JOURNAL OF MEDICAL MICROBIOLOGY

**JMM PROFILE**

Hardie and Fenn, *Journal of Medical Microbiology* 2022;71:001566

DOI 10.1099/jmm.0.001566

MICROBIOLOGY SOCIETY

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# JMM profile: rifampicin: a broad-spectrum antibiotic

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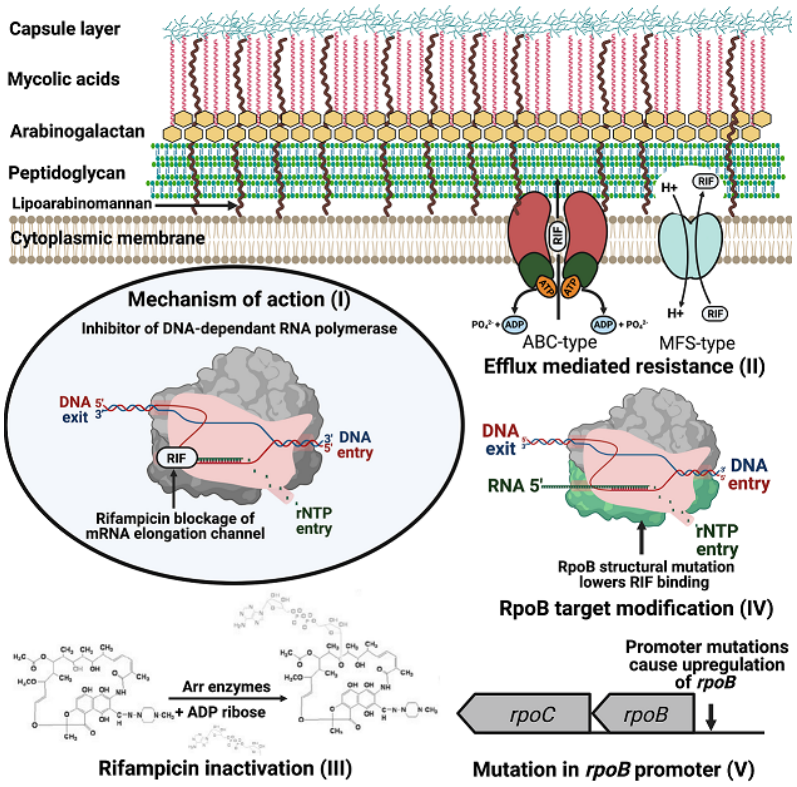


### Graphical abstract

Rifampicin mode of action and resistance mechanisms. After entering bacteria, rifampicin binds to the $\beta$-subunit of RNA polymerase (RpoB) to inhibit RNA synthesis at an early point (between the addition of the second or third nucleotide). Resistance to rifampicin (depending on species) can occur via (i) ADP ribosylation of rifampicin so that it no longer interacts with RNA polymerase (due to being transmissible, black cross on red circle), (ii) efflux (yellow efflux pump), (iii) mutation of RNA polymerase so that it cannot interact with rifampicin (most clinically significant), or (iv) mutation of the*rpoB* promoter that raises the production levels of RNA polymerase thereby outnumbering the rifampicin and leaving some of the polymerase free to function.

### Abstract

Rifampicin (also known as rifampin) inhibits RNA synthesis, and is used to treat tuberculosis, leprosy, staphylococcal infections and legionnaires' disease. It can also protect at-risk populations from *Haemophilus influenzae* type b and *Neisseria meningitidis*. It is a polyketide antibiotic and is on the World Health Organization (WHO) list of essential medicines due to its critical importance to human medicine. The adverse effect of liver toxicity is controlled by testing during prolonged treatment regimes. Rifampicin's red–orange colour can result in the colouration of sweat, tears and urine. Resistance to rifampicin arises from mutation of the target RNA polymerase or ADP ribosylation of the antibiotic or efflux. Mycobacteria may become singularly resistant to rifampicin or as part of multidrug or extensive drug resistance.

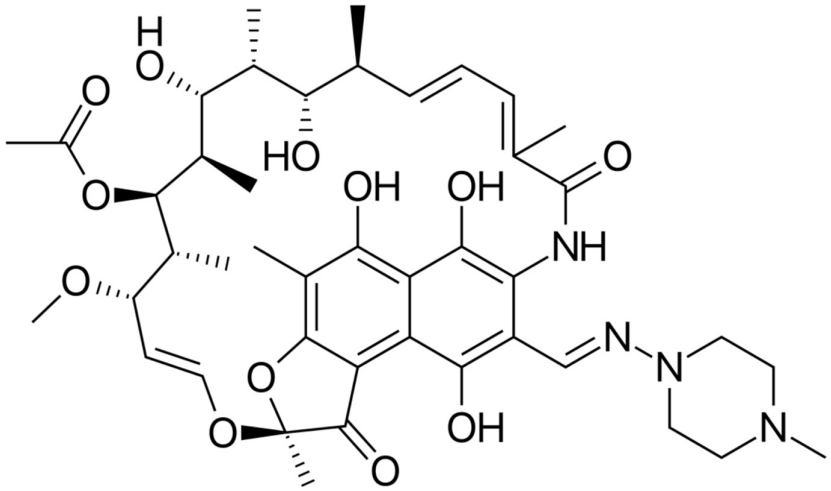


**Fig. 1.** The chemical structure of rifampicin.

## HISTORICAL PERSPECTIVE

Rifamycin compounds were isolated from a Gram positive-bacterium, *Amycolatopsis rifamycinica* (previously referred to as *Streptomyces mediterranei* and *Nocardia mediterranei*), that was grown from French soil in 1957. The more stable semisynthetic rifampicin was produced in 1965. The higher efficacy and good tolerability enabled rifampicin to be marketed in Italy in 1965 and approved by the US Food and Drug Administration (FDA) in 1971. Other rifamycin compounds include rifabutin, rifapentine, rifalazil and rifaximin. Rifampicin is in a select category of agents that are active against slow-growing or non-replicating *Mycobacterium tuberculosis*.

## STRUCTURE/CHEMISTRY

Rifampicin is a polyketide with a naphthoquinone core spanned by an aliphatic ansa chain (Fig. 1). Its red–orange crystalline colour derives from the naphthoquinone chromophore, and it belongs to the chemical class of ansamycin compounds. Rifampicin is the 3-(4-methyl-1-piperazinyl)-aminomethyl derivative of rifamycin, an antibiotic naturally synthesized by the bacterium *Amycolatopsis rifamycinica*. Side chains at positions 3 and/or 4 of the naphthyl moiety of rifamycinsare found inrifabutin, rifapentine, rifalazil and rifaximin. For instance, whilst rifampicin has a methyl group, rifapentine has a cyclopentane ($C_5H_9$) group at position 3 and there is a cyclic spiro-piperidyl side chain at positions 3 and 4 of ribabutin.

## MODE OF ACTION

Rifampicin enters the body following oral administration easily, as it is generally well absorbed from the gastroeintestinal tract, from where it distributes throughout the body, creating a bioavailability in the region of 90%. It enters the bacteria via passive diffusion. Rifampicin targets DNA-dependent RNA polymerase to inhibit RNA synthesis (graphical abstract). Four hydroxyl groups of the rifampicin ansa bridge and naphthol ring form hydrogen bonds with amino acids in RNA polymerase. This binding occurs within the DNA/RNA channel of the RNA polymerase $\beta$-subunit (RpoB). Rifampicin binds a distance from the active site of RNA polymerase. The physical interaction blocks RNA elongation. The RNA transcripts are truncated after two or three nucleotides as the phosphodiester bond formation in RNA is prevented, halting downstream bacterial protein synthesis. Rifampicin reaches the urine, saliva, sweat, tears, sputum and faeces, where it imparts an orange/red colour. It is also secreted in bile and can be recirculated, which prolongs its half-life. *M. tuberculosis* can persist in some people, despite the strain being

Received 20 October 2021; Accepted 30 May 2022; Published 05 August 2022
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Keywords: antimicrobial resistance; rifampin; tuberculosis; RNA synthesis.
Abbreviations: ADP, adenosine diphosphate; AMR, antimicrobial resistance; DNA, deoxyribonucleic acid; FDA, Food and Drug Agency; MIC, minimum inhibitory; MTB, *Mycobacterium tuberculosis*; RNA, ribonucleic acid; RRDR, rifampicin resistance-determining region; USA, United States of America; WHO, World Health Organization.

susceptible to rifampicin. This drug non-responsiveness could emanate from rifampicin regulating host factors such as xenobiotic sensing nuclear receptors that in turn modulate drug efflux [1, 2]. Rifapentine and rifabutin do not activate the nuclear receptors, and may be more effective treatments in this scenario.

## MECHANISMS OF MICROBIAL RESISTANCE

Resistance against rifampicin [3, 4] can arise via three routes: (i) alteration of the target protein RNA polymerase via mutation of *rpoB* at a variety of sites or in the promoter to enable overproduction [5], (ii) ADP ribosylation of the rifampicin molecule itself and (iii) efflux (Fig. 1). The frequency of laboratory selection of spontaneous rifampicin-resistant mutants ranges from $10^{-10}$ to $10^{-7}$, depending on the organism and the methodology used [3]. The resistance can develop alone or in combination with resistance against additional antimicrobials, but may incur a fitness disadvantage given that *rpoB* is an essential gene.

RpoB mutations arise in *Escherichia coli*, *Staphylococcus aureus* and *M. tuberculosis*, and the residues mutated are predominantly in hotspots conserved across the species known as the rifampicin resistance-determining region (RRDR). Most are point mutations, although small insertions (1–2 codons) and deletions (2–3 codons) have been identified. Mutations in *rpoB* have also been reported in clinical isolates of *Haemophilus influenzae*, *Streptococcus pneumoniae* and *Neisseria meningitidis*. A few species are intrinsically resistant to rifampicin due to a refractory RpoB, including spirochetes and mollicutes. Partial cross-resistance resulting from mutation of *rpoB* has been reported in tuberculosis patients between rifampicin, rifabutin and rifapentine, whilst complete cross-resistance has been reported between the same drugs for *S. aureus* [3, 6].

The use of covalent modification (ADP ribosylation) to inactivate rifampicin has been reported in mycobacteria. The gene for an enzyme (termed ARR-2) capable of ADP ribosylating rifampicin has been identified on an integron in a *Pseudomonas aeruginosa* strain. Although the origin of Arr and the mechanism of its entry into mycobacteria remain unclear, the possibility of transfer of ADP ribosylases on plasmids raises a significant public health concern [7].

The clinical implications of changes in efflux are not evident, although they have been reported to occur in mycobacteria. The genome of the opportunistic pathogen *Nocardia farcinica* has duplicated *rpoB*, which would raise the amount of target to dilute out the effectiveness of the antibiotic.

Resistance to rifampicin develops quickly if it is administered alone, but as part of combination therapy or over a short period this resistance development is decreased. The evolution of rifampicin resistance has been prompted by delivering substandard drugs containing degradation products [8]. However, there is hope for the future as alternative ansamycin natural products are being discovered that are active against rifampicin-resistant *M. tuberculosis* [9]. Enterobacteriaceae, *Acinetobacter* and *Pseudomonas* species are intrinsically resistant to rifampicin. The resistance in these species originates from exclusion by the outer membrane. Moreover, rifaximin does not encourage the emergence of resistance in mycobacteria, which may be due to rifaximin not being absorbed in the gastrointestinal tract, and thus being present at very low systemic levels that are subselective [10].

## CLINICAL EFFICACY AND CONTRAINDICATIONS

### Target organisms

Rifampicin is only used for bacterial infections, and is restricted to the treatment of mycobacterial infections and a few other infections with specific indications. It is bactericidal and acts on both intracellular and extracellular bacteria. It is the first-line treatment for *Mycobacterium* spp. (*M. tuberculosis*, *M. avium* complex and *M. leprae*) and *Legionella pneumophila* (see Fig. 2).

Prophylactic treatment of individuals at risk of infection by *N. meningitidis* or *H. influenzae* type b. It can also be used as an alternative treatment for tick-borne pathogens, including *Borrelia burgdorferi* or *Anaplasma phagocytophilum*. Antimicrobial susceptibility testing is advised before treatment of *Listeria*, *Neisseria gonorrhoeae, H. influenzae* and *L. pneumophila* with rifampicin. In combination with other antibacterials, it can be used to treat brucellosis, legionnaires' disease and serious staphylococcal infections. A rifamycin closely related to rifaximin is also approved in the USA for the treatment of travellers' diarrhoea in some circumstances.

### Route of delivery

Rifampicin can be administered orally and intravenously.

### Dosage (general guidelines)

The dosage and duration depend on the age and condition being treated. It is effective against tuberculosis in combination with other antibiotics, including pyrazinamide, isoniazid and ethambutol. Because this treatment is lengthy, taken daily for over 6 months, compliance is sometimes problematic. Rifampicin should be taken an hour before meals or 2 h after meals, as it is more effective on an empty stomach with a glass of water. Shorter treatment regimens are being investigated to reduce the drivers

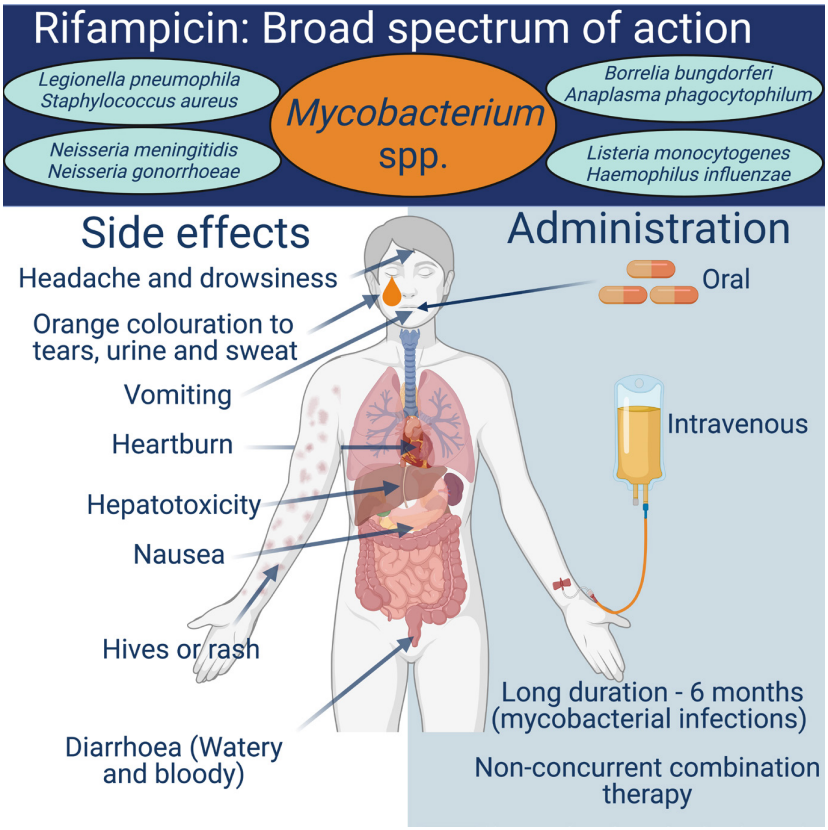


**Fig. 2.** The spectrum, side effects and administration of rifampicin.

of resistance to rifampicin [11], and novel combinations are being investigated as potential strategies to utilize rifampicin as a broader spectrum antimicrobial [2].

### Adverse effects/Toxicity

The side effects of rifampicin are too numerous to list and include the more serious nausea, vomiting, thromboctopenia, diarrhoea, rash and loss of appetite. In addition, tears, sweat and urine may develop an orange colouration.

Rifampicin induces hepatic cytochrome P450 enzymes, leading to liver toxicity, and therefore liver function tests are advised before and during treatment.

### Contraindications

Rifampicin increases the metabolism of certain drugs, making them ineffective.

- Patients undergoing warfarin anticoagulation therapy have to increase the dosage and have their clotting time monitored regularly.
- Unintended pregnancies due to the reduced efficacy of hormonal contraception.
- Anti-retroviral agents (e.g.everolimus, atorvastatin, rosiglitazone, pioglitazone, celecoxib and lorazepam), antifungals (caspofungin and vorconazole) and antibiotics (clarithromycin, gentamicin, amikacin).
- Note: there is uncertainty about synergistic roles with other agents due to the lack of large, robust clinical trials, e.g. vancomycin treatment of methicilin-resistant *S. aureus* (MRSA).

## OPEN QUESTIONS

(1) What are the full implications of *M. tuberculosis* resistance to rifampicin on future therapies? Do *rpoB* mutations concomitantly reduce *M. tuberculosis* fitness or alter disease pathology, transmissibility, or colonization?

(2) Are there additional rifampicin combination therapies that could be beneficial or detrimental?
(3) Are there further One Health uses for rifampicin?
(4) Can the side effects of rifampicin be minimized?
(5) Are there more polyketides to be discovered that can target other bacteria?
(6) Which form of *M. tuberculosis* is targeted by rifampicin – granuloma or another?

**Disclaimer**

This article has presented information in line with UK guidelines, take reference from the Green book and the British National Formulary for the current status.

Funding information
Wellcome Trust Doctoral Training Programme in Antimicrobials and Antimicrobial Resistance provided the funding (grant reference: 108876/Z/15/Z).

Acknowledgements
The figures were rendered using Biorender.com.

Author contributions
K.R.H. and S.J.F.: contributed to the writing and S.J.F.: compiled all the figures.

Conflicts of interest
The authors declare that there are no conflicts of interest.

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