Chapter 4.2.2

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List To- tal	Pop Hits	Pop To- tal	Fold En- richment	Bon-fer- roni	Benja- mini	FDR
KEGG_PATH- WAY	mtu01240: Bio- synthesis of co- factors	3	7.89	7.28E- 02	RV1569	886345	8-amino-7-oxononanoate syn- thase(bioF1)	5	133	1109	5.00E+00	4.11E- 01	4.33E-01	4.33E- 01
					RV2207	887781	nicotinate-nucleotide-dime- thylbenzimidazol phosphoribosyl- transferase(cobT)							
					RV2677C	887711	protoporphyrinogen oxi- dase(hemY)							

Table 0.12: GO analysis of RIF-EMB vs Medium TP2 upregulated DEGs, Chapter 4.2.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
Annotation Cluster 1	Enrichment Score 3.24													
GOTERM_MF_DIRECT	GO:0051082~unfolded protein binding	5	14.71	8.19E-06	RV0351	886497	stress response protein GrpE(grpE)	30	13	2612	3.35E+01	3.85E-04	3.85E-04	3.85E-04
					RV1736C	885213	nitrate reductase-like protein NarX(narX)							
					RV0350	885946	chaperone protein DnaK(dnaK)							
					RV0251C	886667	heat shock protein(hsp)							
					RV2031C	887579	alpha-crystallin(hspX)							
GOTERM_BP_DIRECT	GO:0009408~response to heat	4	11.76	3.23E-04	RV0350	885946	chaperone protein DnaK(dnaK)	21	16	2226	2.65E+01	1.57E-02	5.16E-03	4.41E-03
						888580	RNA polymerase sigma factor SigB(sigB)							
					RV2710	886667	heat shock protein(hsp)							
					RV2031C	887579	alpha-crystallin(hspX)							
GOTERM_BP_DIRECT	GO:0006457~protein folding	3	8.82	7.55E-04	RV0351	886497	stress response protein GrpE(grpE)	21	5	2226	6.36E+01	3.63E-02	9.06E-03	7.74E-03
					RV0251C	886667	heat shock protein(hsp)							
					RV2031C	887579	alpha-crystallin(hspX)							
UP_KW_BIOLOGICAL_PROCESS	KW:0346~Stress response	5	14.71	1.04E-03	RV0351	886497	stress response protein GrpE(grpE)	17	44	1446	9.67E+00	1.76E-02	1.77E-02	1.77E-02
					RV0350	885946	chaperone protein DnaK(dnaK)							

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
Annotation Cluster 1	Enrichment Score 3.24													
UP_KW_MOLECULAR_FUNCTION	KW-0143~Chaperone	3	8.82	2.90E-02	RV2710 RV0251 C RV2031 C RV2031 C RV0351 RV0350	888580 886667 887579 887579 886497 885946	RNA polymerase sigma factor SigB(sigB) heat shock protein(hsp) alpha-crystallin(hspX) alpha-crystallin(hspX) stress response protein GrpE(grpE) chaperone protein DnaK(dnaK)	19	30	1989	1.05E+01	2.76E-01	3.48E-01	3.48E-01
Annotation Cluster 2	Enrichment Score: 0.17791965619286093													
Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
UP_KW_BIOLOGICAL_PROCESS	KW-0805~Transcription regulation	3	8.82	5.98E-01	RV0081 RV2710 RV1129 C	887012 888580 885963	HTH-type transcriptional regulator(Rv0081) RNA polymerase sigma factor SigB(sigB) transcriptional regulator(Rv1129c)	17	176	1446	1.45E+00	1.00E+00	1.00E+00	1.00E+00

Table 0.13: GO analysis of RIF-EMB vs Medium TP3 upregulated DEGs, Chapter 4.2.2.

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
Annotation Cluster 1	Enrichment Score: 4.06													
GOTERM_BP_DIRECT	GO:0071770~DIM / DIP cell wall layer assembly	7	16.28	1.08E-07	RV2933	887686	PpsC(ppsC)	19	33	2226	2.49E+01	3.89E-06	3.78E-06	3.13E-06
					RV2932	888023	PpsB(ppsB)							
					RV2935	888210	PpsE(ppsE)							
					RV1662	885527	polyketide synthase(pks8)							
					RV2934	887172	PpsD(ppsD)							
					RV1663	885523	polyketide synthase(pks17)							
							phthiocerol / phthiodiolone dimycocerosyl transferase(papA5)							
GOTERM_MF_DIRECT	GO:0004312~fatty acid synthase activity	6	13.95	1.37E-07	RV2939	887327						3.55E-06	3.55E-06	3.55E-06
					RV2933	887686	PpsC(ppsC)	17	23	2612	4.01E+01			
					RV2932	888023	PpsB(ppsB)							
					RV2935	888210	PpsE(ppsE)							
					RV1662	885527	polyketide synthase(pks8)							
					RV2934	887172	PpsD(ppsD)							
					RV1663	885523	polyketide synthase(pks17)							
GOTERM_MF_DIRECT	GO:0004315~3-oxoacyl-[acyl-carrier-protein] synthase activity	5	11.63	4.30E-06	RV2933	887686	PpsC(ppsC)	17	20	2612	3.84E+01	1.12E-04	5.58E-05	5.58E-05
					RV2932	888023	PpsB(ppsB)							
					RV2935	888210	PpsE(ppsE)							
					RV1662	885527	polyketide synthase(pks8)							

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
GOTERM_MF_DIRECT	GO:0031177~phosphopantetheine binding	5	11.63	9.28E-06	RV2934	887172	PpsD(ppsD)	17	24	2612	3.20E+01	2.41E-04	8.05E-05	8.05E-05
					RV2933	887686	PpsC(ppsC)							
					RV2932	888023	PpsB(ppsB)							
					RV2935	888210	PpsE(ppsE)							
					RV2934	887172	PpsD(ppsD)							
GOTERM_BP_DIRECT	GO:0006633~fatty acid biosynthetic process	6	13.95	7.25E-05	RV1663	885523	polyketide synthase(pks17)	19	59	2226	1.19E+01	2.61E-03	1.27E-03	1.05E-03
					RV2933	887686	PpsC(ppsC)							
					RV2932	888023	PpsB(ppsB)							
					RV2935	888210	phthiocerol synthesis polyketide							
					RV1662	885527	(pks8)							
GOTERM_BP_DIRECT	GO:0097041~phenolic phthiocerol biosynthetic process	3	6.98	6.09E-04	RV2934	887172	PpsD(ppsD)	19	5	2226	7.03E+01	2.17E-02	7.10E-03	5.89E-03
					RV2933	887686	PpsC(ppsC)							
					RV2932	888023	PpsB(ppsB)							
GOTERM_BP_DIRECT	GO:0097040~phthiocerol biosynthetic process	3	6.98	9.09E-04	RV2934	887172	PpsD(ppsD)	19	6	2226	5.86E+01	3.22E-02	7.95E-03	6.59E-03
					RV2933	887686	PpsC(ppsC)							
					RV2932	888023	PpsB(ppsB)							
GOTERM_BP_DIRECT	GO:0044550~secondary metabolite biosynthetic process	3	6.98	8.75E-03	RV2934	887172	PpsD(ppsD)	19	18	2226	1.95E+01	2.71E-01	6.13E-02	5.08E-02
					RV2933	887686	PpsC(ppsC)							
					RV2932	888023	PpsB(ppsB)							

Table 0.14: GO analysis of RIF-EMB vs Medium TP3 downregulated DEGs, Chapter 4.2.2

Category	Term	Count	%	P-Value	Locus	DAVID ID	Gene name	List Total	Pop Hits	Pop Total	Fold En-rich-ment	Bonfer-roni	Benja-mini	FDR
GOTERM_BP_DIRECT	GO:0009408~re-sponse to heat	2	11.76	4.24E-02	Rv2745c	888315	clgR	7	16	2226	39.75	6.90E-01	1	1
					Rv0251c	886667	hsp							

