Journal of Scientific & Industrial Research
Vol. 66, March 2007, pp. 191-208

Emerging trends in tuberculosis therapy—A review

Yadvendra K Agrawal[1]*, Hardik G Bhatt[1], Hitesh G Raval[2], Pratik M Oza[2], Hitesh B Vaidya[1], Kuntal Manna[1] and Parthajyoti Gogoi[1]

[1]Institute of Pharmacy, NIRMA University, S G Highway, Chharodi, Ahmedabad 382 481
[2]Anand Pharmacy College, Opposite Town Hall, Anand 388 001

Received 31 May 2006; revised 18 September 2006; accepted 30 October 2006

Tuberculosis is one of the most devastating bacterial disease having high rates of morbidity and mortality. *Mycobacterium tuberculosis* invades the host immune system and persists in pulmonary granulomas. Review presents bacterial cell wall or envelope structure, drug targets against *M. tuberculosis* and novel therapeutics agents like quinolone, pyrimidine, nitroimidazopyran and oxazolidinone derivatives. This review also focuses on the newer untouched targets like immunomodulators and isocitrate lyase inhibitors, which can be further exploited.

Keywords: Antimycobacterial agents, Cell envelope, Drugs, MIC, *Mycobacterium tuberculosis*, Therapeutic agents, Targets

Introduction

Tuberculosis (TB) is characterized as a chronic bacterial infection caused by *Mycobacterium tuberculosis,* an aerobic acid-fast bacillus (AFB). Tuberculosis killed about 2 million people every year and the epidemic, which is spreading globally, is assuming alarming proportions. Around 8 million people become infected with TB every year. The WHO *'Fact Sheet'* on TB estimates that between 2000 and 2020, nearly one billion people will get sick and 35 million will die from TB. In India alone, one person dies because of TB every minute. Indian population (approx 50%) reports positive tuberculin test. Every year, about 0.4 million deaths and millions of new cases of TB are reported. Due to demographic factor, socio-economic trends, neglected TB control in many countries and HIV (Human-immuno deficiency virus) infection, this epidemic has been able to adopt such proportion. Efforts to stop these frightening trends are hampered by the lack of financial resources in developing countries, the appearance of the multi-drug resistant strains of *M. tuberculosis* and bad therapy compliance[1-3]. The various pathogenic mycobacterium species are *M. tuberculosis*, *M. scrofulaceium*, *M. Arcanum*, *M. leprae*, *M. kanasasii* and *M. avium-intracellulare complex* (MAC)[4].

*Author for correspondence
Tel: +91-2717-241900-04; Fax: +91-2717-241916
E-mail: drykagrawal@yahoo.com

The review updates on newer molecules, which are synthesized as anti TB agents with their mode of action.

Targets for Antimycobacterial Agents

A key target for antimycobacterial chemotherapy is cell wall biosynthesis. Complex lipoglycan calyx on the mycobacterial cell surface provides a significant physical barrier to intracellular acting drugs. Due to lack of penetration, many antibiotics show no activity against *M. tuberculosis*. Inhibition of synthesis is lethal to bacterium as evidenced by the action of isoniazid and ethambutol. Recent deconvolution of biochemical events leading to cell wall formation has exposed a rich supply of targets[5-10]. The cell envelope of *M. tuberculosis*, which is unusually thick and waxy (Fig. 1), comprises four classes of polymer (peptidoglycan, arabinogalactan, mycolic acids and lipoarabinomannan)[11].

Tuberculosis Therapy

Chemotherapeutic agents[3,4,12-14] for TB may be divided into two main classes, first line agents (isoniazid, rifampicin, streptomycin, pyrazinamide and ethambutol) and second line agents (ehionamide, *p*-amino salicylic acid, cycloserine, rifapentine, clarythromycin, kanamycin, amikacin, ofloxacin, ciprofloxacin, viomycin and capreomycin). Some of

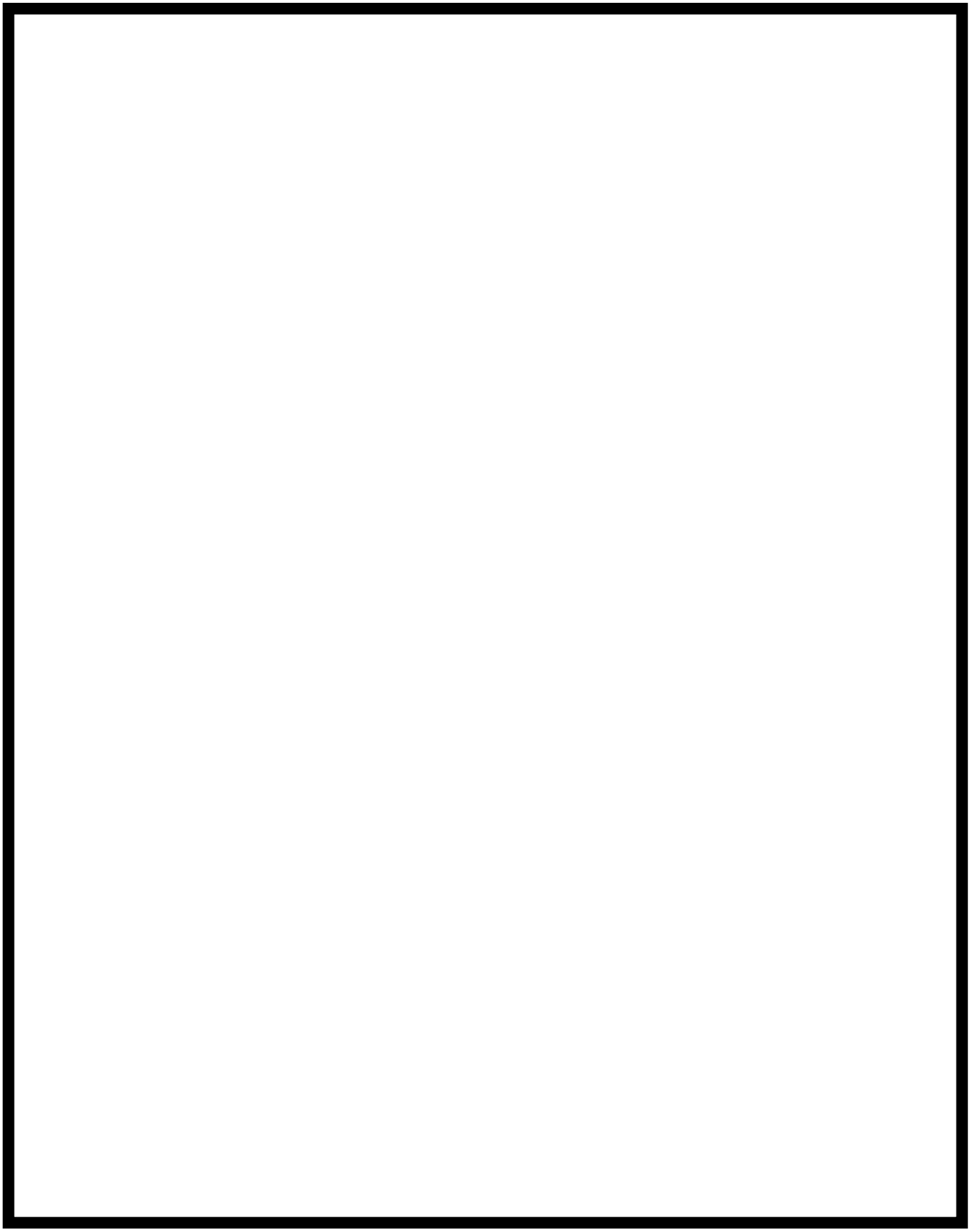


Fig. 1—Mycobacteria cell envelope[11]

the patented targets for antitubercular drugs (Fig. 2) are known to control various cellular processes required for survival or virulence of *M. tuberculosis*. Arrows (Fig. 2) show site of action of first line drugs[15]. To decreases possibility of the emergence of resistant organisms, compounded drug therapy is employed, involving the following: 1) A first phase (about 2 months) consisting of three drugs [isoniazid, rifampicin, pyrazinamide (plus ethambutol, if the organism is suspected to resistant)] used concomitantly; and 2) A second, continuation phase of 4 months, consisting of two drugs (isoniazid, rifampicin). Longer treatment needed in meningitis, bone or joint involvement, drug resistant case, etc.

The recommended treatment regimen is highly effective and rates of severe adverse reactions are low. However, unpleasant side effects and relatively long course of treatment are the drawbacks. Drug resistance and development of MDR-TB is also important. The second line drug used for MDR-TB is more expensive, less effective and more toxic than the four standard regimens. The goal, to develop bactericidal drugs, is a cost-effective manner, which effectively treats the infection of MDR strains of *M. tuberculosis* and latent infection. It is also important that they should have shortened treatment period or reduced frequency of doses.

Antitubercular Activity of Some Novel Drugs

Quinolones

Moxifloxacin (BAY 12-8039) **(1)**, an 8-methoxy-quinolone drug (MIC, 0.25 µg/ml), is reported active against *M. Tuberculosis*[16,17]. The elimination half-life of the drug in man (12 h) supports possibility of once a day treatment[18]. CS-940 **(2)**[19] shows antimycobacterial activity (IC50, 0.25-0.5 µg/ml) and is more potent than ofloxacin, ciprofloxacin and balofloxacin. PD 161148 **(3)**[20] is a third generation fluoroquinolone having potent antimycobacterial activity. Sitofloxacin (DU-6859a) **(4)** has outstanding activity against broad range of bacteria. The potency is believed to reside with its ability to equally inhibit both DNA gyrase and topoisomerase IV with lowest IC50 amongst quinolones[21]. Gemifloxacin (SB-265805) **(5)** is also found to be active[22]. 1-Substitutedaryl-6-fluoroquinolones **(6)**, at 6.25 µg/ml inhibits complete growth of *M. tuberculosis* with selective index[23] SI > 40. Mefloquine (4-aminoquinolinemethanol) **(7)** and its several analogues are active against a variety of bacteria including *Mycobacteria*[24]. WR-3016 **(8)** at 1 µg/ml and WR-3017 **(9)** at 2 µg/ml show potent inhibitory activity *in vitro* in *M. avium* complex (MAC) compared to 16 µg/ml for mefloquine[25].

Pyrimidines

3-N-[(substitutedaryl/heteroaryl) -methylene]-imino-2-methyl-5-thienyl-thieno-[2, 3-d]pyrimidin-4-(3H)-one **(10)**[26] at 2 - 40 µg/ml, 4-(4-chlorophenyl)-6-(4-methylphenyl)-2-aminopyrimidin **(11)**[27] at 6.25 µg/ml and 3-[4-(4-chlorophenyl)-6-(4-methylphenyl)pyrimidin-2yl]iminoisatin **(12)**[27] at 6.20 µg/ml give potent antitubercular activity against *M. tuberculosis* $H_{37}Rv$. 3'-C-branched-chain substituted nucleosides and nucleotides **(13)** are reported as potent inhibitors of *M. tuberculosis* thymidine monophosphate kinase[28]. Some thio analogue of purine **(14)** at 0.78-6.25 µg/ml proved as potent antimycobacterial agents against *M. tuberculosis* $H_{37}Rv$[29].

Nitroimidazopyrans

The 3-substituted nitroimidazopyrans (NAPs) have been screened for antimycobacterial activity[30,31]. In contrast to conventional anti-tubercular drugs, NAPs exhibit bactericidal activity against both replicating and stationary *M. tuberculosis* cells. PA-824 **(15)** at 0.015-0.25 µg/ml and PA-1343 **(16)** at 0.015 µg/ml are potent under clinical trial[32]. A novel lipophilic and orally active agent CGI 17341 (2-ethyl-5-nitro-2,3-dihydro[2-lb]imidazo-oxazole) **(17)**, whose MIC is not altered at pH 5.6 and since the combination of CGI 17341 and RIF is synergistic at a low pH, CGI 17341 may play a significant role in improving the overall sterilizing activity of drug regimens against *M. tuberculosis*[33].

Novel Antimycobacterial Agents with Known Mode of Action

Rhodamine derivative 5372 **(18)**, whose structural motif is similar to that used in 4-thiazolidinones, affects sugar nucleotide biosynthesis during peptidoglycan formation[34,35]. The series of alkyl-sulfonyl amides, which inhibit ß-ketoacyl synthase (KAS), are also the series of target-based drug design **(19)**[36]. Sulfonyl amides mimic putative tetrahedral transition state formed during KAS catalysis. KAS is one of the accessory fatty acid synthases peculiar to mycobacteria. Proteins of antigen 85 complex, which exhibit mycolyl transferase activity, are prime targets in mycolic acid biosynthesis[37]. These proteins

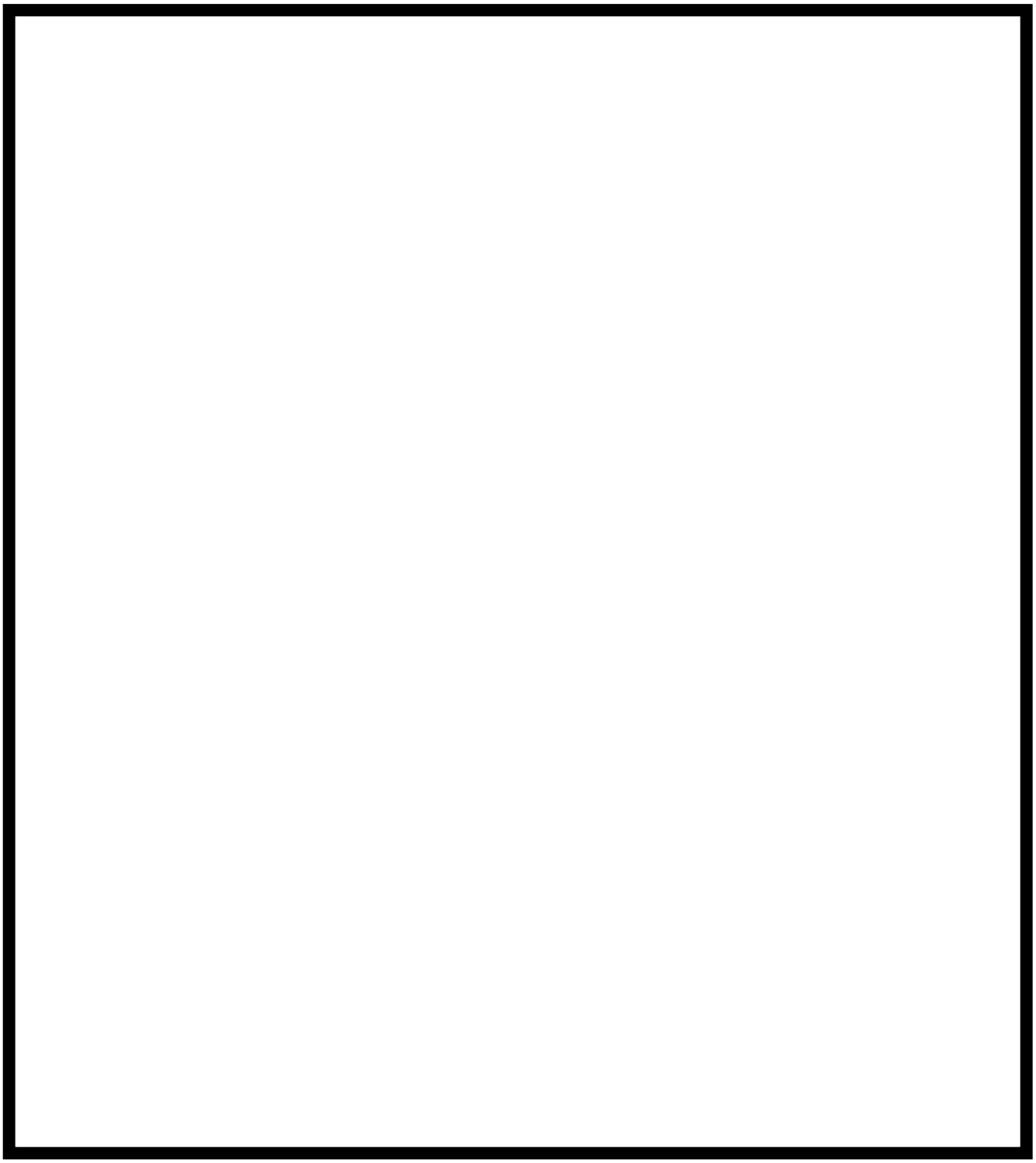
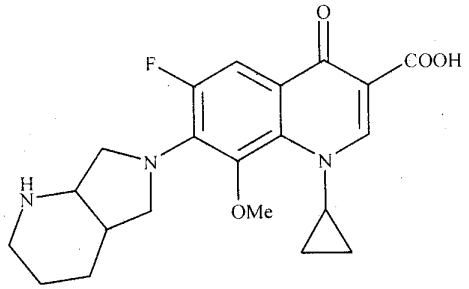
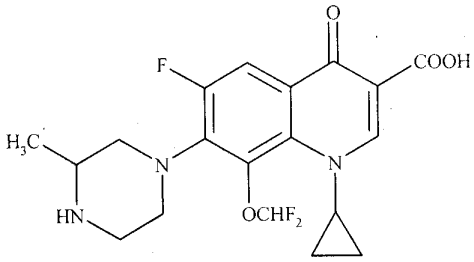


Fig. 2—Patent protected drug targets for *Mycobacterium tuberculosis*[15]

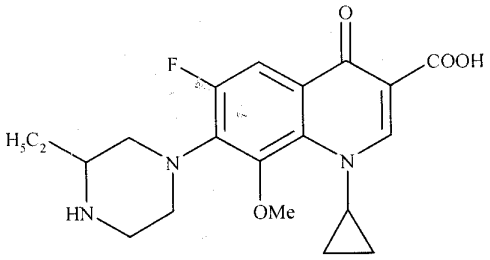
recognize and transfer mycolate from trehalose mono- and dimycolates. A family of 6,6'-diamino-6,6'-dideoxytrehalose-based derivatives with different alkylamines or alkysulfonamide functionality have been synthesized and found active against *M. tuberculosis* $H_{37}R_a$[38]. A potent new antimycobacterial activity with an MIC of 1.3 µg/ml is found in compound **(20)**.



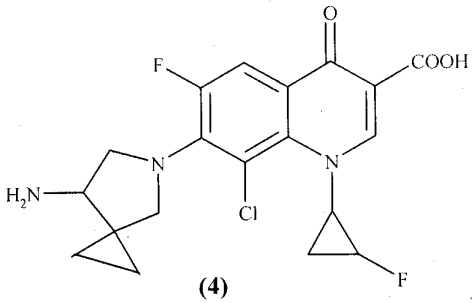
(1)



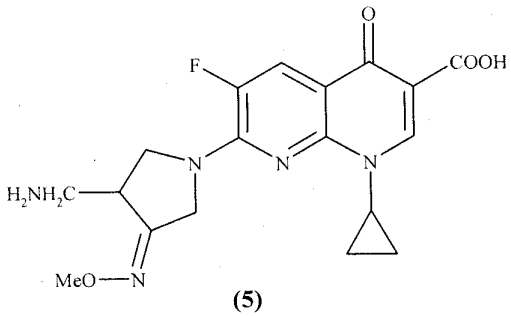
(2)



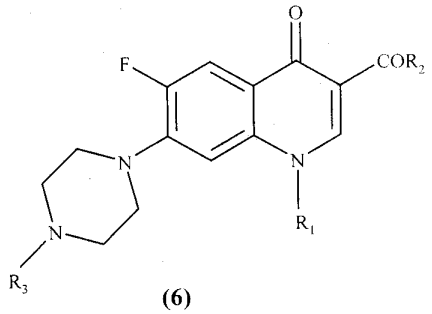
(3)



(4)



(5)

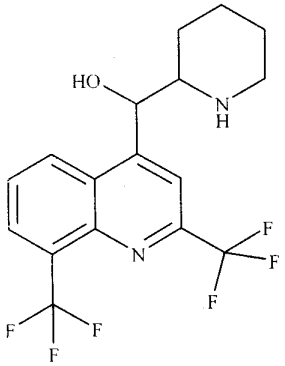


R_1 = 4-amino-2-fluorophenyl

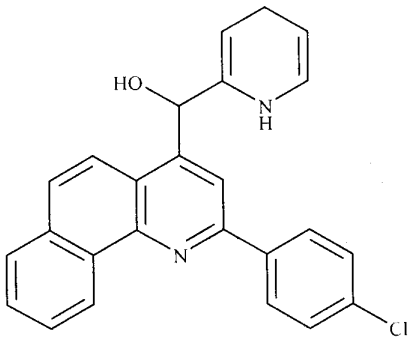
R_2 = OH

R_3 = H, CH_3

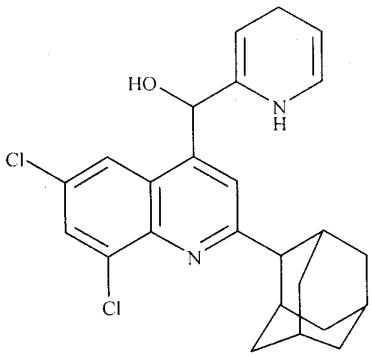
(6)



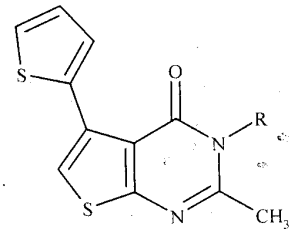
(7)



(8)



(9)

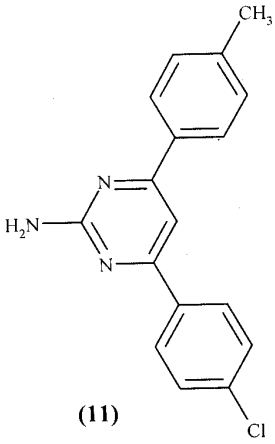


R = NH_2

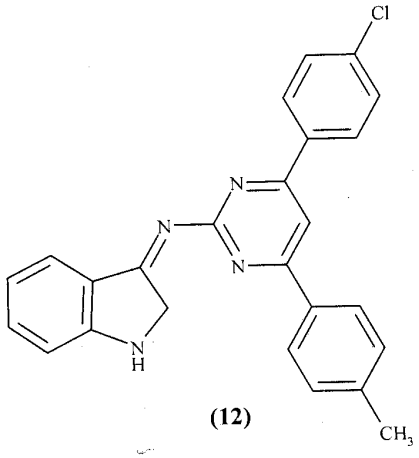
R = N=CHR_1

R_1 = 4-Cl-C_6H_4, 4-OH-C_6H_4, 3-pyridinyl, 2-F, 4-CF_3-C_6H_3

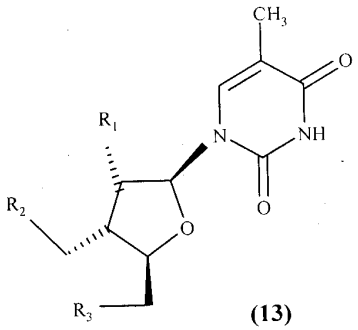
(10)



(11)



(12)

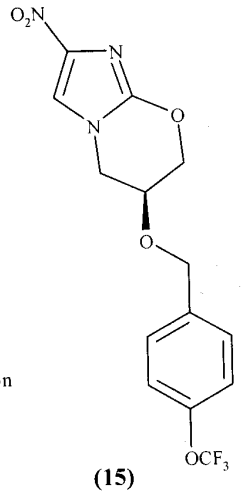


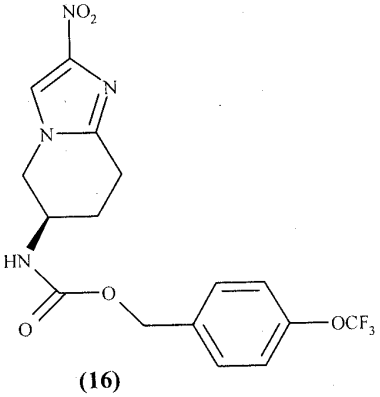
R_1 = H, OH

R_2 = OH, N_3, NH_2, F, CH_2OH

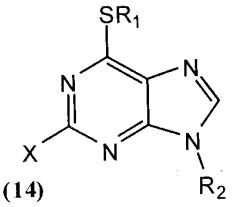
R_3 = OH, OPO_3-2

(13)





(15) **(16)**

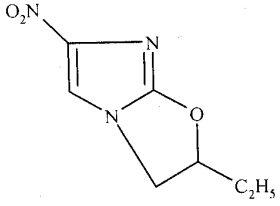


X = H, OH, Cl

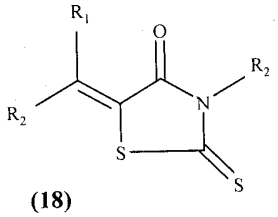
R_1 = Aliphatic, Aromatic substitution

R_2 = H, Carboxymethyl derivatives

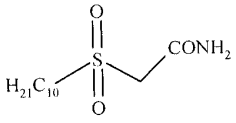
(14)



(17)



(18)



(19)

A drug based on the inhibition of arabinofuranosyl transferases does not produce mechanism-based toxicity as these units are not present in humans, hence they are proposed as potential targets for drug development[39]. Decaprenylphosphoarabinose is the probable donor molecule for the arabinose unit[40]. One of the *C*-phosphonate analogs **(21)** is active at MIC 3.13 µg/ml[41]. The enoyl acyl carrier protein reductase enzyme InhA is a good antimycobacterial target[42]. Indole-5-amides, 4-aryl-substituted piperazines, and various pyrazole derivatives provide useful core templates for good InhA inhibition[43]. Genz-8575 **(22)** at 2.5 µg/ml is a potent InhA inhibitor (91% inhibition at 40 µM) against $H_{37}R$.

A tubulin polymerase inhibitor, SRI-3072 **(23)**[44,45] at 0.15 µg/ml, is effective against the growth of *M. tuberculosis* and has good selectivity index[46] SI-42. It is also an inhibitor of FtsZ (a bacterial tubulin polymerase homologue). From a series of 2,4-diamino-5-deazapteridine derivatives, SRI-20094 **(24)** at 0.13 µg/ml displays potent inhibition of MM6 cells infected with MAC strain NJ3440 and shows excellent inhibition of dihydrofolate reductase (DHFR) of MAC with IC50 of 1.0 nM. SRI-20094 is of potential value for the treatment of *M. avium* infections and in particular for persons co-infected with HIV[47]. D-Alanine racemase is a cytoplasmic enzyme responsible for the conversion of L-alanine to D-alanine, which is a key building block in peptidoglycan biosynthesis. Inhibitors of this enzyme, such as D-cycloserine and fludalanine, possess potent antitubercular activity. 5-Amino-furanoside derivatives are potent inhibitors of this enzyme **(25)**[48]. Most active compound (MIC 3.12 µg/ml) of this series possesses reasonable antimycobacterial activity.

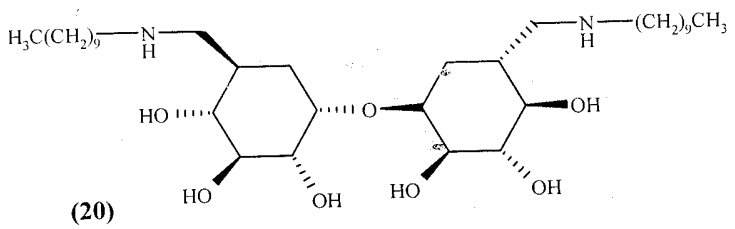
Sulfometuron methyl **(26),** a herbicide, has inhibitory activity against acetolactate synthase, which catalyses a key step in branched-chain amino acid biosynthesis[49]. 4-Thiazolidinone derivative **(27)**[50] possesses moderate antimycobacterial activity and shows good inhibitory activity against bacterial MurB enzyme, which converts UDP-GlcNAc into UDP-MurNAc, an intermediate in the assembly of MurNAc-pentapeptide for cell wall peptidoglycan biosynthesis. Compound **(28)** at MIC of 1.42 µg/ml with micromolar levels of monoamine oxidase inhibition shows potent antitubercular activity[51]. Compound R207910 **(29)**[52] at 0.03 to 0.12 µg/ml is extremely potent against a variety of mycobacterial species with no cross-resistance against a panel of different drug-resistant isolates. This is further supported by identification of the molecule's target as the proton pump for *M. tuberculosis* ATP synthase through genetic analysis of resistant mutants.

Novel Antimycobacterial Agents with Unknown Mode of Action

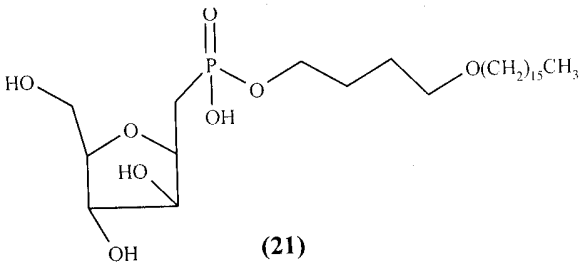
Compound **(30)** of the series 9-benzylpurines at 0.78 µg/ml has very good inhibitory activity against *M. tuberculosis* with moderate toxicity levels against Vero cell lines[53,54] (SI = 10.6), besides a potent antimycobacterial activity against strains resistant to INH, RIF, and EMB. Experiments with infected bone marrow macrophages also suggest that the compound is capable of attacking *M. tuberculosis* inside these cells[55]. A series of pyrrole derivatives show very good MIC of 0.4 µg/ml and a selectivity index[56] of 20. BM 212 **(31)** of pyrrole series, which has very good MIC of 0.7 µg/ml and moderate toxicity levels against Vero cells[57] (SI = 5.6), shows no significant cross-resistance with INH, RIF, EMB, and streptomycin with intracellular antimycobacterial activity in macrophage assays.

Oxidation state of sulfur atom has an impact on the MICs for resistant strains. 9-Sulfonylated or sulfenylated-6-mercaptopurines **(32)** show potent antimycobacterial activity at 0.39-3.39 µg/ml with very good selectivity index[58] (45-200). Compound **(33)**, α, β-unsaturated acyclic sugar ketones, possesses good antimycobacterial activity at 3.1 µg/ml[59,60]. Some of 1,2,4,5-tetraoxacycloalkanes **(34)** also possess notable antimycobacterial activity at 3.12 µg/ml[61]. Xanthone derivatives exhibit antitubercular activity[62]. Compound **(35)**[63] of this series shows antimycobacterial activity at 4 µg/ml. Compound **(36)** among 3-phenyl-6,8-dichloro-2*H*-1,3-benzoxazine-2,4(3*H*)-dithiones showed antitubercular activity[64, 65]. The best compound of the series showed moderate activity against mycobacteria with MIC of 4 µg/ml.

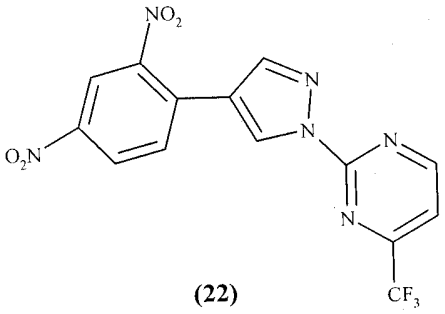
Widespread activity of quinoxaline-1-oxides is observed from enzymatic, single-electron reduction of quinoxaline 1,4-dioxides under hypoxic conditions leading to DNA damage[66]. Of this series, compound **(37)** shows a very good antitubercular activity at 0.1 µg/ml and selectivity towards mycobacteria[67-69] (SI > 125). A series of compounds containing an alkyl-mercaptan group attached to an electron-deficient carbon atom produce good antimycobacterial activity[70]; compound **(38)** at 1.3 µg/ml is most active. Alkyl-mercaptan functionality is



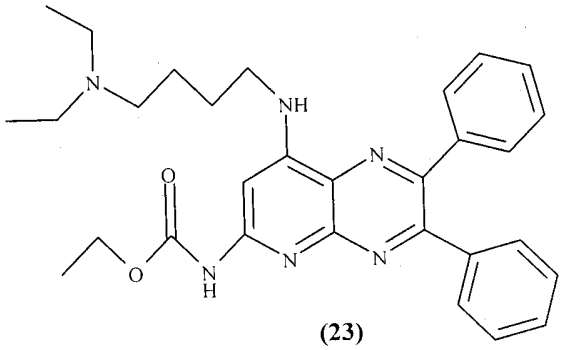
(20)



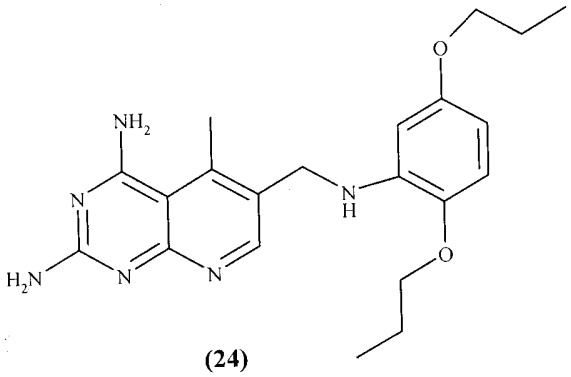
(21)



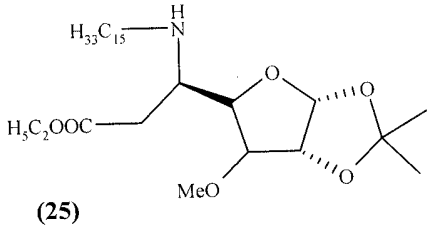
(22)



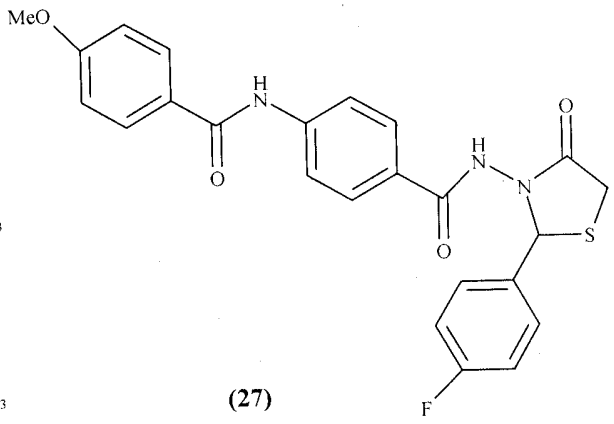
(23)



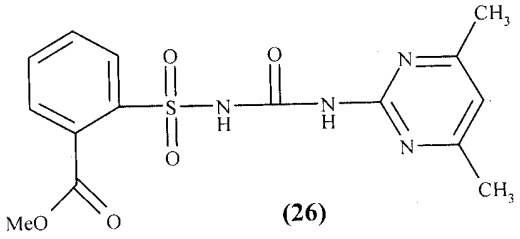
(24)



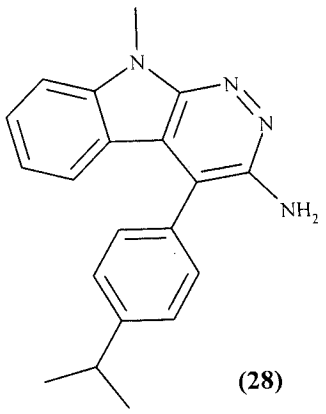
(25)



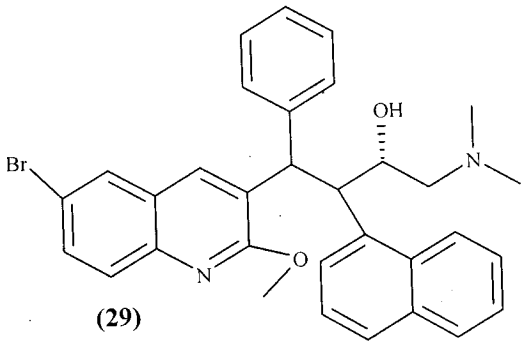
(27)



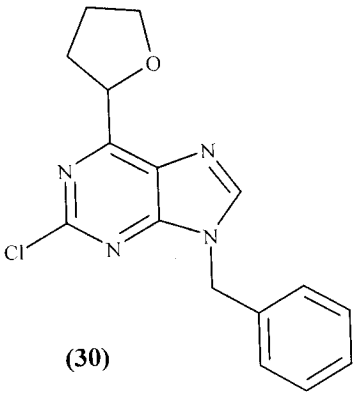
(26)



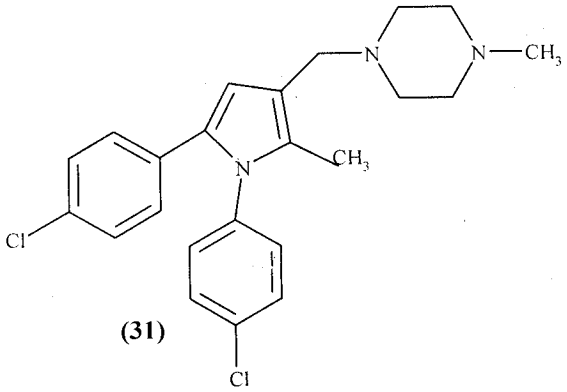
(28)



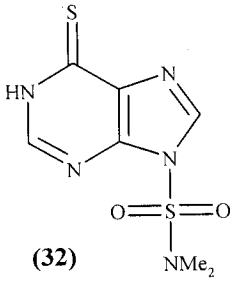
(29)



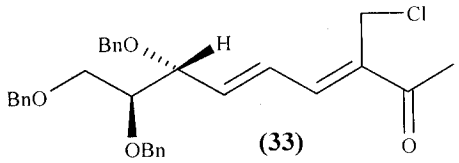
(30)



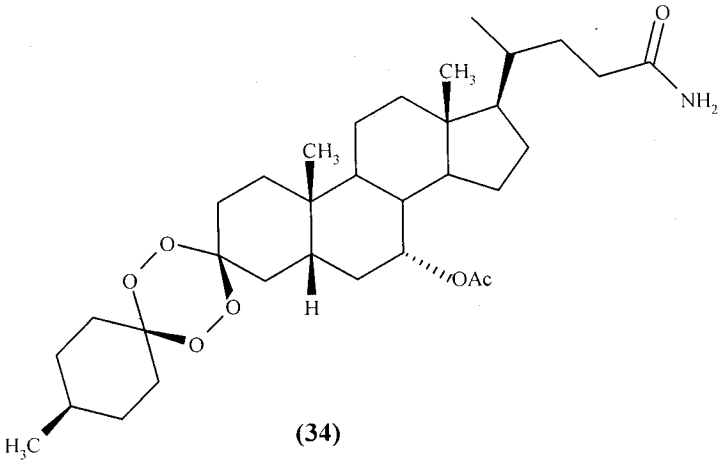
(31)



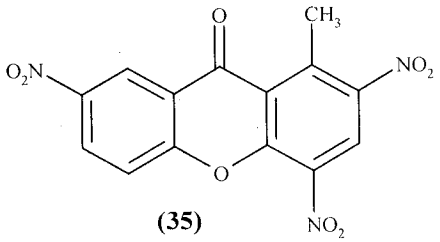
(32)



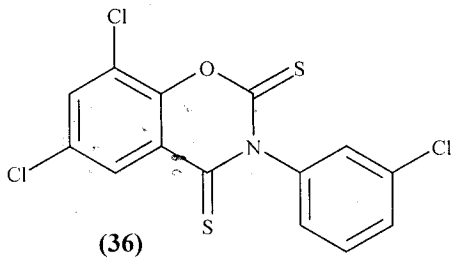
(33)



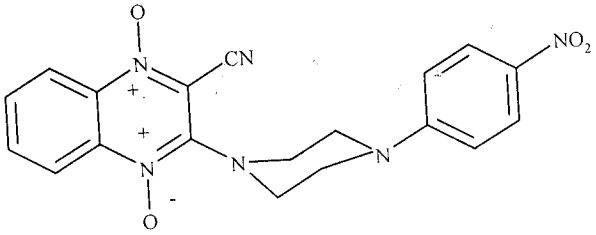
(34)



(35)



(36)



(37)

attached to a benzimidazole ring, a heterocycle that, along with structurally related benzothiazole, is often found in molecules with antimycobacterial activity[71,72]. Structural hybrids of isoniazid and quinolones, 4-quinolylhydrazones, show marked antitubercular activity[73]. Of this series, compound **(39)** at 0.78 µg/ml shows poor selectivity for mycobacteria (SI = 6.67). Compound **(40)**, among 1,3-thiazine derivatives display good antimycobacterial activity[74,75]. A derivative of 3-[4'-Y-(1,1'-biphenyl)-4-yl]-*N*,*N*-dimethyl-3-(4-X-phenyl)-2-propen-1-amine **(41)** represents new class of moderately potent antimycobacterial agents[76]. These molecules possess good antitubercular activity against $H_{37}R_a$ and $H_{37}R_v$ isolates with MIC as low as 1.6 µg/ml.

Compound **(42)** of fluorobenzyl derivatives represents successful use of an electron-withdrawing group attached to sulfur as a template for antimycobacterial activity[77]. Compound **(43)** represents the series having alkylmercapto group with chloropyrimidine derivatives[78]. Molecules, where heterocyclic ring is attached to a mercaptopropyl chain, exhibit good antimycobacterial activity at 0.78 µg/ml. Compound **(44)** of toluidine derivatives at 4 µg/ml show moderate to good antimycobacterial activity[79,80] and is also moderately selective versus mammalian Vero cells (SI = 16).

Deazapteridine derivatives, compound **(45)**[55], exhibit moderate activity against *M. tuberculosis*[81]. Even though cytotoxic properties of the compounds have previously been correlated with their tubulin binding capacity, active antitubercular molecules do not inhibit polymerization of mycobacterial FtsZ. Naturally occurring antibiotic pyrrolnitrin, a topical antifungal agent, has moderate antimycobacterial activity at 8 µg/ml. Compound **(46)** represents the given series. Antifungal activity is due to inhibition of protein kinase III, which is involved in osmosensing signal transduction pathway. Best MIC observed for the series is 1 µg/ml, but this is accompanied across all derivatives by pronounced levels of cytotoxicity (SI < 1)[55]. Oxazolidinone derivaties, a new class of synthetic antimicrobial agents, possess a unique mechanism of action in inhibiting protein synthesis[82]. Linezolid **(47)** inhibits multi-drug resistant isolates *in vitro* at 2 µg/ml[83]. PNU 100480 **(48)** is potential antimycobacterial agent. Several analogues of this series possess activity (MICs <0.125 µg/ml), which is also supported by good antitubercular activity against multiple strains of *M. tuberculosis*[84].

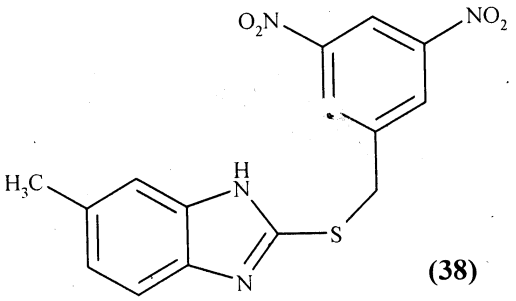
A class of putative monoxazolidinone analogues, compound **(49),** show notable activity against *M. tuberculosis* at 0.5 µg/ml and a moderate selectivity index[85] (SI=16). A series of galactopyranosyl amino alcohols, compound **(50)**, a dimeric hybrid of a galactofuranosyl ethambutol analogue, display potent *in vitro* activity at 1.56 µg/ml against *M. tuberculosis*[55]. Compound **(51)**, based on the thiazoline template[86], shows good MICs of 0.3 µg/ml with low toxicity. SAR study shows that replacing thiazoline with a thiazolidinone substantially reduced activity.

Novel Natural and Synthetic Antitubercular Agents

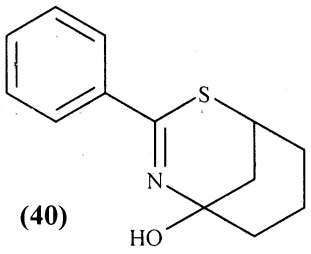
Evaluation of a series of 6-chloro-3-phenyl-4-thioxo-2H-1,3-benzoxazine-2(3H)-ones **(52)** and 6-chloro-3-phenyl-2H-1, 3-benzoxazine-2, 4(3H)-dithiones **(53)** revealed compounds having potent antimycobacterial activity against *M. tuberculosis*[87]. Pseudopteroxazole **(54)** and seco-pseudopteroxazole **(55)**, active diterpenoid alkaloids containing the uncommon benzoxazole moiety[88] isolated from *Pseudopterogorgia elisabethae*, possess anti tuberculosis activity.

Tryptanthrin **(56)** is a novel indoloquinazolinone alkaloid, which at 0.5-1 µg /ml showed potency against various strains of *M. tuberculosis*[89]. Intra and extra-cellular activities of phenazine derivatives are compared to clofazimine **(57)** and rifampicin against *M. tuberculosis* $H_{37}Rv$ (ATCC 27294)[90]. One of the phenazine derivatives, B4169 **(58)**, potently inhibits bacterium at 0.015 µg/ml, while corresponding value for clofazimine is 0.06 µg/ml. Several other derivatives of phenazines, like B4128 **(59)**, also show significant intracellular activity against *M. tuberculosis* infected monocyte-derived macrophages at 0.001 µg/ml and are superior to both clofazimine and rifampicin. The compounds are also more active than clofazimine against multi drug resistant strains.

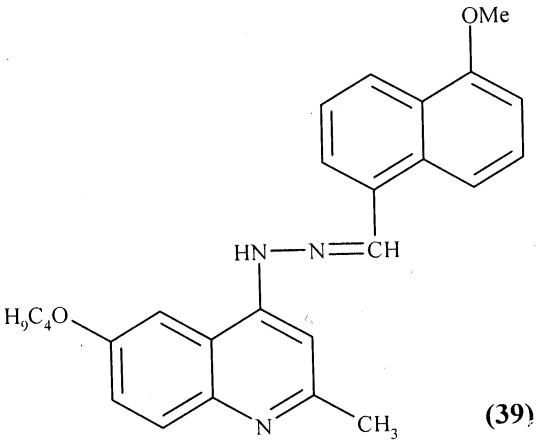
Calanolide A **(60)**, a naturally occurring pyranocoumarin[91] with dual activity against TB and HIV infections[55,92], is an inhibitor of HIV-1 reverse transcriptase[93] and also displays good *in vitro* activity towards *M. tuberculosis*. In a preliminary assessment of its activity, calanolide A is comparable with the positive control isoniazid and remains effective against rifampin and streptomycin resistant TB strains[55]. In addition, calanolide B **(61)** is readily available in substantial quantities from renewable



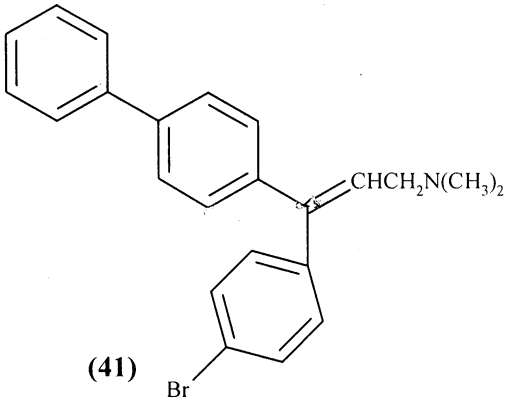
(38)



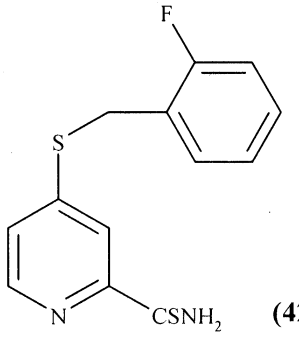
(40)



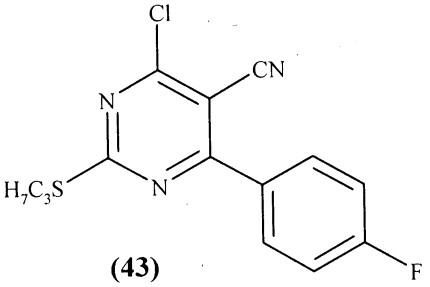
(39)



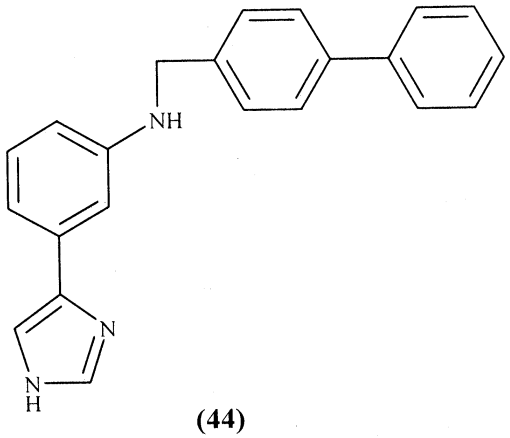
(41)



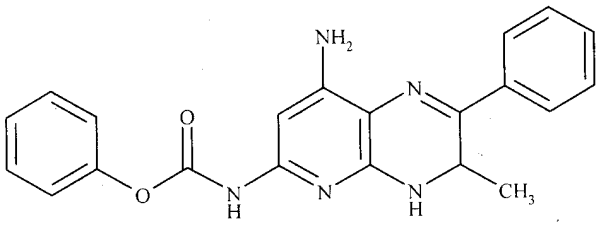
(42)



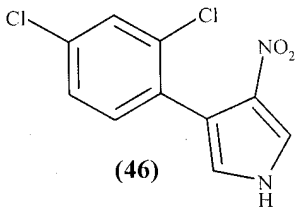
(43)



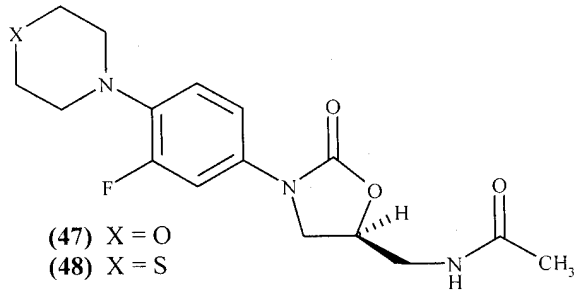
(44)



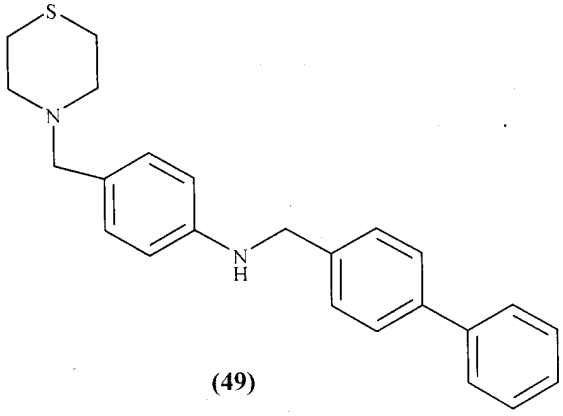
(45)



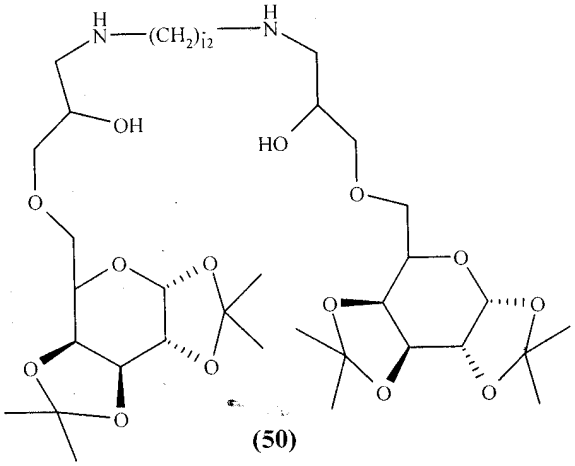
(46)



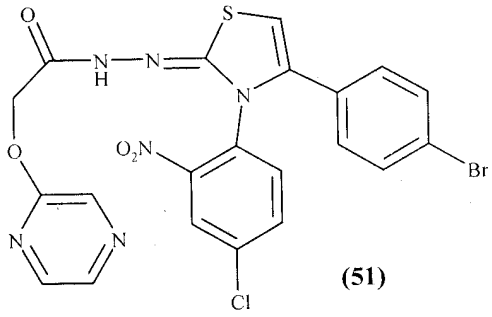
(47) X = O
(48) X = S



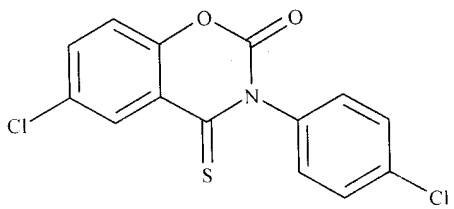
(49)



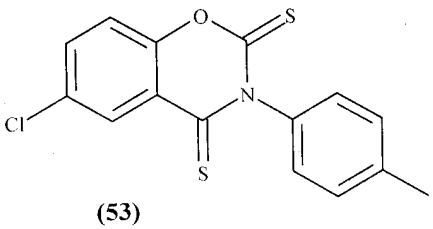
(50)



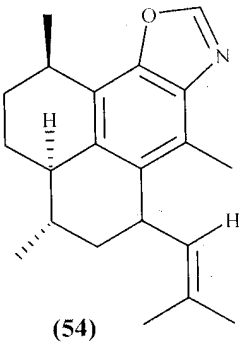
(51)



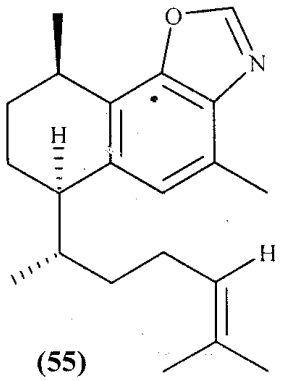
(52)



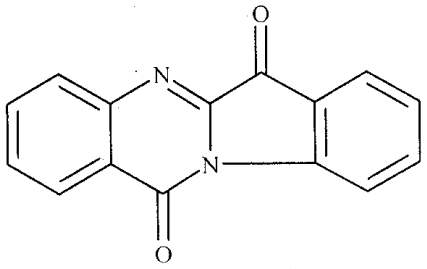
(53)



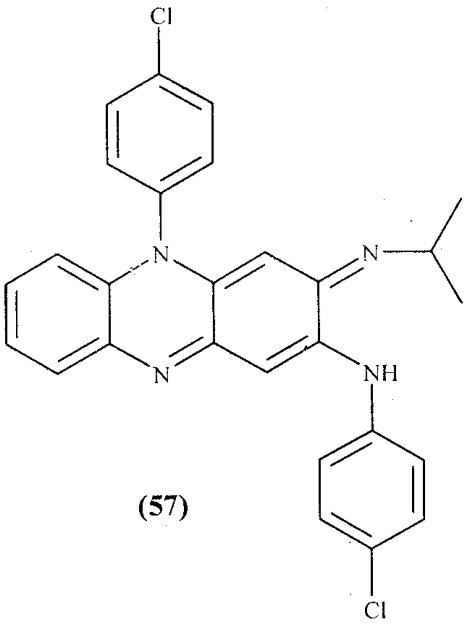
(54)



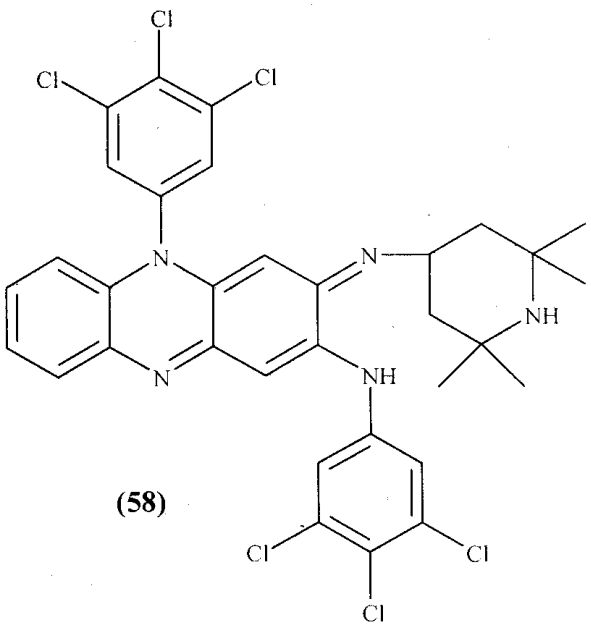
(55)



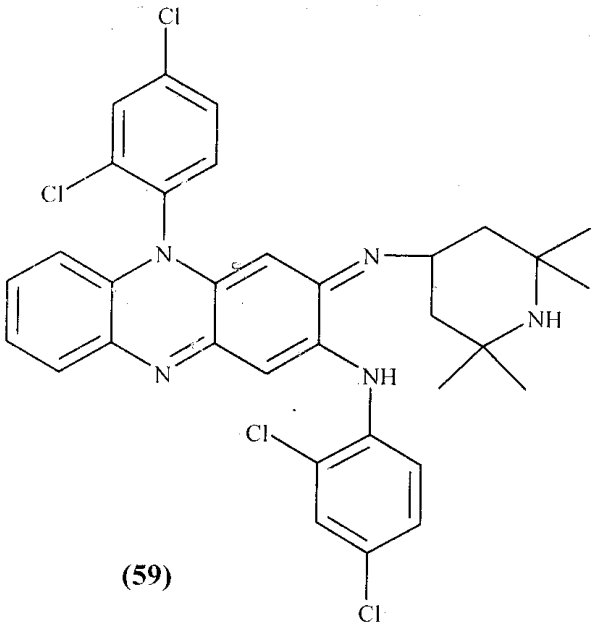
(56)



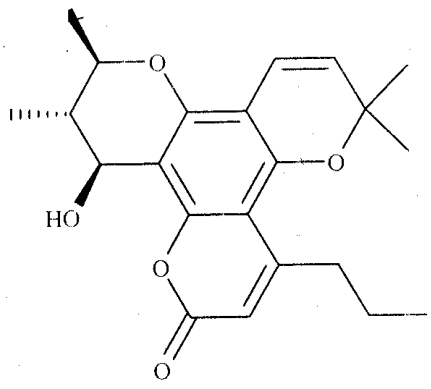
(57)



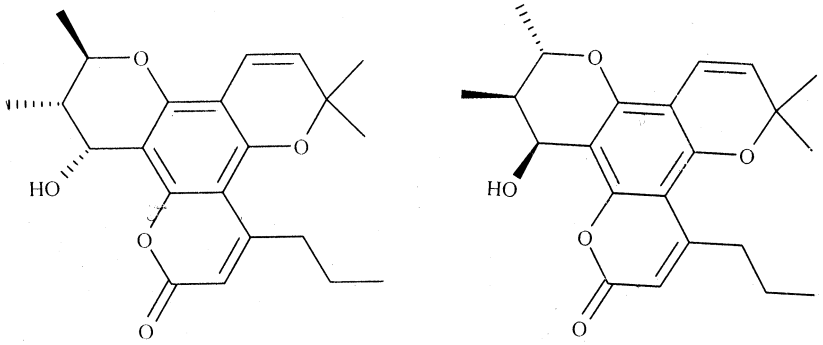
(58)



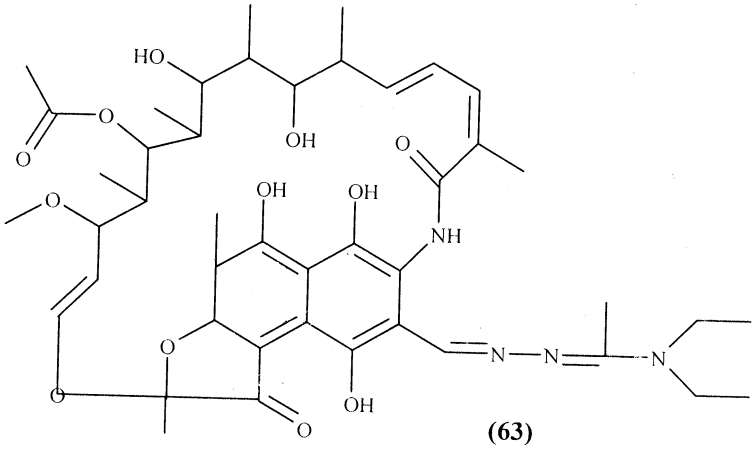
(59)



(60)



(61) (62)



(63)

natural sources like *Calophyllum* seed oil[94] having a similar spectrum of activity to calanolide A against *mycobacteria*. Other analogues are obtained either from plant extracts or by synthesis and compound **(62)** has been patented for its antimycobacterial properties[95]. Rifametane (SPA-S-565) **(63)**, a new semi-synthetic rifamycin, has a bactericidal spectrum and potency similar to that of rifampicin, but with better pharmacokinetic properties[96]. This is reflected in the fact that although the MIC90 values of the 2 compounds are the same against 20 strains of *M. tuberculosis*, in TB-infected mice, rifametane proves to be the more effective orally[97].

Newer Untouched Targets for Antitubercular Therapy

Immunomodulators

Immunomodulators might be useful in TB treatment. Gamma interferon has been administered by aerosol to MDR TB patients and shown to have some bacteriological effects[98]. Thalidomide, which block TNFα production, has beneficial effects on weight gain in both HIV positive and negative TB patients[99].

Isocitrate lyase Inhibitors

The persistence of *M. tuberculosis* in mice is facilitated by isocitrate lyase, an obligate enzyme for metabolism of fatty acids[100,101]. Biochemical studies suggest that in chronically infected lung tissues, fatty acids may be a major source of carbon and energy in *M. tuberculosis* metabolism. The data shows that whilst isocitrate lyase is important for survival of *M. tuberculosis* in the lung during the persistent phase of infection, it is not essential for growth in the acute stage[101]. Combination therapy of existing TB drugs with an isocitrate lyase inhibitor might be expected to expedite eradication of TB infections since conventional anti-microbials target cell-wall biosynthesis and chromosome replication.

Conclusions

One of the main lines of enquiry for recent research is the *M. tuberculosis* cell wall or envelope. From all the reported molecules, certain molecules exhibit potent activity against *M. tuberculosis*. Novel quinolones like CS-940 shows IC50 of 0.25-0.5 μg/ml and some thio analogue of purines exhibit low MIC. PA-824 and PA-1343 are the novel nitroimidazopyran

derivatives, the most advanced tubercular specific agents, at 0.015 - 0.25 µg/ml. Another significant development in the field of novel antimicrobial is oxazolidinones derivatives with unique mechanism of action of inhibiting protein synthesis. Some phenazine derivatives like B-4169 and B-4128 also show activity with MIC around 0.015 µg/ml. Immunomodulators and isocitrate lyase inhibitors can be further exploited to improve the treatment regimen. Mode of action of certain lead should be defined to greatly increase its chances for the development as a drug. SAR studies can be improved to assess the true potential of each series. Efforts must be made to identify compounds acting upon the key targets. Further, work should be carried out on reported molecules and the other emerging ones from the focused studies, which should lead to newer therapeutic agents in upcoming years.

References

1 Lemke T L, in *Foye's Principles of Medicinal Chemistry* by T L Lemke & D A Williams, **5th edn** (Lippincott Williams & Wilkins, USA) 2002, 904-906.

2 Panchagnula R, Agrawal S & Kaul C L, Fixed dose combinations in treatment of tuberculosis, *Indian J Pharm Sci,* **63** (2001) 1-9.

3 www.who.org

4 Petri W A, in *Goodman and Gilman's The Pharmacological Basis of Therapeutics* by J G Hardman & L E Limbird, **11th edn** (MaGraw Hill,) 2006, 1203-1223.

5 Kuo M R, Morbidoni H R, Alland D, Sneddon S F, Gourlie B B, Staveski M M, Leonard M, Gregory J S, Janjigian A D, Lee C, Musser J M, Kreiswirth B, Iwamoto H, Perozzo R, Jacobs W R, Sacchettini J C & Fidock D A, Targeting tuberculosis and malaria through inhibition of enoyl reductase, *J Biol Chem*, **278** (2003) 20851-20859.

6 Lee R, Monsey D, Weston A, Duncan K, Rithner C & McNeil M, Enzymatic synthesis of UDP-galactofuranose and an assay for UDP-galactopyranose mutase based on high-performance liquid chromatography, *Anal Biochem*, **242** (1996) 1-7.

7 Lee R E, Brennan P J & Besra G S, *Mycobacterial arabinan* biosynthesis: The use of synthetic arabinoside acceptors in the development of an arabinosyl transfer assay, *Glycobiology*, **7** (1997) 1121-1128.

8 Ma Y, Mills J A, Belisle J T, Vissa V, Howell M, Bowlin K, Scherman M S & McNeil M, Determination of the pathway for rhamnose biosynthesis in mycobacteria: Cloning, sequencing and expression of the *Mycobacterium tuberculosis* gene encoding alpha-D-glucose-1-phosphate thymidylyltransferase, *Microbiology*, **143** (1997) 937-945.

9 Nassau P M, Martin S L, Brown R E, Weston A, Honsey D, McNeil M R & Duncan K, Galactofuranose biosynthesis in *Escherichia coli* K-12: Identification and cloning of UDP-galactopyranose mutase, *J Bacteriol*, **178** (1996) 1047-1052.

10 Takayama K, Wang C & Besra G S, Pathway to synthesis and processing of mycolic acids in *Mycobacterium tuberculosis*, *Clin Microbiol Rev*, **18** (2005) 81-101.

11 http://www.chemsoc.org/chembytes/ezine/1998/evans.htm

12 Sensi P & Grassi C G, in *Berger's Medicinal Chemistry and Drug Discovery* by M E Wolff, **5th edn** (John-Wiley and sons Inc., USA) 1996, 576-635.

13 Beale J M, in *Wilson and Giswold's Textbook of Organic Medicinal Chemistry* by J M Beale & J H Block, **9th edn** (Lippincott Williams & Wilkins, USA) 2004, 535-580.

14 Zeind C S, Gourley G K & Chandler-Toufieli D M, in *Textbook of Therapeutics, Drug and Disease Management*, by E T Harfindal & D R Gourley, **7th edn** (Lippincott Williams & Wilkins, USA) 2004, 1427-1450.

15 Vohra R, Gupta M, Chaturvedi R & Singh Y, Attack on the scourge of tuberculosis: patented drug targets, *Recent Patents on Anti-Infective Drug Discovery*, **1** (2006) 95-106.

16 Ji B, Lounis N, Maslo C, Truffot-Pernot C, Bonnafous P & Grosset J, *In vitro* and *in vivo* activities of moxifloxacin and clinafloxacin against *Mycobacterium tuberculosis*, *Antimicrob Ag Chemother*, **42** (1998) 2066-2069.

17 Miyazaki E, Miyazaki M, Chen J M, Chaisson R E & Bishai W R, Moxifloxacin (BAY12-8039), a new 8-methoxyquinolone is active in a mouse model of tuberculosis, *Antimicrob Ag Chemother*, **43** (1999) 85-89.

18 Sullivan J T, Woodruff M, Lettieri J, Agarwal V, Krol G J, Leese P T, Watson S & Heller A H, Pharmacokinetics of a once-daily oral dose of moxifloxacin (Bay 12-8039), a new enantiomerically pure 8-methoxy quinolone, *Antimicrob Ag Chemother*, **43** (1999) 2793-2797.

19 Biedenbach D J, Sutton L D & Jones R N, Antimicrobial activity of CS-940, a new trifluorinated quinolone, *Antimicrob Ag Chemother*, **39** (1995) 2325-2330.

20 Zhao B Y, Pine R, Domagala J & Drlica K, Fluoroquinolone action against clinical isolates of *Mycobacterium tuberculosis*: Effects of a C-8 methoxyl Group on survival in liquid media and in human macrophages, *Antimicrob Ag Chemother*, **43** (1999) 661-666.

21 Onodera Y, UchidaY, Tanaka M & Sato K, Dual inhibitory activity of sitafloxacin(DU-6859a) against DNA gyrase and topoisomerase IV of *Streptococcus pneumoniae*, *J Antimicrob Chemother*, **44** (1999) 533-536.

22 Roy B N, Karnik M A & Sankaran R, Recent advances in research on antituberculars, *J Ind Chem Soc*, **79** (2002) 320-335.

23 Sheu J Y, Chen Y L, Tzeng C C, Hsu S L, Fang K C & Wang T C, Synthesis, and antimycobacterial and cytotoxic evaluation of certain fluoroquinolone derivatives, *Helv Chem Acta*(Eng), **86** (2003) 2481-2489; *Chem Abstr*, **140** (2004) 16634.

24 Kunin C M & Ellis W Y, Antimicrobial activities of mefloquine and a series of related compounds, *Antimicrob Ag Chemother,* **44** (2000) 848-852.

25 Inderlied C B, Bermudez L, Ellis W Y, Aralar P A, Kolonoski P & Petrofsky M, *Poster 999, presented at the 40th Interscience Conf on Antimicrobial Agents and Chemotherapy*, Toronto, Ontario, Canada, 17 - 20 September, 2000,. Derwent World Drug Alert abstract WD-2000-013271.

26 Chambhare R V, Bobade A S & Khasde B G, Synthesis of novel 3-N-[(substitutedaryl/heterotryl)methylene]-imino-2-methyl-5-thienyl-thieno[2,3-d]pyrimidine-4-(3H)-ones as

possible antimicrobial agents, *Ind J Het Chem*, **12** (2002) 67-68.

27 Sriram D, Yogeeswari P, Pandeya S N & Ananthan S, Synthesis and antituberculous activity of n-mannich bases of 3-[4-(4-chlorophenyl)-6-(4-methylphenyl)pyrimidin-2-yl]iminoisatin derivatives, *Scientia Pharmaceutica (Eng)*, **70** (2002) 39-48; *Chem Abstr*, **138** (2003) 89754.

28 Vanheusden V, Munier-Lehmann H, Busson R, Froeyen M, Dugue L, Heyerick A, Keukeleire D D, Pochet S, Busson R, Herdiwijn P & Calenbergh S V, 3'-C-Branched-chain-substituted nucleosides and nucleotides as potent inhibitors of *Mycobacterium tuberculosis* thymidine monophosphate kinase, *J Med Chem*, **46** (2003) 3811-3821.

29 Pathak A K, Pathak V, Seitz L E, Reynolds R C & Suling W J, Anti-mycobacterial agents: 1-Thio analogues of purine, *J Med Chem*, **47** (2004) 273-276.

30 Baker W R, Mitscher L A, Arain M A, Shaver R & Stover C K, Tuberculosis: A search for novel therapy starting with natural products, *Annu Rep Med Chem*, **31** (1996) 161-170.

31 Baker W R, Shaopei C & Keeler E L, *US Pat* 6, 087, 358 (2000); *Chem Abstrt*, **133** (2000) 89525.

32 Stover C K, Warrener P, VanDevanter D R, Sherman D R, Arain T M, Langhorne M H, Anderson S W, Towell J A, Yuan Y, McMurray D N, Kreiswirth B N, Barry C E & Baker W R, A small-molecule nitroimidazopyran drug candidate for the treatment of tuberculosis, *Nature*, **405** (2000) 962-966.

33 Ashtekar D R, Rabi C P, Nagrajan K, Vishvanathan N, Bhatt A D & Rittel W, *In vitro* and *in vivo* activities of the nitroimidazole CGI 17341 against *Mycobacterium tuberculosis*, *Antimicrob Ag Chemother*, **37** (1993) 183-186.

34 Ma Y, Stern R J, Scherman M S, Vissa V D, Yan W, Jones V C, Zhang F, Franzblau S G, Lewis W H & McNeil M R, Drug targeting *Mycobacterium tuberculosis* cell wall synthesis: genetics of dTDP-rhamnose synthetic enzymes and development of a microtiter plate-based screen for inhibitors of conversion of dTDP-glucose to dTDP-rhamnose, *Antimicrob Ag Chemother*, **5** (2001) 1407-1416.

35 Andres C J, Bronson J J, D'Andrea S V, Deshpande M S, Falk P J, Grant-Young K A, Harte W E, Ho H T, Misco P F, Robertson J G, Stock D, Sun Y & Walsh A W, 4-Thiazolidinones: Novel inhibitors of the bacterial enzyme MurB, *Bioorg Med Chem Lett*, **10** (2000) 715-717.

36 Jones P B, Parrish N M, Houston T A, Stapon A, Bansal N P, Dick J D & Townsend C A, A new class of anti-tuberculosis agents, *J Med Chem*, **43** (2000) 3304-3314.

37 Kremer L, Maughan W N, Wilson R A, Dover L G & Besra G S, The *M. tuberculosis* antigen 85 complex and mycolyltransferase activity, *Lett Appl Microbiol*, **34** (2002) 233-237.

38 Rose J D, Maddry J A, Comber R N, Suling W J, Wilson L N & Reynolds R C, Synthesis and biological evaluation of trehalose analogs as potential inhibitors of mycobacterial cell wall biosynthesis, *Carbohydr Res*, **337** (2002) 105-120.

39 Wolucka B A, McNeil M R, deHoffman E, Chojnaki T & Brennan P J, Recognition of the lipid intermediate for arabinogalactan/arabinomannan biosynthesis and its relation to the mode of action of ethambutol on mycobacteria, *J Biol Chem*, **269** (1994) 23328-23335.

40 Scherman M S, Kalbe-Bournonville L, Bush D, Xin Y, Deng L & McNeil M, Polyprenylphosphate-pentoses in mycobacteria are synthesized from 5-phosphoribose pyrophosphate, *J Biol Chem*, **271** (1996) 29652-29658.

41 Centrone C A & Lowary T L, Synthesis and antituberculosis activity of C-phosphonate analogues of decaprenolphosphoarabinose, a key intermediate in the biosynthesis of mycobacterial arabinogalactan and lipoarabinomannan. *J Org Chem*, **67** (2002) 8862-8870.

42 Banerjee A, Dubnau E, Quemard A, Balasubramanian V, Um K S, Wilson T, Collins D, de Lisle G & Jacobs W R Jr, InhA, a gene encoding a target for isoniazid and ethionamide in *M. tuberculosis*., *Science*, **263** (1994) 227-230.

43 Kuo M R, Morbidoni H R, Alland D, Sneddon S F, Gourlie B B, Staveski M M, Leonard M, Gregory J S, Janjigian A D, Lee C, Musser J M, Kreiswirth B, Iwamoto H, Perozzo R, Jacobs W R, Sacchettini J C & Fidock D A., Targeting tuberculosis and malaria through inhibition of enoyl reductase, *J Biol Chem*, **278** (2003) 20851-20859.

44 Temple C G, in *Chemistry and Biology of Pteridines*, by C Curius, S Ghisla & N Blau (Walter de Gruyter, Berlin, Germany) 1990, 1009-1014.

45 Temple C G, Wheeler G P, Elliott R D, Rose J D & Comber R N, Montgomery J A, 1,2-dihydropyrido[3,4-b]pyrazines: Structure-activity relationships, *J Med Chem*, **26** (1983) 91-95.

46 White E L, Suling W J, Ross L J, Seitz L E & Reynolds R C, 2-Alkoxycarbonylaminopyridines: Inhibitors of *Mycobacterium tuberculosis* FtsZ., *J Antimicrob Chemother*, **50** (2002) 111-114.

47 Suling W J, Seitz L E, Pathak V, Westbrook L, Barrow E W, Zywno-Van-Ginkel S, Reynolds R C, Piper J R & Barrow W W, Antimycobacterial activities of 2,4-diamino-5-deazapteridine derivatives and effects on mycobacterial dihydrofolate reductase, *Antimicrob Ag Chemother*, **44** (2000) 2784-2793.

48 Tripathi R. P, Tripathi R, Tiwari V K, Bala L, Sinha S, Srivastava A, Srivastava R & Srivastava B S, Synthesis of glycosylated beta-amino acids as new class of antitubercular agents, *Eur J Med Chem*, **37** (2002) 773-781.

49 Yadav N, McDevitt R E, Benard S & Falco S C, Single amino acid substitutions in the enzyme acetolactate synthase confer resistance to the herbicide sulfometuron methyl, *Proc Natl Acad Sci*, **83** (1986) 4418-4422.

50 Andres C J, Bronson J J, D'Andrea S V, Deshpande M S, Falk P J, Grant-Young K A, Harte W E, Ho H T, Misco P F, Robertson J G, Stock D, Sun Y & Walsh A W, 4-Thiazolidinones: novel inhibitors of the bacterial enzyme MurB, *Bioorg Med Chem Lett*, **10** (2000) 715-717.

51 Velezheva V S, Brennan P J, Marshakov V Y, Gusev D V, Lisichkina I N, Pergudov A S, Tchernousova L N, Smirnova T G, Andreevskaya S N & Medvedev A E, Novel pyridazino[4,3-*b*]indoles with dual inhibitory activity against *M. tuberculosis* and monoamine oxidase, *J Med Chem*, **47** (2004) 3455-3461.

52 Andries K, A diarylquinoline drug active on the ATP synthase of *Mycobacterium tuberculosis*, *Science*, **307** (2005) 223-227.

53 Gundersen L, Nissen-Meyer J & Spilsberg B, Synthesis and antimycobacterial activity of 6-arylpurines: The requirements for the N-9 substituent in active anti-mycobacterial purines, *J Med Chem*, **45** (2002) 1383-1386.

54 Pathak A K, Pathak V, Seitz L E, Suling W J & Reynolds R C, Anti-mycobacterial agents: 1-Thio analogues of purine, *J Med Chem*, **47** (2004) 273-276.

55 Ballell L, Field R A, Duncan K & Young R J, New small-molecule synthetic antimycobacterials, *Antimicrob Ag Chemother*, **49** (2005) 2153-2163.

56 Deidda D, Lampis G, Fioravanti R, Biava M, portea G, Zanetti S & Pompei R, Bactericidal activities of the pyrrole derivative BM212 against multidrug-resistant and intramacrophagic *Mycobacterium tuberculosis* strains, *Antimicrob Ag Chemother*, **42** (1998) 3035-3037.

57 Biava M, BM 212 and its derivatives as a new class of anti-mycobacterial active agents, *Curr Med Chem*, **9** (1995) 1859-1869.

58 Scozzafava A, Mastrolenzo A & Supuran C T, Anti-mycobacterial activity of 9-sulfonylated/sulfenylated-6-mercaptopurine derivatives, *Bioorg Med Chem Lett*, **11** (2001) 1675-1678.

59 Pathak R, Shaw A K, Bhaduri A P, Chandrasekhar K V G, Sinha S, Srivastava A, Srivastava K K, Chaturvedi V, Srivastava R, Srivastava B S & Arora S, Higher acyclic nitrogen containing deoxy sugar derivatives: a new lead in the generation of antimycobacterial chemotherapeutics. *Bioorg Med Chem*, **10** (2002) 1695-1702.

60 Pathak R, Pant C S, Shaw A K, Bhaduri A P, Gaikwad A N, Sinha S, Srivastava A, Srivastava K K, Chaturvedi V, Srivastava R & Srivastava B S, Baylis-Hillman reaction: Convenient ascending syntheses and biological evaluation of acyclic deoxy monosaccharides as potential anti-mycobacterial agents, *Bioorg Med Chem*, **10** (2002) 3187-3196.

61 Dong Y, Matile H, Chollet J, Kaminsky R, Wood K & Vennerstrom J L, Synthesis and antimalarial activity of 11 dispiro-1,2,4,5-tetraoxane analogues of WR 148999. 7,8,15,16-tetraoxadispiro[5.2.5.2]hexadecanes substituted at the 1 and 10 positions with unsaturated and polar functional groups, *J Med Chem*, **42** (1999) 1477-1480.

62 Ghosal S, Biswas K & Chauduri R K, Chemical constituents of Gentianaceae. XXIV. Anti-*Mycobacterium tuberculosis* activity of naturally occurring xanthones and synthetic analogs, *J Pharm Sci*, **67** (1978) 721-722.

63 Savini L, Chiasserini L, Gaeta A & Pellerano C, Synthesis and anti-tubercular evaluation of 4-quinolylhydrazones, *Bioorg Med Chem*, **10** (2002) 2193-2198.

64 Waisser K, Gregor J, Dostál H, Kubicová L, Klimešová V & Kaustová J, Influence of the replacement of the oxo function with the thioxo group on the antimycobacterial activity of 3-aryl-6,8-dichloro-2H-1,3-benzoxazine-2,4(3H)-diones and 3-arylquinazoline-2,4(1H,3H)-diones, *Farmaco*, **56** (2001) 803-807.

65 Waisser K, Bures O, Holy P, Kunes J, Oswald R, Jiraskova L, Pour M, Klimesova V, Palat K, Kaustova J, Danse H M & Mollmann U, Antimycobacterial 3-aryl-2H-1,3-benzoxazine-2,4(3H)-diones, *Pharmazie*, **58** (2003) 83-94.

66 Ganley B, Chowdhury G, Bhansali J, Daniels J S & Gates K S, Redox-activated, hypoxia-selective DNA cleavage by quinoxaline 1,4-di-N-oxide, *Bioorg Med Chem*, **9** (2001) 2395-2401.

67 Carta A, Loriga M, Paglietti G, Mattana A, Fiori P L, Mollicotti P, Sechi L & Zannetti S, Synthesis, anti-mycobacterial, anti-trichomonas and anti-candida *in vitro* activities of 2-substituted-6,7-difluoro-3-methylquinoxaline 1,4-dioxides, *Eur J Med Chem*, **39** (2004) 195-203.

68 Ortega M A, Montoya M E, Jaso A, Zarranz B, Tirapu I, Aldana I & Monge A, Anti-mycobacterial activity of new quinoxaline-2-carbonitrile and quinoxaline-2-carbonitrile 1,4-di-N-oxide derivatives, *Pharmazie*, **56** (2001) 205-207.

69 Zarranz B, Jaso A, Aldana I & Monge A, Synthesis and antimycobacterial activity of new quinoxaline-2-carboxamide 1,4-di-N-oxide derivatives, *Bioorg Med Chem*, **11** (2003) 2149-2156.

70 Waisser K, Klimesova V & Odlerová Z, Design of compounds active against *Mycobacterium tuberculosis, Folia Pharm Univ Carol*, **18** (1995) 31-37.

71 Gasparova R, Lacova M, El-Shaaer H M & Odlerová Z, Synthesis and antimycobacterial activity of some new 3-heterocyclic substituted chromones, *Farmaco*, **52** (1997) 251-253.

72 Vicini P, Geronikaki A, Incerti M, Busonera B, Poni G, Cabras C A & La Colla P, Synthesis and biological evaluation of benzo[d]isothiazole, benzothiazole and thiazole Schiff bases, *Bioorg Med Chem*, **11** (2003) 4785-4789.

73 Savini L, Chiasserini L, Gaeta A & Pellerano C, Synthesis and anti-tubercular evaluation of 4-quinolylhydrazones, *Bioorg Med Chem*, **10** (2002) 2193-2198.

74 Koketsu M, Tanaka K, Takenaka Y, Kwong C D & Ishihara H, Synthesis of 1,3-thiazine derivatives and their evaluation as potential antimycobacterial agents, *Eur J Pharm Sci*, **15** (2002) 307-310.

75 Koketsu M, Takenaka Y, Hiramatsu S & Yshihara H, Facile preparation of 1,3-selenazine derivatives by reaction of primary selenoamides with α,ß-unsaturated aldehydes of in the presence of BF_3:Et_2O, *Heterocycles*, **55** (2001) 1181-1188.

76 De Souza A O, Santos R R Jr, Ferreira-Julio J F, Rodríguez J A, Melo P S, Haun M, Sato D N & Durán N, Synthesis, antimycobacterial activities and cytotoxicity on V79 of 3-[4'-Y-(1,1'-biphenyl)-4-yl]-N,N-dimethyl-3-(4-X-phenyl)-2-propen-1-amine derivatives, *Eur J Med Chem*, **36** (2001) 843-850.

77 Klimesova V, Palat K, Waisser K & Klimes J, Combination of molecular modeling and quantitative structure-activity relationship analysis in the study of antimycobacterial activity of pyridine derivatives, *Int J Pharm*, **207** (2000) 1-6.

78 Agarwal N, Srivastava P, Raghuwanshi S K, Upadhyay D N, Sinha S, Shukla P K & Ram V J, Chloropyrimidines as a new class of antimicrobial agents, *Bioorg Med Chem*, **10** (2002) 869-874.

79 Biava M, Fioravanti R, Porretta G C, Sleiter G, Ettorre A, Deidda D, Lampis G & Pompei R, New toluidine derivatives with antimycobacterial and antifungal activities, *Med Chem Res*, (1997) 228-250.

80 Biava M, Fioravanti R, Porretta G C, Sleiter G, Ettorre A, Deidda D, Lampis G & Pompei R, Toluidine derivatives with antimycobacterial and antifungal activities, *Med Chem Res*, (1998) 523 -533.

81 Suling W J & Maddry J A, Antimycobacterial activity of 1-deaza-7,8-dihydropteridine derivatives against *Mycobacterium tuberculosis* and *Mycobacterium avium* complex *in vitro, J Antimicrob Chemother*, **47** (2001) 451-454.

82 Diekema D J & Jones R N, Oxazolidinones, A review, *Drugs*, **59** (2000) 7-16.

83 Zurenko G E, Yagi B H, Schaadt R D, Allison J W, Kilburn J O, Glickman S E, Hutchinson D K, Barbachyn M R & Brickner S J, *In vitro* activities of U-100592 and U-100766, novel oxazolidinone antibacterial agents, *Antimicrob Ag Chemother*, **40** (1996) 839-845.

84 Barbachyn M R, Hutchinson, Brickner S J, Cynamon M H, Kilburn J O, Klemens S P, Glickman S E, Grega K C, Hendges S K, Toops D S, Ford C W & Zurenko G E, Identification of a novel oxazolidinone (U-100480) with potent antimycobacterial activity, *J Med Chem*, **39** (1996) 680-685.

85 Arima K, Imanaka H, Kousaka M, Fukuda A & Tamura G, Studies on pyrrolnitrin, a new antibiotic. I. Isolation and properties of pyrrolnitri, *J Antibiot Ser A*, **18** (1968) 201-204.

86 Bonde C G & Gaikwad N J, Synthesis and preliminary evaluation of some pyrazine containing thiazolines and thiazolidinones as antimicrobial agents, *Bioorg Med Chem*, **12** (2004) 2151-2161.

87 Waisser K, Gregor J, Kubicova L, Klimesova V, Kunes J, Machacek M & Kaustova J, New groups of antimycobacterial agents: 6-chloro-3-phenyl-4-thioxo-2H-1,3-benzoxazine-2(3H)-ones and 6-chloro-3-phenyl-2H-1,3-benzoxazine-2,4(3H)-dithiones, *Eur J Med Chem*, **35** (2000) 733-741.

88 Rodriguez A D, Ramirez C, Rodriguez II & Gonzalez E, Novel antimycobacterial benzoxazole alkaloids from the West Indian Sea whip *Pseudopterogorgia elisabethae, Org Letts*, **1** (1999) 527-530.

89 Mitscher L A & Baker W, Tuberculosis: A search for novel therapy starting with natural products, *Med Res Rev*, **18** (1997) 363-374.

90 Van Rensburg C E J, Joone G K, Sirgel F A, Matlola N M & O'Sullivan J F, *In vitro* Investigation of the Antimicrobial Activities of Novel Tetramethylpiperidine-Substituted Phenazines against *Mycobacterium tuberculosis, Chemother*, **46** (2000) 43-48.

91 McKee T C, Covington C D, Fuller R W, Bokesch H R, Young S, Cardellina II J H, Kadushin M R, Soejarto D D, Stevens P F, Cragg G M & Boyd M R, Pyranocoumarins from tropical species of the genus *Calophyllum*: A chemotaxonomic study of extracts in the National Cancer Institute collection, *J Nat Prod*, **61** (1998) 1252-1256.

92 Currens M J, Gulakowski R J, Mariner J M, Moran R A, Buckheit R W Jr., Gustafson K R, McMahon J B & Boyd M R, Antiviral activity and mechanism of action of calanolide A against the human immunodeficiency virus type-1, *J Pharmacol Exp Therap*, **279** (1996) 645-651.

93 Dharmaratne H R, Wanigasekera W M, Mata-Greenwood E & Pezzuto J M, Inhibition of human immunodeficiency virus type 1reverse transcriptase activity by cordatolides isolated from *Calophyllum cordato-oblongum*, *Planta Medica*, **64** (1998) 460-461.

94 Spino C, Dodier M & Sotheeswaran S, Anti-HIV coumarins from *Calophyllum* seed oil, *Biorg Med Chem Lett*, **8** (1998) 3475-3478.

95 Xu Z Q, Lin Y M & Flavin M, *Pat* WO 200021514-A2.

96 Bruzzese T, Rimaroli C, Bonabello A, Mozzi G, Ajay S & Cooverj N D, Pharmacokinetics and tissue distribution of rifametane, a new 3-azinomethyl-rifamycin derivative, in several animal species, *Arzneimittel-Forschung*, **50** (2000) 60-71.

97 Strippoli V, Bruzzese T, Galli R & Siminetti N, The antibacterial activity of a new 3-azinomethylrifamycin. *Il Farmaco Ed Sci*, **43** (1988) 619-625.

98 Condos R, Rom W M & Schluger N W, Treatment of multidrug-resistant pulmonary tuberculosis with interferon-gamma via aerosol, *Lancet*, **349** (1997) 1513-1515.

99 Tramontana J M, Utaipat T, Molloy A, Akarasewi P, Burroughs M, Makonkawkeyoon S, Johnson B, Klausner J D, Rom W & Kaplan G, Thalidomide treatment reduces tumour necrosis factor production and enhances weight gain in patients with pulmonary tuberculosis, *Molec Med*, **1** (1995) 384-387.

100 Bishai W, Lipid lunch for persistent pathogen, *Nature*, **406** (2000) 683-685.

101 McKinney J D, Honer zu Bentrup K, Munoz Elias E J, Miczak A, Chen B, Chan W T, Swenson D, Sacchettini J C, Jacobs W R Jr & Russell D G, Persistence of *Mycobacterium tuberculosis* in macrophages and mice requires the glyoxylate shunt enzyme isocitrate lyase, *Nature*, **406** (2000) 735-738.