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Commentary

New antimicrobial agents on the horizon

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ABSTRACT

Antibiotic resistance issues necessitate the continued discovery and development of new antibacterial agents. Efforts are ongoing in two approaches to find new compounds that are effective against antibiotic-resistant pathogens. These efforts involve modification of existing classes including fluoroquinolones, tetracyclines, aminoglycosides, and β-lactams and identification of inhibitors against previously unexploited antibacterial targets. Examples of both approaches are described here with emphasis on compounds in late pre-clinical or clinical stages of development.

1. Introduction

As a result of decreasing attention to antimicrobial research within the pharmaceutical industry, we are approaching a critical need for new agents to treat increasingly resistant bacterial infections [1]. Organisms such as MRSA (methicillin-resistant *Staphylococcus aureus*) and methicillin-resistant coagulase negative staphylococci are still among the major pathogens of interest in both community and hospital-acquired infections; however, of particular concern are multidrug-resistant Gram-negative bacteria causing severe nosocomial infections that are not sensitive to most currently approved antibiotics. Although new antimicrobial agents are not being developed as rapidly as in previous decades, new approaches to the treatment of infectious diseases are still emerging. This commentary describes some of the newer antibacterial agents that have been described at the most recent international infectious disease meetings, with a focus on those investigational compounds that are in clinical trials. There appears to be a surprising number of potentially useful agents in development, providing some hope for the future.

One approach to new agents has been to continue to modify existing classes of previously successful antibiotics. New fluoroquinolones, aminoglycosides, tetracyclines and β-lactams are currently under development to treat multidrug resistant pathogens. In addition, combinations of β-lactamase inhibitors with new, and old, β-lactams are being developed to counteract the hydrolytic activity of newer β-lactamases. Analogs of these familiar drug classes will be discussed in this commentary with respect to their inhibitory properties and their development status.

Newer approaches to antimicrobial therapy include identification of inhibitors that are operative against previously unexploited essential bacterial targets such as peptide deformylase or metabolic pathways such as fatty acid biosynthesis. Although compounds in these areas have been explored in the past, progress of some of the newer inhibitors into clinical trials has been meeting with increased success. In addition, researchers are using previously validated antibacterial targets to discover new compounds that are chemically distinct and that interact with a bacterial target in different ways than existing compounds. Fig. 1 summarizes the cellular sites of action for both current antibacterial drugs and targets for agents under development. Perspectives on the feasibility of investigational drugs in all these categories will be provided in this commentary, with perhaps a more optimistic outlook than predicted for the past five years.

2. New antibacterial agents in development

2.1. DNA replication interactions

Inhibitors of bacterial DNA replication target proteins have provided valuable antibiotics over the years. There appear to be enough differences between the bacterial and human DNA replication processes to allow for the discovery and development of selective agents. In general, these compounds have proven to be bactericidal and effective against a number of important human pathogens. The associated targets continue to be exploited in the search for new antibacterial agents.

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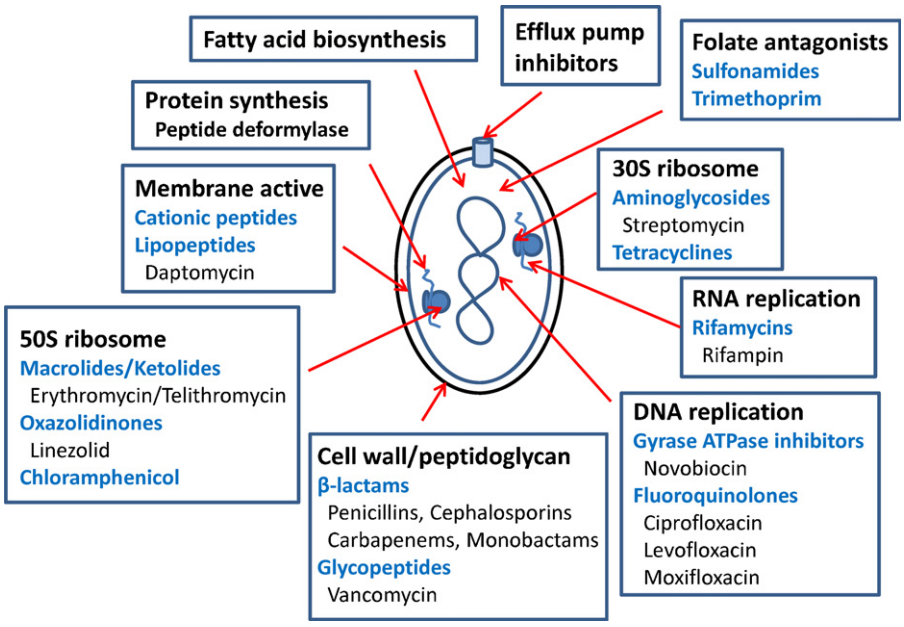


Fig. 1. Diagram of a bacterial cell showing the targets for existing drug classes with examples of approved drugs and targets for agents currently in development.

2.1.1. Fluoroquinolones

Inhibitors of bacterial topoisomerases, exemplified by several marketed fluoroquinolones, have been important and widely used antibacterial agents for several decades [2–4]. One reason for the continued effectiveness of the fluoroquinolones is that they inhibit two essential bacterial enzymes: DNA gyrase (GyrA subunit) and topoisomerase IV (ParC or GrlA subunit), although to varying extents depending on the pathogen. Other topoisomerase inhibitors have been identified that bind to different locations on these targets or target the partner subunits of these enzyme heterodimers (GyrB and/or ParE or GrlB), often allowing for coverage of quinolone-resistant strains [2]. Research and development of new inhibitors of these two validated antibacterial targets shows no signs of ending soon [4]. We describe below several examples of bacterial topoisomerase inhibitors that have either entered human clinical trials or are in late stages of pre-clinical research.

Fluoroquinolones continue to be developed. JNJ-Q2 (Fig. 2, **1**), originally discovered at Johnson & Johnson, recently completed a Phase 2 clinical trial as a new agent for the treatment of skin infections, and will be further developed by Furiex Pharmaceuticals for possible use in skin and respiratory infections. This compound has excellent antibacterial activity against Gram-positive pathogens with MIC_{90} values of 0.12 μg/ml for *Streptococcus pneumoniae* and 0.25 μg/ml for methicillin-resistant *Staphylococcus aureus* (MRSA) where the majority of isolates were ciprofloxacin-resistant [5]. Improved activity against enteric and non-fermenter Gram-negative isolates was not observed although MICs were comparable or lower than those obtained with moxifloxacin. The enhanced antibacterial activity against existing fluoroquinolone-resistant Gram-positive strains is believed to be due to relatively balanced and potent activity against both the DNA gyrase and topoisomerase IV target enzymes in these organisms. This balanced potent inhibition of two essential bacterial targets resulted in a lower potential to select for resistant mutants as might be predicted.

Nemonoxacin, a novel non-fluorinated quinolone under development by Taigen Biotechnology (Fig. 2, **2**), possesses improved in vitro antibacterial activity against Gram-positive pathogens as well as *Haemophilus influenzae* and *Moraxella catarrhalis* compared to ciprofloxacin and levofloxacin [6] although MIC_{90}s against *Enterococcus faecalis* and *E. faecium* were 4 and 8 μg/ml, respectively. It also displayed improved antibacterial activity over moxifloxacin against quinolone-resistant MRSA (MIC_{90} of 1 versus 4 μg/ml) and *S. pneumoniae* (MIC_{90} of 2 versus 8 μg/ml). Antibacterial activity against Gram-negative bacilli is comparable to levofloxacin and ciprofloxacin [7] with limited effectiveness against *Pseudomonas aeruginosa* and *Acinetobacter baumannii* [6]. Efficacy has been demonstrated in several mouse infection models including systemic, lung, and urinary tract infections against several bacterial pathogens including MRSA, *S. pneumoniae, E. faecalis,* and *Escherichia coli* [8]. In Phase 1 clinical studies, nemonoxacin was found to be safe and well-tolerated with favorable pharmacokinetics after oral administration [9].

Another fluoroquinolone in later stages of clinical development is delafloxacin (Fig. 2, **3**), licensed by Wakunaga to Rib-X Pharmaceuticals. This compound was also developed for a time by Abbott as ABT-492 [10]. Delafloxacin displays excellent antibacterial activity against Gram-positive organisms, including both methicillin-susceptible *S. aureus* and MRSA, with MIC_{90}s of 0.008 and 0.5 μg/ml, respectively [10]. Excellent antibacterial activity was also observed against streptococci including quinolone-resistant pneumococci (MIC_s $\leq$0.03 μg/ml). Delafloxacin possesses antibacterial activity against Gram-negative pathogens although reduced against quinolone-resistant isolates and especially against non-fermenters. Twelve Phase 1 and three Phase 2 clinical trials involving more than 1300 individuals have been completed with oral and intravenous formulations for community-acquired pneumonia (CAP) and skin and skin structure infection (SSSI) indications [11]. These studies have shown delafloxacin to be both clinically efficacious and safe.

2.1.2. Topoisomerase/gyrase inhibitors

NXL101 (Fig. 2, **4**) is representative of a new class of quinoline DNA gyrase and topoisomerase IV inhibitors with a Gram-positive spectrum of antibacterial activity including MRSA and fluoroquinolone-resistant isolates with MIC_{90}s of $\leq$1 μg/ml [11]. Efficacy was demonstrated in mouse sepsis and tissue infections with MRSA including fluoroquinolone-resistant strains [13]. Interestingly, this compound displayed target preference profiles opposite those of fluoroquinolones, i.e., there was a preference for gyrase over topoisomerase in staphylococci with the converse observed for *E. coli*. In addition, different mutations were selected in the quinolone

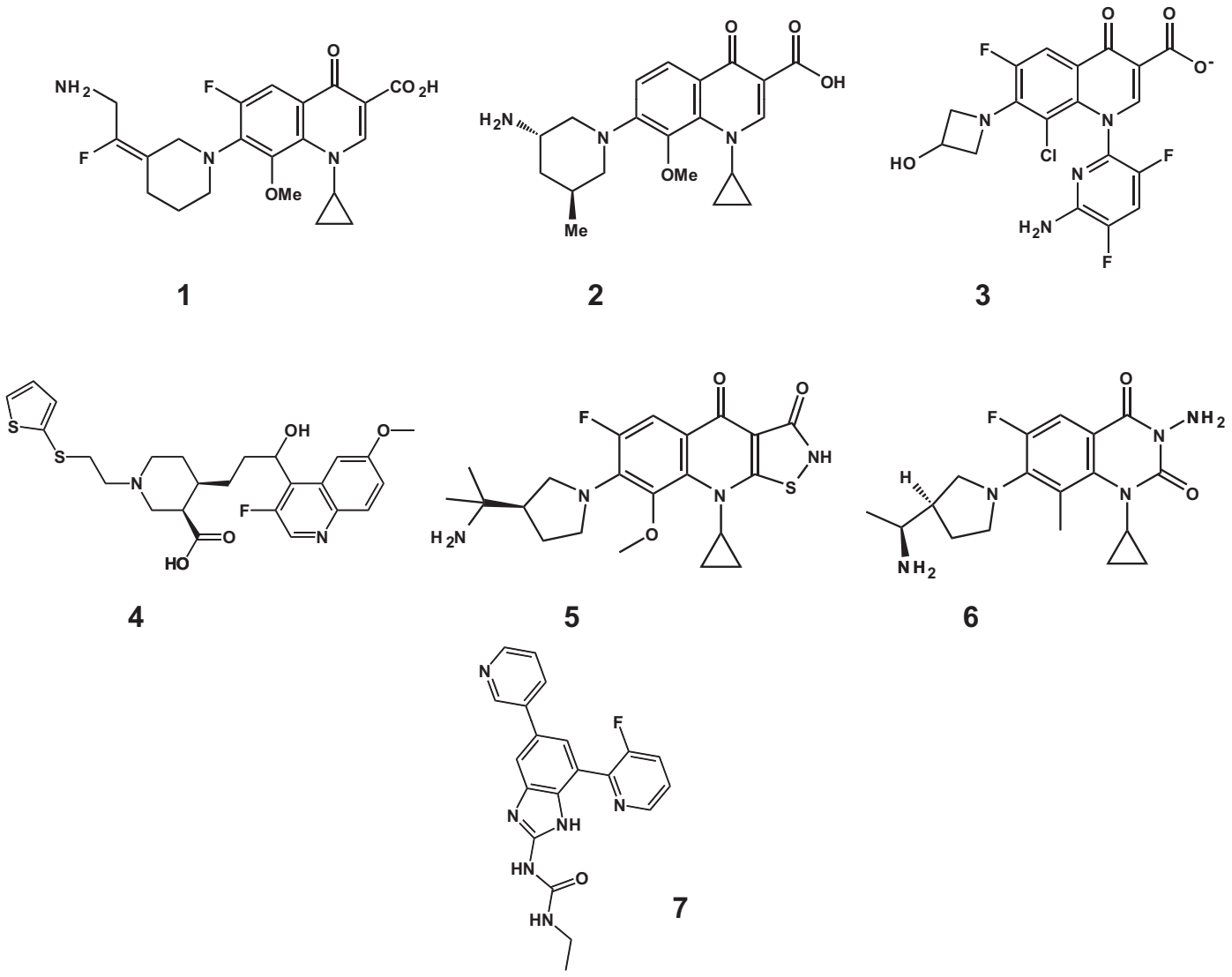


Fig. 2. Structures of bacterial DNA topoisomerase inhibitors. Compounds: **1**, JNJ-Q2; **2**, nemonoxacin; **3**, delafloxacin; **4**, NXL101; **5**, ACH-702; **6**, PD0326448; **7**, VRT-752586.

resistance determining regions (QRDRs) [14] than those reported for fluoroquinolones. The antibacterial activity profile and apparent lack of or reduced cross-resistance with existing fluoroquinolone strains resulted in the initiation of Phase 1 clinical trials by Novexel in 2006 for hospital-based infections including CAP and SSTI caused by Gram-positive pathogens. There was also the potential for both oral and intravenous dosage formulations. Clinical trials were discontinued in 2008 due to QT interval prolongation observed in healthy human subjects during early clinical development [15]. It is unclear whether other candidates from this class are currently under investigation.

ACH-702 (Fig. 2, **5**) is an isothiazoloquinolone (ITQ) structurally related to the quinolone class [16]. This compound possesses potent antibacterial activity against Gram-positive bacterial pathogens including quinolone-resistant strains and many Gram-negative organisms as well although activity is limited against quinolone-resistant Enterobacteriaceae and non-fermenters [17]. It is particularly active against MRSA including quinolone-resistant isolates with an MIC_{90} of 0.25 μg/ml. The mechanism of action of ITQs was determined in staphylococci and found to be based on excellent balanced inhibition of both bacterial DNA gyrase and topoisomerase IV, essentially allowing it to act as a dual target inhibitor. This probably accounts for the effectiveness of ACH-702 against quinolone-resistant strains and the low propensity to select for resistant mutants. The compound displayed efficacy in three mouse infection models and currently in development for topical indications due to extensive metabolism after systemic administration [17].

Researchers from Pfizer Global Research and Development recent described a new class of bacterial gyrase and topoisomerase inhibitors designated quinazolinediones (QDs) also structurally related to the quinolones [18,19]. PD 0305970 and PD0326448 (Fig. 2, **6**) were developed as new antibacterial agents that were orally active and covered some gaps in coverage of the quinolone class. QDs were found to target the same enzymes (gyrase and topoisomerase IV) as the quinolones but at sites on different subunits: GyrB and ParE versus GyrA and ParC [20]. These compounds display highly potent in vitro and in vivo activities versus susceptible and resistant Gram-positive and fastidious pathogens with an MIC_{90} of 0.25 μg/ml reported for highly quinolone-resistant isolates of *S. aureus* [18]. Activity versus Gram-negative organisms was generally less than that seen with quinolones probably at least in part due to efflux [18,19]. Efficacy against *Streptococcus pyogenes* and *E. faecalis* was demonstrated in a mouse sepsis model [21]. The current development status of these compounds is unclear at present.

Another new class of bacterial DNA gyrase and topoisomerase IV inhibitors, the aminobenzimidazoles as exemplified by VRT-752586 (Fig. 2, **7**), was recently reported by Vertex Pharmaceuticals. Potent antibacterial activity was observed against susceptible and antibiotic-resistant staphylococci, enterococci, streptococci with MIC_{90}s ranging from 0.5 to 4 μg/ml, and against the respiratory Gram-negative pathogens, *H. influenzae* and *M. catarrhalis*, although some *H. influenzae* isolates displayed MICs of >16 μg/ml [22]. The spectrum of activity suggested utility in both nosocomial and community settings including use for SSSI and respiratory infections caused by drug-susceptible and -resistant pathogens. The compound exhibited good pharmacokinetic properties in animals and demonstrated efficacy in animal models of infection [22]. VRT-752586 possesses dual targeting activities in *S. aureus*, *E. faecalis*, and *S. pneumoniae* inhibiting both the GyrB and ParE subunits of DNA gyrase and topoisomerase IV, respectively. The essential ATPase activities of these enzymes are required for the strand-passage reaction and represent a mechanism of action distinct from the fluoroquinolones for antibacterial drug discovery. This dual targeting mechanism results in excellent antibacterial activity and low frequency of resistance development at concentrations that would be expected in clinical situations [23]. Also, since there are no currently marketed antibiotics targeting GyrB/ParE ATPase activity, compounds such as VRT-752586 or derivatives should not be cross-resistant with fluoroquinolones or any other antibiotics. Therefore, the aminobenzimidazoles represent a promising new class of antibacterial agents for combating the problem of drug resistance that inhibit a relatively less exploited target. However, a 16-fold shift in MICs was observed in the presence of 50% human serum [22], suggesting that high protein binding may affect in vivo potency. Pharmacokinetic/pharmacodynamics studies in animal models should provide insights regarding the actual effects on bacterial infections. The current development status of this compound class is unknown at present.

2.2. Fatty acid biosynthesis inhibitors

Bacterial fatty acid biosynthesis is an essential process that is involved in the formation of such cellular constituents as lipopolysaccharides, phospholipids, lipoteichoic acid, and lipoproteins. Inhibition of this pathway leads to disruption of membrane structure and function and cell death. This fatty acid synthase II (FASII) pathway in bacteria is quite distinct from that in mammalian cells (FASI) suggesting the possibility of inhibitor selectivity. Fatty acid biosynthesis enzymes in bacteria have been validated as targets for antibacterial drugs in that there are several examples of inhibitors that have effects on both the targets in vitro and on the growth of bacterial cells [24,25]. These include compounds such as cerulenin, thiolactomycin, isoniazid, triclosan, and, most recently, platensimycin and platencin [26,27]. However, the validity of bacterial fatty acid biosynthetic enzymes as drug targets has recently been challenged [28] as data were reported that showed that streptococci and staphylococci could utilize unsaturated fatty acids from human serum in sufficient quantities as to render de novo synthesis of fatty acids unnecessary for survival. In addition, FASII knock-out mutants in *Streptococcus agalactiae* were found to grow in the presence of human serum or exogenous fatty acids. Thus, FASII as a therapeutic target is a controversial issue, as the clinical significance for these observations is unknown. Clearly, more basic research is warranted prior to major resource investment into these targets.

However, others have continued research and development of inhibitors of fatty acid biosynthesis. Recently, a compound from Affinium Pharmaceuticals, AFN-1252 (Fig. 3, **8**), entered Phase 1 human clinical trials as an oral anti-staphylococcal antibiotic. This compound is closely related to inhibitors previously described by researchers at GlaxoSmithKline [29] and targets FabI, an essential enzyme that catalyzes the reduction of trans-2-enoyl-acyl carrier protein (trans-2-enoyl-ACP) to acyl-ACP, the final step in elongation

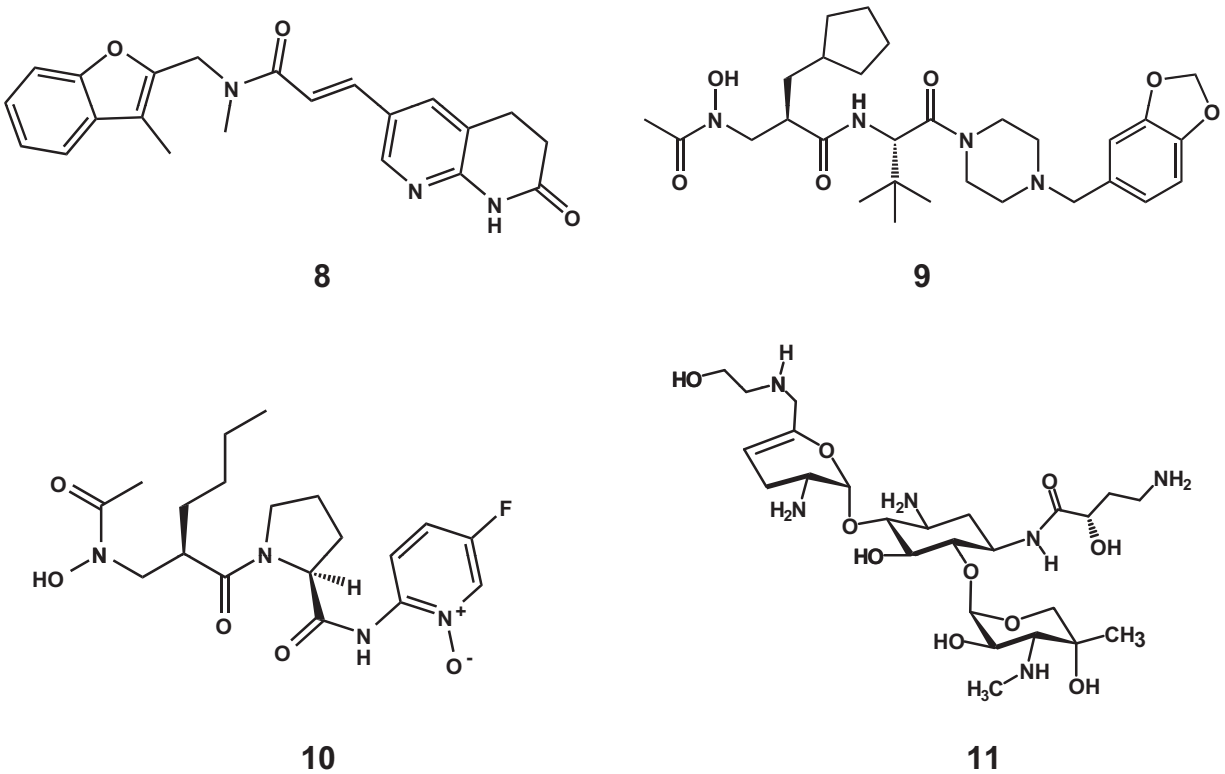


Fig. 3. Structures of fatty acid biosynthesis inhibitor, peptide deformylase inhibitors, and neoglycoside. Compounds: **8**, AFN-1252; **9**, BB 83698; **10**, LBM415; **11**, ACHN-490.

cycle for bacterial fatty acid biosynthesis [25]. However, AFN-1252 has a narrow antibacterial spectrum as alternative enzymes or rescue pathways are present in other pathogens [30]. FabI is the sole enoyl-ACP reductase present in staphylococci, *H. influenzae*, *M. catarrhalis*, and *E. coli*. AFN-1252 was identified through an iterative structure-guided chemistry effort and optimized for the staphylococcal enzyme. It displayed excellent in vitro selectivity with an IC_{50} >100 μM for human FASI and 14 nM for *S. aureus* FabI.

AFN-1252 inhibited all methicillin-susceptible and methicillin-resistant isolates of *S. aureus* and *Staphylococcus epidermidis* tested at concentrations ≤0.12 μg/ml with MIC_{90}s of ≤0.008 μg/ml for multi-drug resistant strains [31]. Interestingly, this compound was less active against vancomycin-resistant than vancomycin-susceptible *S. aureus* isolates (MIC_{90}s of 0.06 versus ≤0.008 μg/ml, respectively). As expected from the target profile, AFN-1252 was inactive against non-staphylococcal Gram-positive pathogens and Gram-negative pathogens with MICs ≥4 μg/ml. However, despite the possession of FabI as their sole enoyl-ACP reductase enzyme, *M. catarrhalis*, *E. coli*, and *Klebsiella pneumoniae* were non-susceptible to AFN-1252 possibly due to efflux [31]. AFN-1252 has demonstrated in vivo efficacy in a murine subcutaneous abscess model using a strain of MRSA [32] and in other animal infections including the thigh infection model [33].

2.3. Protein synthesis inhibitors

Inhibition of bacterial protein synthesis has yielded several important classes of antibacterial agents including the macrolides, tetracyclines, and, more recently, the oxazolidinones. Although primarily bacteriostatic, these drugs have been invaluable in the treatment of bacterial infections for decades. Recent advances in our knowledge of prokaryotic ribosome structures have provided new opportunities to identify additional bacterial protein synthesis inhibitors.

2.3.1. Peptide deformylase inhibitors

Bacterial peptide deformylase (Pdf), encoded by the *def* gene, is the enzyme that catalyzes the removal of the N-formyl group from the N-terminal methionine following translation during protein synthesis. This enzyme has several properties that support that it is a good antibacterial target [34,35]. Pdf is present in all pathogenic bacteria and *def* has been shown to be an essential gene for bacterial growth and survival in both Gram-negative and Gram-positive bacteria [36]. There is also no functional homologue in mammalian cells which suggests that inhibitors would have a good chance for selectivity against bacteria, although a human mitochondrial homolog of Pdf has been reported that is inhibited by inhibitors of the bacterial enzyme [37]. A major area of concern is that mutants that lack Fmt, the transformylase that formylates methionine can survive in the laboratory [38] thus predicting the possibility for rapid resistance development against novel inhibitors. At least two compounds, BB 83698 ([39]; Fig. 3, **9**) and LBM415 ([40]; Fig. 3, **10**), were previously advanced into Phase 1 clinical trials, however, development of both were halted in 2004 for reasons that were not reported [41].

Despite the potential risks suggested by the prior failure of two Pdf inhibitor compounds to advance in clinical development and potential resistance emergence issues, efforts have continued to find new inhibitors of this antibacterial target. Researchers at GlaxoSmithKline have recently described GSK1322322, a 4-hydrazinopyrimidine whose chemical structure has not been published to date, which was the result of a structure-based design program. Structural information was used to design multiple non-peptidic inhibitors that were optimized in an iterative chemistry effort that resulted in clinical candidate GSK1322322. This compound showed good in vitro antibacterial activity against Gram-positive pathogens *S. aureus, S. pneumoniae*, and *S. pyogenes* and the Gram-negative respiratory pathogens *H. influenzae* and *M. catarrhalis* with MICs ranging from 0.5 to 4 μg/ml [42]. Efficacy was observed in a rat respiratory infection model against both *S. pneumoniae* and *H. influenzae* and in a mouse groin abscess model against MRSA after oral administration. This compound was safe and well-tolerated in Phase 1 clinical studies and has advanced to Phase 2a clinical trials focused on acute bacterial skin and skin structure (abSSSI) infections [43].

2.3.2. Aminoglycosides

Ever since the discovery of streptomycin in the 1940s, the aminoglycoside class of antibiotics has been an important component of our antibacterial arsenal [44]. Their mechanism of action involves binding to the 16S rRNA subunit of the 30S bacterial ribosome at the A site and interfering with normal protein synthesis. They have been particularly valuable in the treatment of infections caused by Gram-negative pathogens; however, their usefulness has been eroded over the years by increased resistance and by toxicity issues. Their spectrum of activity, bactericidal properties, and ability to act synergistically with other antibiotic classes has led to a renewal of research into the development of new and improved aminoglycoside antibiotics [44]. At least one of these newer aminoglycosides has advanced into human clinical trials.

ACHN-490 (Fig. 3, **11**), a next generation neoglycoside under development at Achaogen, Inc., is a semi-synthetic compound derived from sisomicin [45]. Structural modifications render this compound resistant to the effects of aminoglycoside modifying enzymes (AMEs) thus allowing for activity against aminoglycoside-resistant bacterial isolates. In fact, virtually all AMEs that modify gentamicin and amikacin do not affect ACHN-490 in a variety of bacterial species [46]. Included in the Gram-negative pathogen coverage are isolates that possess extended-spectrum β-lactamases (ESBLs), AmpC cephalosporinases, carbapenemases, and metallo-β-lactamases as well as fluoroquinolone resistance [45]. MIC_{90} values for *E. coli, Klebsiella* spp., and *Enterobacter* spp. were 1–2 μg/ml. However, MIC_{90}s for *Proteus mirabilis*, *Acinetobacter* spp., and *P. aeruginosa* were 8, 32, and 64 μg/ml, respectively. Efflux is believed to account for the higher MICs observed in these Gram-negative pathogens [46]. However, resistance has also been reported in isolates of Enterobacteriaceae harboring ArmA and RmtC 16S rRNA methylases [47]. The MIC_{90} was found to be 2 μg/ml for staphylococci.

Achaogen recently reported data that demonstrated the efficacy of ACHN-490 in a murine model of a urinary tract infection with uropathogenic *E. coli* (UPEC) and indicated that further testing using drug-resistant UPEC would provide additional non-clinical data to support utility in the treatment of UTIs caused by resistant isolates [48]. These results, combined with the fact that ACHN-490 produced high C_{max} and AUC values in human Phase 1 trials, suggest that this compound has the potential to be an effective therapeutic option for UTIs caused by UPEC and other pathogens. ACHN-490 has advanced into Phase 2 clinical trials for pneumonia and cUTI [49].

SelectX Pharmaceuticals has also reported the development of a new aminoglycoside, SXP2523, which is claimed to be significantly differentiated from any other Gram-negative antibiotics either in development or on the market [50]. This compound has potent antibacterial activity against MDR Gram-negative clinical isolates including *P. aeruginosa*, *K. pneumoniae*, *A. baumannii*, and *E. coli*, as well as *S. aureus* (MIC_{90}s < 4 μg/ml). Other desirable properties include bactericidal with rapid killing kinetics, low frequencies of resistance selection, and synergy with β-lactam and glycopeptide antibiotics. Efficacy has been demonstrated in several animal infection models. SXP2523 is also predicted to have an excellent

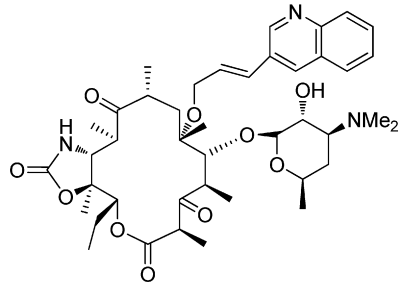
safety margin and therapeutic index. The structure of this compound has not been made public and the current stage of development is not clear.

2.3.3. Macrolides

New macrolides that are currently under development are 14-membered ketolides, with a 3-ketone substitution rather than the 3-cladinose found in macrolides such as erythromycin (Fig. 4). Ketolides are noted for their potent activity against Gram-positive respiratory pathogens, especially against the erythromycin-resistant pneumococci containing *mef*E and *erm*B mutations [51].

Cethromycin (ABT-773, Fig. 4, **12**) from Advanced Life Sciences is a once-a-day orally bioavailable ketolide with an NDA (New Drug Application) considered for the treatment of outpatient community acquired bacterial pneumonia (CABP) [52]. In addition, the FDA has granted it orphan drug status for biodefense indications including anthrax, plague and tularemia. Its spectrum of activity is similar to other ketolides like telithromycin, covering both Gram-negative and Gram-positive respiratory pathogens [53]. In vitro studies showed that cethromycin MICs tended to be 2–4-fold lower than telithromycin MICs against recent pneumococcal isolates [54]. Both ketolides were 2-fold less potent than azithromycin against the Gram-negative respiratory pathogens *M. catarrhalis* and *H. influenzae* [51]. Full development studies of cethromycin have been completed, including two double-blind non-inferiority clinical trials conducted in mild- to moderate-community-acquired pneumonia with clarithromycin as the comparator [52]. Although an FDA Advisory Committee voted in June 2009 that the drug was safe, the efficacy was questioned, with suggestions that a superiority trial be designed, enrolling more patients with macrolide-resistant pathogens, using mortality as an endpoint [54]. However, no new trials are currently in progress [43].

Solithromycin (CEM-101), a fluoroketolide with fluorine substituted at position 1 (Fig. 4, **14**), demonstrated potent activity in vitro against common pathogens associated with community-acquired respiratory diseases or skin infections [55]. When tested against macrolide-susceptible and macrolide-resistant pneumococci, solithromycin MIC ranges were 0.002–0.016 μg/ml against macrolide-susceptible pneumococci and 0.004–1 μ/ml against recent macrolide-resistant *S. pneumoniae* isolates [56]. Like many ketolides, solithromycin displayed some activity against MRSA, with an MIC_{50}/MIC_{90} values of 0.06 and



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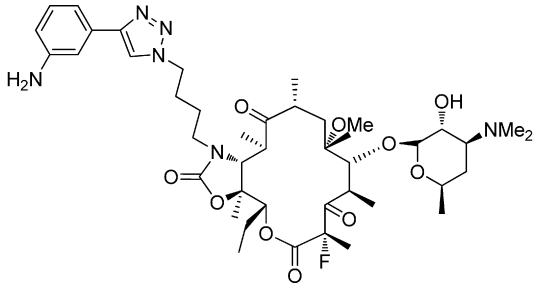

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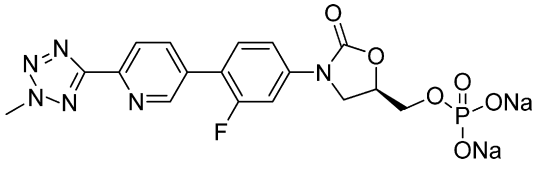

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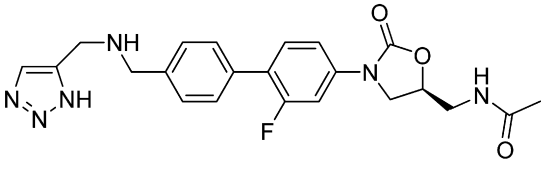

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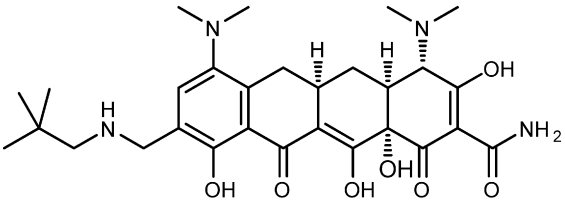

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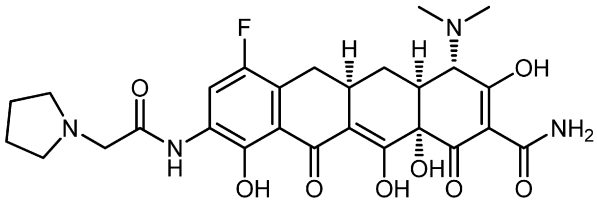

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Fig. 4. Structures of ketolides, oxazolidinones and tetracyclines. Compounds **12**, cethromycin; **13**, solithromycin; **14**, torezolid phosphate; **15**, radezolid; **16**, omadacycline (PTK0796); **17**, TP-434.

>4 mg/ml, respectively [55]. Solithromycin has successfully completed preclinical testing and escalating dose Phase 1 studies, and is currently entering Phase 2 therapeutic trials under the direction of Cempra Pharmaceuticals [57].

2.3.4. Oxazolidinones

Oxazolidinones are bacteriostatic agents with activity primarily against Gram-positive cocci, although some compounds in the class have demonstrable activity against *H. influenzae*. The only compound that is clinically available in this class is linezolid; when it was approved in 2000 by the FDA, it represented the first new molecular class of antibacterial agent in over 20 years. Linezolid has remained relatively robust in its low selection of resistance in Gram-positive cocci, although it has been recently challenged with the identification of a mobile methyltransferase that confers resistance by methylating C-8 of the 23S rRNA base A2503 at the site of oxazolidinone inhibitory activity [58].

Torezolid (TR-700, DA-7157, DA-70157), a novel oxazolidinone is the active component of torezolid phosphate (TR-701, DA-7218, DA-70218), licensed by Trius Pharmaceuticals from Dong-A Pharmaceuticals, a South Korean company. Torezolid MICs tend to be 4–8-fold lower than linezolid MICs against staphylococcal, enterococcal and streptococcal isolates [59]. Both oxazolidinones have MIC_{50} values $\geq$ 8 mg/ml against most strains of *H. influenzae*. Following acceptable preclinical evaluations, the compound successfully completed Phase 1 dose-ranging studies for IV and oral formulations [60] and a Phase 2 clinical trial in complicated SSSI with oral dosing [61].

Radezolid (RX-1741); a novel biaryloxazolidinone from Rib-X Pharmaceuticals (Fig. 4, **15**), has recently completed two Phase 2 clinical trials for CAP and for uncomplicated SSSI [62]. In vitro potency of this oxazolidinone is generally 2–4-fold greater than linezolid against Gram-positive cocci; against *H. influenzae* radezolid MIC_{50}/MIC_{90} values were 0.5/1 μg/ml, at least 16-fold more potent than linezolid [63]. When tested against *S. aureus* clinical isolates bearing the *cfr* methyltransferase, linezolid MICs were 16–64 μg/ml compared to wild type MICs for linezolid of $\leq$2 μg/ml; the highest oxazolidinone MICs were associated with the presence of both the *cfr* gene and mutations in ribosomal protein L3 [58]. In contrast to linezolid, MICs for the investigational oxazolidinones were increased less dramatically. Radezolid MICs ranged from 2 to 8 μg/ml against these linezolid-resistant isolates, whereas torezolid MICs did not exceed 2 μg/ml for any of these strains [58]. Both torezolid and radezolid are being developed for intravenous (IV), as well as oral, usage [60,62].

2.3.5. Tetracyclines

Tetracyclines, broad-spectrum bacteriostatic antibiotics that inhibit bacterial growth by binding to the 30S ribosome, have been a staple of the antibacterial armamentarium for over 50 years. Widespread resistance to the original set of tetracyclines emerged due to ribosomal protection or efflux mechanisms. However, in the past decade, a new family of tetracyclines has been developed, based on the glycylcyclines as exemplified by tigecycline [47]. As shown below, these new tetracyclines can evade common resistance mechanisms while retaining the same activity spectrum as tigecycline against both Gram-positive and Gram-negative pathogens. None of these new protein synthesis inhibitors, however, have useful activity against *P. aeruginosa* [64].

Omadacycline (PTK0797), an aminomethylcycline (Fig. 4, **16**), was discovered by scientists at Paratek and is being co-developed with Novartis. Its broad spectrum of activity is similar to that of tigecycline in vitro, with potent bacteriostatic activity against Gram-positive cocci including MRSA, multidrug-resistant *S. pneumoniae* and vancomycin-resistant enterococci. MICs of 0.5–8 mg/ml were reported for susceptible and drug-resistant *E. coli*, *K. pneumoniae* and *H. influenzae* [65,66]. Although 75% of the *K. pneumoniae* and *A. baumannii* tested had PTK0796 MICs <4 μg/ml, MICs were uniformly $\geq$ 8 μg/ml against the Proteae and *P. aeruginosa* [66]. Omadacycline is currently in Phase 3 comparative clinical studies for the treatment of complicated SSSI [43]. Because it has the advantage of being orally bioavailable, it can be developed for use in both hospital and community settings where IV and oral formulations provide dosing versatility [67].

TP-434 [68] is a fluorocycline from Tetraphase that has entered Phase 2 clinical trials for the treatment of community-acquired complicated intra-abdominal infections (cIAIs) [69]. In vitro studies demonstrated that TP-434 inhibited protein synthesis equally in the presence and absence of the ribosomal protection mechanism Tet(M) [70]. TP-434 MICs against MRSA (n = 137) and vancomycin-resistant *E. faecium* (n=43) isolates were reported as $\leq$ 0.5 μg/ml [22]. MIC_{90} values were $\leq$ 2 μg/ml against a set of 372 Enterobacteriaceae isolates, including *K. pneumoniae* strains producing carbapenemases and extended-spectrum β-lactamases (ESBLs) [70]. Supportive in vivo data in animal models of septicemia, pyelonephritis and lung infections [71] suggest that this fluorocycline may be useful in treating some of the more recalcitrant infections caused by both multidrug-resistant enteric bacteria and Gram-positive cocci. An oral formulation that is being investigated [69] would add to the attractiveness of this agent.

2.4. Cell wall inhibitors

The bacterial cell wall has been an attractive and effective drug target throughout the history of antibiotics. Agents that interfere with bacterial cell wall synthesis include glycopeptides and the β-lactam antibiotics. Although Gram-positive bacteria have become resistant to these agents primarily through mutations in their killing targets, the penicillin-binding proteins (PBPs), resistance to the β-lactams in Gram-negative bacteria is most frequently due to the production of inactivating enzymes [72]. Unfortunately, the efficacy of the β-lactams, including the penicillins, cephalosporins, monobactams and carbapenems, has been severely compromised by the rapid expansion of hydrolytic capabilities of over 1000 β-lactamase variants with broad-spectrum activities [73]. As a result, new β-lactams, or β-lactamase inhibitors, have been designed that can counteract the hydrolytic activity of multiple β-lactamases, several of which may appear in a single bacterial strain.

2.4.1. Monocyclic β-lactams

Although monocyclic β-lactams have been described in the literature for over 30 years, only the monobactam aztreonam has been widely used clinically, and with only limited commercial success. Nonetheless, the exclusively Gram-negative spectrum of monobactams is still attractive in this era of infections caused by multidrug-resistant Gram-negative pathogens. The stability of monobactams in the presence metallo-β-lactamases (MBLs) has prompted the study of combinations of β-lactams that may be useful in treating infections caused by a variety of β-lactamase-producing organisms, including those with transferable zinc-containing β-lactamases [74].

BAL 30376 (Fig. 5, **18**, **19**, **20**) was a unique triple combination from Basilea Pharmaceutica that utilized the siderophore monobactam BAL19764 (Fig. 5, **18**) for rapid entry into Gram-negative bacteria by using iron uptake pathways. BAL19764 was formerly known as Syn2416 which originated from SynPhar Laboratories, now NAEJA Pharmaceuticals. This siderophore monobactam, with lability to hydrolysis by AmpC cephalosporinases and ESBLs, was combined with the bridged monobactam BAL29880 (Fig. 5, **19**)and with clavulanic acid (Fig. 5, **20**) to inhibit AmpC enzymes and

Table 1
Antibacterial spectrum of activity for new antimicrobial agents.

Antimicrobial class	Compound	Spectrum of activity				Reference
		Gram-positive	Respiratory Gram-negative/Atypicals	Enteric Gram-negative	Non-fermenters	
Fluoroquinolones	JNJ-Q2	*S. aureus*/MRSA MDR[a]-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	Fluoroquinolone-susceptible strains	NA[b]	[5]
	Nemonoxacin	Fluoroquinolone-susceptible *S. aureus* MDR-streptococci	*H. influenzae*	Fluoroquinolone-susceptible strains	Fluoroquinolone-susceptible strains	[7]
	Delafloxacin	*S. aureus*/MRSA MDR-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	Fluoroquinolone-susceptible strains	Fluoroquinolone-susceptible strains	[10]
Topoisomerase/gyrase inhibitors	NXL101	*S. aureus*/MRSA MDR-streptococci	NA	NA	NA	[12]
	ACH-702	*S. aureus*/MRSAMDR-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	Fluoroquinolone-susceptible strains	Fluoroquinolone-susceptible strains	[17]
	Quinazoline-diones	*S. aureus*/MRSA MDR-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	Some strains	Some strains	[18]
	VRT-752586	*S. aureus*/MRSA MDR-streptococci	*M. catarrhalis*	NA	NA	[22]
Fatty acid synthesis inhibitors	AFN-1252	*S. aureus*/MRSA	NA	NA	NA	[30,31]
Peptide deformylase inhibitors	GSK1322322	*S. aureus*/MRSA MDR-streptococci	*H. influenzae* *M. catarrhalis*	NA	NA	[42]
Aminoglycosides	ACHN-490	*S. aureus*/MRSA	ND[c]	Not *P. mirabilis*, indole+ *Proteae*	Aminoglycoside-susceptible	[46,47]
	SXP2523	*S. aureus*/MRSA	ND	Many strains	Many strains	[50]
Macrolides	Cethromycin	*S. aureus*/some MRSA MDR-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	NA	NA	[51,52,54]
	Solithromycin	*S. aureus*/some MRSA MDR-streptococci	*H. influenzae* *L. pneumophila* *M. catarrhalis*	NA	NA	[55–57]
Oxazolidinones	Torezolid	MDR-enterococci/VRE *S. aureus*/MRSA MDR-streptococci	*Chlamydia pneumoniae* *L. pneumophila*	NA	NA	[59,60]
	Radezolid	MDR-enterococci/VRE *S. aureus*/MRSA MDR-streptococci	*H. influenzae* *M. catarrhalis*	NA	NA	[63]
Tetracyclines	Omadacycline	MDR-enterococci/VRE *S. aureus*/MRSA MDR-streptococci	ND	Many strains No *Proteae*	ND	[65,66]
	TP-434	MDR-enterococci/VRE *S. aureus*/MRSA MDR-streptococci	*L. pneumophila*	Many strains	ND	[69,70]
tRNA synthetase inhibitors	GSK'052	ND	ND	All strains tested	Some strains of *P. aeruginosa*	[88]
Monocyclic β-lactams	BAL30376	NA	ND	Many strains unless multiple AmpC/ESBLs	Many strains of *P. aeruginosa*	[72,75]
	BAL30072	NA	ND	Non-ESBL producing enterics	*P. aeruginosa*, including MBLs[d]	[74]

Table 1 (*Continued*)

Antimicrobial class	Compound	Spectrum of activity				Reference
		Gram-positive	Respiratory Gram-negative/Atypicals	Enteric Gram-negative	Non-fermenters	
β-lactamase inhibitor combinations	NXL104 + ceftazidime	NA	ND	ESBL- and serine carbapenemase-producing enterics	Many strains of *P. aeruginosa*	[77,78]
	NXL-104 + ceftaroline	*S. aureus*/MRSA MDR-streptococci	ND	ESBL- and serine carbapenemase-producing enterics	NA	[79]
	CXA-101 + tazobactam	Group A and Group B streptococci	*H. influenzae*	Many ESBL- and serine carbapenemase-producing enterics	Most strains of *P. aeruginosa*	[80,81]
	MK-7655 + imipenem	MDR-streptococci	*H. influenzae* *M. catarrhalis*	ESBL- and serine carbapenemase-producing enterics	Most strains of *P. aeruginosa*	[82]

[a] MDR, multidrug resistant.
[b] NA, no useful activity.
[c] ND, no data reported.
[d] MBL, metallo-β-lactamase.

ESBLs, respectively [75]. Although microbiological activity was noted for some MBL-producing strains, higher MICs were observed for organisms that produced multiple β-lactamases in different classes. This combination does not seem to be under further development, but the concept of a combination drug with multiple β-lactamase inhibitors remains of interest.

More recently, the siderophore monosulfactam BAL30072 (Fig. 4, **21**), also from Basilea, was designed to act as a "Trojan horse" agent that readily enters the Gram-negative cell using an iron uptake system before binding to essential cell wall synthesizing enzymes and causing cell death [72]. Although iron transport is involved in cell entry, the broad spectrum Gram-negative antibacterial activity of the sulfactam is dependent upon binding to several bifunctional PBPs that cause rapid cell death and lysis. The spectrum of activity of this compound includes many non-ESBL-producing Enterobacteriaceae, as well as *P. aeruginosa*, including strains that produce metallo-β-lactamases. MIC_{50}/MIC_{90} values for Bal30072 against *A. baumannii* isolates were 0.5 and 4 μg/ml, respectively [74]. Unlike other siderophore-bearing monocyclic β-lactams, resistance selection was very low, as the sulfactam does not select for mutants in the iron uptake pathways, reinforcing the observation that it must not depend solely upon iron uptake for its antibacterial activity [74]. BAL30072 has entered Phase 1 dose-ranging studies [43].

A second siderophore monocyclic β-lactam, MC-1 has been introduced by Pfizer scientists [76] In vitro data indicated that this monocarbam also had broad-spectrum Gram-negative activity including many resistant isolates of Enterobacteriaceae and *P. aeruginosa*. Its development status is not known.

2.4.2. β-Lactamase inhibitor combinations

Combinations of a β-lactam with a β-lactamase inhibitor have been successfully used to combat β-lactamase-mediated resistance since the introduction of clavulanic acid combinations in the mid-1980s, followed by sulbactam and tazobactam combination drugs. However, the emergence of so many recent β-lactamase variants, appearing in multiple forms within a single organism, has made these early combinations ineffective. In order to counteract the effects of enzymes that may hydrolyze multiple classes of β-lactams, many companies are continuing to develop new inhibitors for these enzymes.

NXL104 is a novel bicyclic, non-β-lactam β-lactamase inhibitor initially developed by Novexel that is now under clinical development through a collaborative agreement between AstraZeneca and Forest Laboratories (Fig. 5, **22**). This inhibitor has broad spectrum inhibitory activity that includes class A and class C β-lactamases, including the class A KPC carbapenemase family of enzymes [77]. The NXL104-ceftazidime combination provides a drug with in vitro activity against multidrug-resistant Enterobacteriaceae that produce ESBLs and serine carbapenemases (KPC enzymes), as well activity against many *P. aeruginosa* isolates [78]. The NXL104-ceftaroline combination provides coverage against most of the same Enterobacteriaceae that do not express high enzyme levels; pseudomonal isolates are not sensitive, due to the intrinsically high ceftaroline MICs against *P. aeruginosa* [79]. However, due to the inherent activity of ceftaroline against MRSA, its combination with NXL104 provides an agent with potential clinical value against both Gram-positive and Gram-negative pathogens. NXL104 has been combined with ceftazidime in Phase 2 studies for the treatment of cUTI (complicated urinary tract infections) and combined with ceftaroline in Phase 2 studies for the treatment of cUTI and cIAI [43].

CXA-101 (FR 264205, Fig. 5, **23**) is an extended-spectrum cephalosporin that was selected for development based on its potent anti-pseudomonal activity [80]. Although the compound is

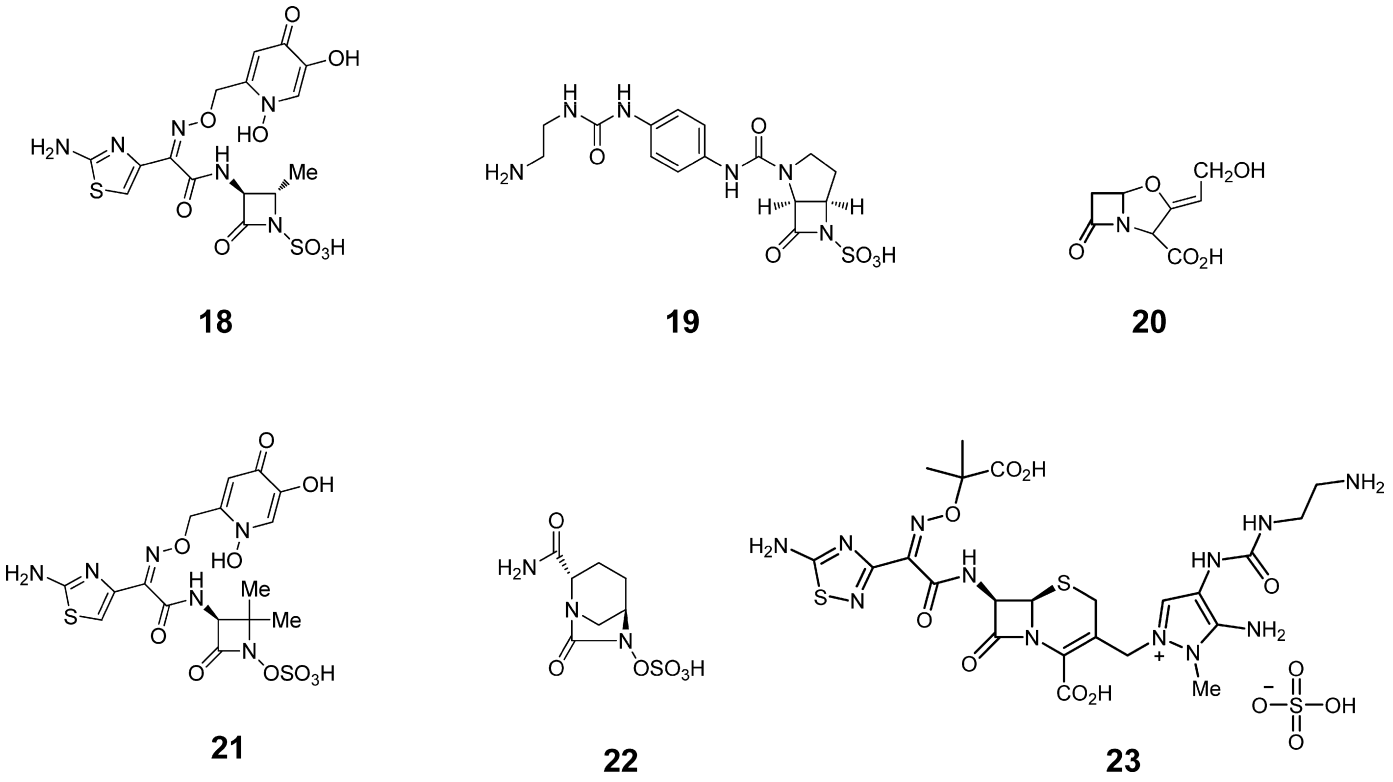

Fig. 5. Structures of cell wall inhibitors and β-lactamase inhibitors. Compounds BAL 30376, a triple combination of **18**, BAL 19764, **19**, BAL29880, and **20**, clavulanic acid; **21**, BAL 30072, **22**, NXL-104; **23**, CXA-101.

labile to hydrolysis by ESBLs and carbapenemases, activity against ESBL-producing Enterobacteriaceae can be restored in vitro by the addition of 8 μg/ml of tazobactam [81], an inhibitor that can be dosed at high levels. It is currently being studied in combination with tazobactam by Cubist Pharmaceuticals in trials initiated by Calixa Therapeutics. A Phase 2 clinical trial for the treatment of cUTI has been completed, with a Phase 2 trial in progress for treatment of cIAI [43].

MK-7655 is a class C and class A β-lactamase inhibitor that was first described publicly by scientists from Merck in September 2010 [82]. When tested in vitro at concentrations of ≤8 μg/ml, it restored the antibacterial activity of imipenem against class A (serine) carbapenemase-producing Enterobacteriaceae and multi-drug-resistant *P. aeruginosa* isolates [82]. Although the structure of MK-7655 has not been published by the Merck scientists, the structure is in the same family as the bicyclic NXL104 structure, and corresponds to a compound described in "Merck patent WO2009091856)" in the patent literature [83]. MK-7655 is currently undergoing Phase 1 clinical trials, with studies conducted in the presence of imipenem [43].

2.5. Efflux pump inhibitors

Efflux pumps are ubiquitous among Gram-negative bacteria, accounting for much of the intrinsic resistance observed for many antibiotic classes. Using an approach similar to the development of β-lactamase inhibitors, several companies have established programs to identify novel compounds that will inhibit specific pumps, especially those in the Resistance/Nodulation/Cell Division Family (RND) that affect many known antibiotics in Gram-negative organisms [84]. Mpex Pharmaceuticals has been at the forefront of the companies working in this area, with a variety of inhibitors of RND pumps that have been shown to potentiate the activity of β-lactams, oxazolidinones, fluoroquinolones and/or macrolides in Enterobacteriaceae or *P. aeruginosa* [85]. Compounds identified include narrow spectrum and broad spectrum inhibitors, with the clinical candidate MP-601,205 that entered Phase 1 clinical trials in cystic fibrosis patients [85]. Development, however, was discontinued due to tolerability issues, a potential problem with broad spectrum pump inhibitors that are non-specific in their action. Although no other clinical candidate has yet to be identified, Mpex scientists are currently working with GlaxoSmithKline to identify a lead compound for development.

2.6. Peptides

Higher order species including humans possess antimicrobial peptides (AMPs) that act as a first line of defense against microbial invaders. These will only be briefly mentioned here and were recently reviewed by Findlay et al. [86]. Many, but not all, AMPs work through selective membrane disruption that leads to depolarization, lysis, and cell death. Much work has been done in attempts to advance peptides as antimicrobial therapeutic agents so that there are now over 1200 peptides listed in the antimicrobial peptide database [87]. AMPs offer such advantages as lack of cross-resistance to existing agents and reduced development of resistance, but offer disadvantages in regard to pharmacokinetics, delivery, and toxicity. None to date have been successfully marketed as a systemic agent although topical applications seem quite promising. Research in this area is expected to be quite active for some time.

3. Commentary

Concern has been raised in the infectious disease community about whether we have reached the stage of untreatable infections [47]. Although the pharmaceutical industry has continued to shrink its efforts in antibiotic discovery and development, many

smaller companies are providing a diversity of compounds that are advancing into early stages of development (Table 1). Some of these new agents are based on well-established antibacterial drugs, whereas novel targets or novel molecular scaffolds represent an opportunity for innovative mechanisms to be the source for clinical development candidates. There is always substantial risk associated with inhibitors against targets that have not yet been clinically validated; more basic research would undoubtedly help with this issue. One should also consider the fact that inhibitors against single bacterial targets may not result in the most optimal drugs, due to rapid emergence of resistance as a result of point mutations in the target gene [89]. It is likely that there will be high attrition among the first compounds related to these newer approaches. Nonetheless, the impetus to develop high risk/high benefit drugs is encouraging. Although rate of resistance development may ultimately determine the utility of these new agents, alternative approaches continue to move forward.

References

[1] Boucher HW, Talbot GH, Bradley JS, Edwards JE, Gilbert D, Rice LB, et al. Bad bugs, no drugs: no ESKAPE! An update from the Infectious Diseases Society of America. Clin Infect Dis 2009;48:1–12.

[2] Bradbury BJ, Pucci MJ. Recent advances in topoisomerase inhibitors. Curr Opin Pharmacol 2008;8:574–81.

[3] Mitscher LA. Bacterial topoisomerase inhibitors: quinolone and pyridine antibacterial agents. Chem Rev 2005;105:559–92.

[4] Wiles JA, Bradbury BJ, Pucci MJ. New quinolone antibiotics: a survey of the literature from 2005 to 2010. Expert Opin Ther Pat 2010;20:1295–319.

[5] Morrow BJ, He W, Amsler KM, Foleno BD, Macielag MJ, Lynch AS, et al. In vitro antibacterial activities of JNJ-Q2, a new broad-spectrum fluoroquinolone. Antimicrob Agents Chemother 2010;54:1955–64.

[6] Lauderdale TL, Shiau YR, Lai JF, Chen HC, King CH. Comparative in vitro activities of nemonoxacin (TG-873870), a novel nonfluorinated quinolone, and other quinolones against clinical isolates. Antimicrob Agents Chemother 2010;54:1338–42.

[7] Adam HJ, Laing NM, King CR, Lulashnyk B, Hoban DJ, Zhanel GG. In vitro activity of nemonoxacin, a novel nonfluorinated quinolone, against 2440 clinical isolates. Antimicrob Agents Chemother 2009;53:4915–20.

[8] Li CR, Li Y, Li GQ, Yang XY, Zhang WX, Lou RH, et al. In vivo antibacterial activity of nemonoxacin, a novel non-fluorinated quinolone. Antimicrob Agents Chemother 2010;65:2411–5.

[9] Chung DT, Tsai CY, Chen SJ, Chang LW, King CH, Hsu CH, et al. Multiple-dose safety, tolerability, and pharmacokinetics of oral nemonoxacin (TG-873870) in healthy volunteers. Antimicrob Agents Chemother 2010;54:411–7.

[10] Harnett SJ, Fraise AP, Andrews JM, Jevons G, Brenwald NP, Wise R. Comparative study of the in vitro activity of a new fluoroquinolone, ABT-492. J Antimicrob Chemother 2004;53:783–92.

[11] Rib-X Company Website, http://www.rib-x.com.

[12] Levasseur P, Delachaume C, Lowther J, Hodgson J. Minimum inhibitory concentrations (MIC) and mutation prevention concentrations (MPC) of NXL101, a novel topoisomerase IV inhibitor, against *Staphylococcus aureus* including multi-resistant strains. In: Proceedings of the 45th interscience conference on antimicrobial agents and chemotherapy; 2005 [abstract F-507].

[13] Lowther J, Levasseur P, Girard A, Delachaume C, Borgonovi M, Hodgson J. Efficacy of NXL101, a novel topoisomerase inhibitor, against multi-resistant *Staphylococcus aureus* in murine septicaemia and thigh muscle infections. In: Proceedings of the 46th interscience conference on antimicrobial agents and chemotherapy; 2006 [abstract F1-1999].

[14] Black MT, Stachyra T, Platel D, Girard AM, Claudon M, Bruneau JM, et al. Mechanism of action of the antibiotic NXL101, a novel nonfluoroquinolone inhibitor of bacterial type II topoisomerases. Antimicrob Agents Chemother 2008;52:3339–49.

[15] Ryder NS. Discontinued drugs in 2008: anti-infectives. Expert Opin Investig Drugs 2010;19:1–21.

[16] Chu DT, Fernandes PB, Claiborne AK, Shen L, Pernet AG. Structure-activity relationships in quinolone antibacterials: design, synthesis, and biological activities of novel isothiazoloquinolones. Drugs Exp Clin Res 1988;14:379–83.

[17] Pucci MJ, Podos SD, Thanassi JA, Leggio MJ, Bradbury BJ, Deshpande M. In vitro and In VIVO PROfile of ACH-702, an isothiazoloquinolone, against bacterial pathogens. Antimicrob Agents Chemother 2011;55:2860–71.

[18] Huband MD, Cohen MA, Zurack M, Hanna DL, Skerlos LA, Sulavik MC, et al. In vitro and in vivo activities of PD 0305970 and PD 0326448, new bacterial gyrase/topoisomerase inhibitors with potent antibacterial activities versus multidrug-resistant Gram-positive and fastidious organism groups. Antimicrob Agents Chemother 2007;51:1191–201.

[19] Tran TP, Ellsworth EL, Sanchez JP, Watson BM, Stier MA, Showalter HD, et al. Structure-activity relationships of 3-aminoquinazolinediones, a new class of bacterial type-2 topoisomerase (DNA gyrase and topo IV) inhibitors. Bioorg Med Chem Lett 2007;17:1312–20.

[20] Pan XS, Gould KA, Fisher LM. Probing the differential interactions of quinazolinedione PD 0305970 and quinolones with gyrase and topoisomerase IV. Antimicrob Agents Chemother 2009;53:3822–31.

[21] Ellsworth EL, Tran TP, Showalter HD, Sanchez JP, Watson BM, Stier MA, et al. 3-Aminoquinazolinediones as a new class of antibacterial agents demonstrating excellent antibacterial activity against wild-type and multidrug resistant organisms. J Med Chem 2006;49:6435–8.

[22] Mani N, Gross CH, Parsons JD, Hanzelka B, Müh U, Mullin S, et al. In vitro characterization of the antibacterial spectrum of novel bacterial type II topoisomerase inhibitors of the aminobenzimidazole class. Antimicrob Agents Chemother 2006;50:1228–37.

[23] Grossman TH, Bartels DJ, Mullin S, Gross CH, Parsons JD, Liao Y, et al. Dual targeting of GyrB and ParE by a novel aminobenzimidazole class of antibacterial compounds. Antimicrob Agents Chemother 2007;51:657–66.

[24] Heath RJ, Rock CO. Fatty acid biosynthesis as a target for novel antibacterials. Curr Opin Investig Drugs 2004;5:146–53.

[25] Zhang YM, White SW, Rock CO. Inhibiting bacterial fatty acid synthesis. J Biol Chem 2006;281:17541–4.

[26] Wang J, Soisson SM, Young K, Shoop W, Kodali S, et al. Platensimycin is a selective FabF inhibitor with potent antibiotic properties. Nature 2006;441:358–61.

[27] Wang J, Kodali S, Lee SH, Galgoci A, Painter R, Dorso K, et al. Discovery of platencin, a dual FabF and FabH inhibitor with in vivo antibiotic properties. Proc Natl Acad Sci USA 2007;104:7612–6.

[28] Brinster S, Lamberet G, Staels B, Trieu-Cuot P, Gruss A, Poyart C. Type II fatty acid synthesis is not a suitable antibiotic target for Gram-positive pathogens. Nature 2009;458:83–6.

[29] Payne DJ, Miller WH, Berry V, Brosky J, Burgess WJ, Chen E, et al. Discovery of a novel and potent class of FabI-directed antibacterial agents. Antimicrob Agents Chemother 2002;46:3118–24.

[30] Karlowsky JA, Laing NM, Baudry T, Kaplan N, Vaughan D, Hoban DJ, et al. In vitro activity of API-1252, a novel FabI inhibitor, against clinical isolates of *Staphylococcus aureus* and *Staphylococcus epidermidis*. Antimicrob Agents Chemother 2007;51:1580–1.

[31] Karlowsky JA, Kaplan N, Hafkin B, Hoban DJ, Zhanel GG. AFN-1252, a FabI inhibitor, demonstrates a Staphylococcus-specific spectrum of activity. Antimicrob Agents Chemother 2009;53:3544–8.

[32] Weiss WJ, Hafkin B, Kaplan N, Pulse ME, Nguyen P, Bolleddula J. Efficacy of AFN-1252 and vancomycin in the mouse subcutaneous abscess with a methicillin-resistant *S. aureus*. In: Proceedings of the 47th interscience conference on antimicrobial agents and chemotherapy; 2007 [abstract F1-329].

[33] Banevicius MA, Deryke CA, Kaplan N, Vaughan D, Nicolau DP. In vivo pharmacodynamic profiling of API-1252 against *Staphylococcus aureus* in a murine thigh model. In: Proceedings of the 46th interscience conference on antimicrobial agents and chemotherapy; 2006 [abstract F1-0759].

[34] Guay DR. Drug forecast—the peptide deformylase inhibitors as antibacterial agents. Ther Clin Risk Manag 2007;3:513–25.

[35] Leeds JA, Dean CR. Peptide deformylase as an antibacterial target: a critical assessment. Curr Opin Pharmacol 2006;6:445–52.

[36] Mazel D, Pochet S, Marlière P. Genetic characterization of polypeptide deformylase, a distinctive enzyme of eubacterial translation. EMBO J 1994;13:914–23.

[37] Lee MD, She Y, Soskis MJ, Borella CP, Gardner JR, Hayes PA, et al. Human mitochondrial peptide deformylase, a new anticancer target of actinonin-based antibiotics. J Clin Invest 2004;114:1107–16.

[38] Margolis PS, Hackbarth CJ, Young DC, Wang W, Chen D, Yuan Z, et al. Peptide deformylase in *Staphylococcus aureus*: resistance to inhibition is mediated by mutations in the formyltransferase gene. Antimicrob Agents Chemother 2000;44:1825–31.

[39] Ramanathan-Girish S, McColm J, Clements JM, Taupin P, Barrowcliffe S, Hevizi J, et al. Pharmacokinetics in animals and humans of a first-in-class peptide deformylase inhibitor. Antimicrob Agents Chemother 2004;48:4835–42.

[40] Osborne CS, Neckermann G, Fischer E, Pecanka R, Yu D, Manni K, et al. In vivo characterization of the peptide deformylase inhibitor LBM415 in murine infection models. Antimicrob Agents Chemother 2009;53:3777–81.

[41] Silver LL. Challenges of antibacterial discovery. Clin Microbiol Rev 2011;24:71–109.

[42] Bouchillon S, Hackel M, Hoban D, Zalacain M, Butler D. In vitro activity of GSK1322322, a novel peptide deformylase Inhibitor, against 4,836 pathogens from skin and soft tissue infections and respiratory tract infections. In: Proceedings of the 50th interscience conference on antimicrobial agents and chemotherapy; 2010 [abstract F1-2112].

[43] US National Institutes of Health Clinical Trials Website, http://clinicaltrials.gov/ct2/home.

[44] Houghton JL, Green KD, Chen W, Garneau-Tsodikova S. The future of aminoglycosides: the end or renaissance? Chembiochem 2010;11:880–902.

[45] Armstrong ES, Miller GH. Combating evolution with intelligent design: the neoglycoside ACHN-490. Curr Opin Microbiol 2010;13:565–73.

[46] Aggen JB, Armstrong ES, Goldblum AA, Dozzo P, Linsell MS, Gliedt MJ, et al. Synthesis and spectrum of the neoglycoside ACHN-490. Antimicrob Agents Chemother 2010;54:4636–42.

[47] Livermore DM, Mushtaq S, Warner M, Zhang JC, Maharjan S, Doumith M, et al. Activity of aminoglycosides, including ACHN-490, against carbapenem-resistant Enterobacteriaceae isolates. J Antimicrob Chemother 2011;66:48–53.

[48] Pulse M, Weiss WJ, Reyes N, Kostrub CF, Nguyen P, Renick P. Efficacy of ACHN-490 in a murine urinary tract infection model with *Escherichia coli*. In: Proceedings of the 48th annual meeting of the Infectious Diseases Society of America, Vancouver, British Columbia, Canada; 2010 [abstract 3254].

[49] Achaogen Company Website, http://www.achaogen.com.
[50] SelectX Pharmaceuticals Company Website, http://www.selectxpharm.com.
[51] Schmitz FJ, Schwarz S, Milatovic D, Verhoef J, Fluit AC. In vitro activities of the ketolides ABT-773 and telithromycin and of three macrolides against genetically characterized isolates of *Streptococcus pneumoniae*, *Streptococcus pyogenes*, *Haemophilus influenzae* and *Moraxella catarrhalis*. J Antimicrob Chemother 2002;50:145–8.
[52] Rafie S, MacDougall C, James CL. Cethromycin: a promising new ketolide antibiotic for respiratory infections. Pharmacotherapy 2010;30:290–303.
[53] Patel SN, Pillai DR, Pong-Porter S, McGeer A, Green K, Low DE. In vitro activity of ceftaroline, ceftobiprole and cethromycin against clinical isolates of *Streptococcus pneumoniae* collected from across Canada between 2003 and 2008. J Antimicrob Chemother 2009;64:659–60.
[54] CDER Company Website. Summary Minutes of the Anti-Infective Drugs Advisory Committee; June 8, 2009, Freedom of Information Office, FDA, http://www.fda.gov/downloads/AdvisoryCommittees/CommitteesMeetingMaterials/Drugs/Anti-InfectiveDrugsAdvisoryCommittee/UCM183844.pdf.
[55] Farrell DJ, Castanheira M, Sader HS, Jones RN. The in vitro evaluation of solithromycin (CEM-101) against pathogens isolated in the United States and Europe (2009). J Infect 2010;61:476–83.
[56] McGhee P, Clark C, Kosowska-Shick KM, Nagai K, Dewasse B, Beachel L, et al. In vitro activity of CEM-101 against *Streptococcus pneumoniae* and *Streptococcus pyogenes* with defined macrolide resistance mechanisms. Antimicrob Agents Chemother 2010;54:230–8.
[57] Cempra Company Website, http://www.cempra.com/research/antibacterials/.
[58] Locke JB, Morales G, Hilgers MG, Kedar GC, Rahawi S, Picazo JJ, et al. Elevated linezolid resistance in clinical cfr-positive *Staphylococcus aureus* isolates is associated with co-occurring mutations in ribosomal protein L3. Antimicrob Agents Chemother 2010;54:5352–5.
[59] Brown SD, Traczewski MM. Comparative in vitro antimicrobial activities of torezolid (TR-700), the active moiety of a new oxazolidinone, torezolid phosphate (TR-701), determination of tentative disk diffusion interpretive criteria, and quality control ranges. Antimicrob Agents Chemother 2010;54:2063–9.
[60] Trius Company Website, http://triusrx.com/.
[61] Prokocimer P, Bien P, Surber J, Mehra P, DeAnda C, Bulitta JB, et al. A phase 2 randomized, double-blind, dose-ranging study evaluating the safety, tolerability, population pharmacokinetics, and efficacy of oral torezolid phosphate in patients with complicated skin and skin structure infections (cSSSI). Antimicrob Agents Chemother 2011;55:583–92.
[62] Rib-X Company Website, http://www.rib-x.com/.
[63] Lawrence L, Danese P, DeVito J, Franceschi F, Sutcliffe J. In vitro activities of the Rx-01 oxazolidinones against hospital and community pathogens. Antimicrob Agents Chemother 2008;52:1653–62.
[64] Boucher HW, Talbot GH, Bradley JS, Edwards JE, Gilbert D, Rice LB, et al. Bad bugs, no drugs: no ESKAPE! An update from the Infectious Diseases Society of America. Clin Infect Dis 2009;48(January (1)):1–12.
[65] Macone A, Donatelli J, Dumont T, Levy SB, Tanaka SK. In vitro activity of PTK0796 (BAY 73-6944) against Gram-positive and Gram-negative organisms. In: Abstract 2439 43rd interscience conference on antimicrobial agents and chemotherapy; 2003.
[66] Traczewski MM, Brown SD. PTK 0796 (BAY 73-6944): in vitro potency and spectrum of activity compared to ten other antimicrobial compounds. In: Abstr. 2458 43rd interscience conference on antimicrobial agents and chemotherapy; 2003.
[67] Torok ME, Chapman AL, Lessing MP, Sanderson F, Seaton RA. Outpatient parenteral antimicrobial therapy: recent developments and future prospects. Cur Opin Investig Drugs 2010;11:929–39.
[68] Walker K. Interscience conference on antimicrobial agents and chemotherapy—50th annual meeting—research on promising new agents: part 1. IDrugs 2010;13:743–5.
[69] Tetraphase Company Website, www.tphase.com.
[70] Hunt D, Xiao X, Clark R, O'Brien W, Fyfe C, Grossman T, et al. TP-434 is a novel broad-spectrum fluorocycline. Poster F1-2157. In: 50th interscience conference on antimicrobial agents and chemotherapy; 2010.
[71] Murphy T, Slee A, Sutcliffe J. TP-434 is highly efficacious in animal models of infection, poster F1-2161. In: 50th interscience conference on antimicrobial agents and chemotherapy; 2010.
[72] Page MGP, Heim J. New molecules from old classes: revisiting the development of beta-lactams. IDrugs 2009;12(September):561–5.
[73] Jacoby GA. Bush K company website, β-Lactamase classification and amino acid sequences for TEM, SHV and OXA extended-spectrum and inhibitor resistant enzymes; 2011 April 12, 2011, http://www.lahey.org/Studies/.
[74] Page MGP, Dantier C, Desarbre E. In vitro properties of BAL30072, a novel siderophore sulfactam with activity against multiresistant Gram-negative bacilli. Antimicrob Agents Chemother 2010;54(June):2291–302.
[75] Livermore D, Mushtaq S, Warner M. Activity of BAL30376 (monobactam BAL19764 + BAL29880 + clavulanate) versus Gram-negative bacteria with characterized resistance mechanisms. J Antimicrob Chemother 2010 Nov;65:2382–95.
[76] Huband MD, Gootz TD, Flanagan ME, Quinn JP, Mullins LM, Penzien JB, et al. In vitro antibacterial activity of MC-1: a new siderophore-monocarbam conjugate versus recent Gram-negative bacterial clinical isolates. In: Abstr. F1-2129 50th interscience conference on antimicrobial agents and chemotherapy; 2010.
[77] Stachyra T, Levasseur P, Pechereau MC, Girard AM, Claudon M, Miossec C, et al. In vitro activity of the β-lactamase inhibitor NXL104 against KPC-2 carbapenemase and *Enterobacteriaceae* expressing KPC carbapenemases. J Antimicrob Chemother 2009;64:326–9.
[78] Mushtaq S, Warner M, Livermore DM. In vitro activity of ceftazidime + NXL104 against *Pseudomonas aeruginosa* and other non-fermenters. J Antimicrob Chemother 2010;65:2376–81.
[79] Mushtaq S, Warner M, Williams G, Critchley I, Livermore DM. Activity of chequerboard combinations of ceftaroline and NXL104 versus beta-lactamase-producing Enterobacteriaceae. J Antimicrob Chemother 2010;65:1428–32.
[80] Takeda S, Nakai T, Wakai Y, Ikeda F, Hatano K. In vitro and in vivo activities of a new cephalosporin, FR264205, against *Pseudomonas aeruginosa*. Antimicrob Agents Chemother 2007;51:826–30.
[81] Livermore DM, Mushtaq S, Ge Y. Chequerboard titration of cephalosporin CXA-101 (FR264205) and tazobactam versus beta-lactamase-producing Enterobacteriaceae. J Antimicrob Chemother 2010;65:1972–4.
[82] Young K, Raghoobar SL, Hairston NN, Painter RE, Racine F, Dorson KL, et al. In vitro activity of the class A and C (-lactamase inhibitor MK-7655. In: Abstr. F1-2139 50th interscience conference on antimicrobial agents and chemotherapy; 2010.
[83] Bush K, Macielag MJ. New β-lactam antibiotics and β-lactamase inhibitors. Exp Opin Therapeut Pat 2010;20:1277–93.
[84] Pages JM, Sandrine AF, Mahamoud A, Bolla JM, Davin-Regli A, Chevalier J, et al. Efflux pumps of Gram-negative bacteria, a new target for new molecules. Curr Topics Med Chem 2010;10:1848–57.
[85] Lomovskaya O, Zgurskaya HI, Totrov M, Watkins WJ. Waltzing transporters and 'the dance macabre' between humans and bacteria. Nat Rev Drug Discov 2007;6:56–65.
[86] Findlay B, Zhanel GG, Schweizer F. Cationic amphiphiles, a new generation of antimicrobials inspired by the natural antimicrobial peptide scaffold. Antimicrob Agents Chemother 2010;54:4049–58.
[87] Wang G, Li X, Wang Z. APD2: the updated antimicrobial peptide database and its application in peptide design. Nucleic Acids Res 2009;37:D933–7.
[88] http://anacor.com/gsk052.php.
[89] Silver LL. Multi-targeting by monotherapeutic antibacterials. Nat Rev Drug Discov 2007;6:41–55.