

Boron-Containing Compounds as Antimicrobial Agents to Tackle Drug-Resistant Bacteria

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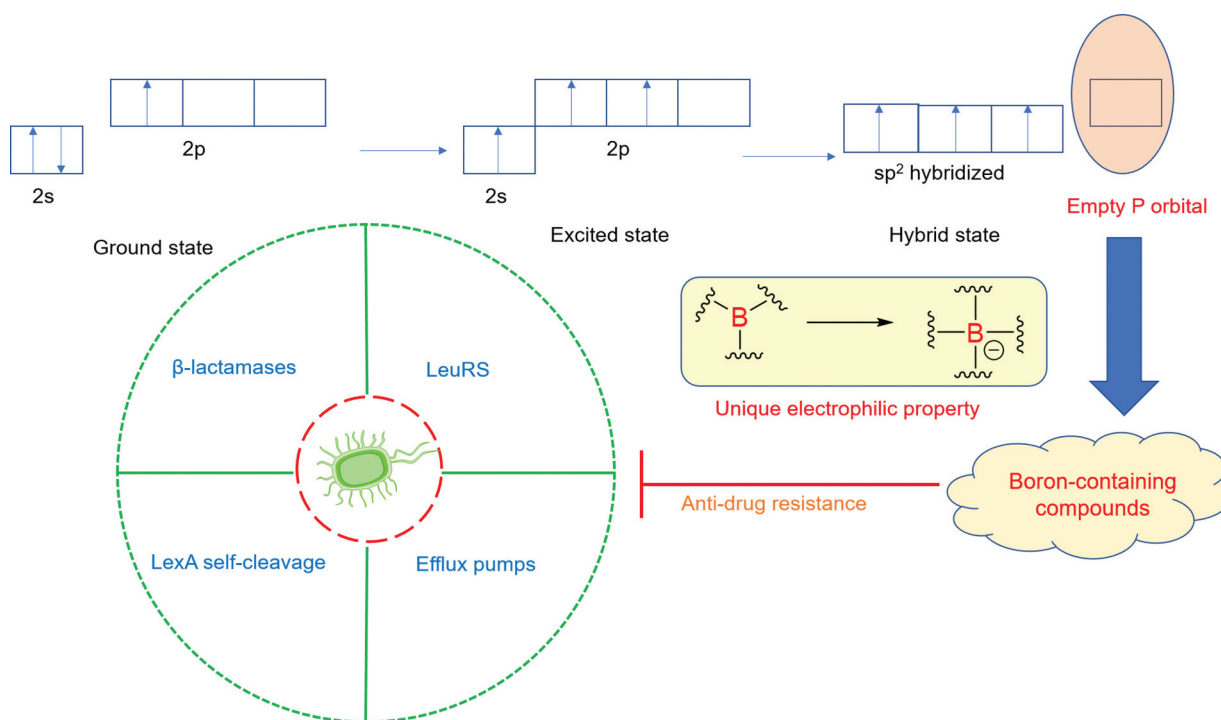
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Abstract

Keywords

- ▶ boron-containing compounds
- ▶ bacterial infections
- ▶ drug-resistant
- ▶ antibacterial agents
- ▶ adjuvants

Bacterial infections, especially those caused by drug-resistant bacterial pathogens, are crucial diseases that damage human health. In recent decades, several important boron-containing drugs have been marketed as anticancer agents or anti-infective adjuvants. Among them, vaborbactam revitalizes the antibacterial effects of meropenem against bacteria by inhibiting β -lactamases, opening a new field for addressing bacterial resistance. In this article, the chemical features of boron atoms and the typical antibacterial agents and adjuvants of boron-containing compounds are reviewed. In this work, boron-containing agents are classified into four categories according to their action mechanisms: β -lactamase inhibitors, leucyl-tRNA synthetase inhibitors, LexA self-cleavage inhibitors, and NorA efflux pump inhibitors. This review provides actionable insights for addressing the increasingly severe drug-resistant infections of bacterial pathogens.

Introduction

Bacterial pathogens affect our daily lives and pose a serious threat to public health. Once people are infected with pathogenic bacteria, they will suffer from corresponding diseases or even die.¹ This phenomenon did not change until Alexander Fleming first discovered the antibiotic drug, penicillin, in 1920s.² However, with the increase in clinical use of antibiotics, bacterial resistance has emerged and is considered one of the most intractable public health problems worldwide today.³ Despite many efforts to address the problem, only a limited number of new antibiotics have been launched in the past two decades.^{4,5} Most of the newly developed antibiotics are “like” or “better than like” drugs, to which bacteria quickly develop resistance.^{6,7} Therefore, there is an urgent need for medicinal chemists to discover antibacterial agents or adjuvants with new mechanisms of action.

Boron, with an empty p orbital, is an element adjacent to carbon in the periodic table, which has several unique and valuable properties to be used in medicinal chemistry.⁸ Boron-containing compounds generally have better bioavailability, pharmacokinetic properties, and target selectivity.⁹ While boron is not a panacea for all diseases, it expands the arsenals of disease treatment. Boron-containing compounds are currently used to treat diseases including cancer,^{10–12} inflammation,^{10,13} and viral^{14,15} and bacterial infections^{16,17} (▶ Fig. 1).

As shown in ▶ Fig. 1, five boron-containing drugs have been approved for clinical use over the past two decades including bortezomib,¹⁸ ixazomib,¹⁹ tavaborole (AN2690),²⁰ crisaborole (AN2728),²¹ and vaborbactam. In addition, several boron-containing compounds have entered clinical assays, including AN2898 for the treatment of psoriasis, and acoziborole (SCYX-7158) for the treatment of human African trypanosomiasis (sleeping sickness).

In this article, we first illustrate the structural characteristics of boron and its potential as an antimicrobial and adjuvant to combat bacterial resistance and then give a broad overview of the research progress of boron-containing

compounds as agents to address bacterial resistance, as well as various biological targets, namely β -lactamases, leucyl-tRNA synthetase (LeuRS), LexA self-cleavage, and NorA efflux pumps. We will comprehensively discuss how these boron-containing compounds function through the four targets and provide some ideas for future use of the compounds in antimicrobial research.

Structural Characteristics of Boron

Boron appears in nature in the form of boron-containing complexes, not as a single atom.²² In natural products, it is not common for boron to occur as a C–B bond, but rather as a B–O bond. The bond length of B–O (1.40 Å) is shorter than that of C–O (1.43 Å), suggesting a high energy of the B–O bond. Thus, boron-containing compounds containing B–O bonds are generally more stable than their carbon-based counterparts. This structural feature suggests that microorganisms might lack metabolic enzymes to break down boron-containing compounds.^{23,24}

Boron belongs a $2s^2 2p^1$ valence electron structure, with three valence electrons and four valence orbitals. Three sp^2 hybrid bonds are formed in a plane, orthogonal to the unbound empty p orbital. The presence of the empty p orbital mediates the physicochemical properties and reaction modes of neutral sp^2 boron compounds (▶ Fig. 2A). As a consequence, boronic acids, with $pK_a = 7–9$ for different substituted phenylboronic acids (▶ Fig. 2B), show a trigonal planar geometry and allow for conversion to sp^3 -hybridized boron by coordination of a hydroxyl group or other nucleophiles, forming a carbon-like conformation of tetrahedral intermediates (▶ Fig. 2C). This is the general basis for the mechanism of boron-containing compounds used as protease inhibitors (▶ Fig. 2D).

Taken together, the low propensity of the B–O bond to be hydrolyzed by bacteria, coupled with the conversion ability of the boron atoms, provides a foundation for the use of boron-containing antibiotics or adjuvants for addressing bacterial resistance.

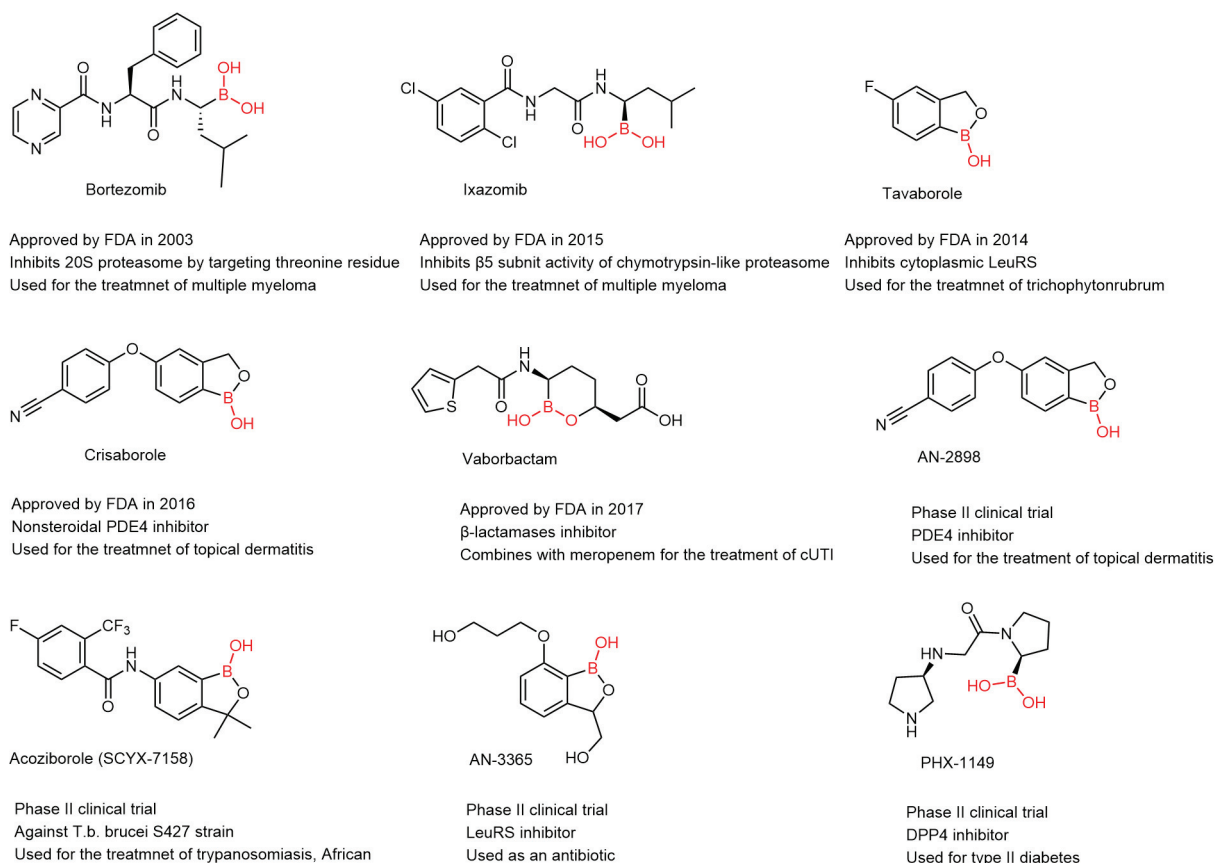


Fig. 1 Boron-containing drugs already marketed and in clinical trials. cUTI, complicated urinary tract infection; DPP4, dipeptidyl peptidase 4; LeuRS, leucyl-tRNA synthetase; PDE4, phosphodiesterase 4.

β -Lactamase Inhibitors

Antibacterial Mechanism of β -Lactam Antibiotics

Penicillins, cephalosporins, monobactams, and carbapenems are the four classes of classical β -lactam antibiotics (\rightarrow Fig. 3A), which are most often used medications in the clinical treatment of bacterial infections and account for a considerable share of clinical usage. Alexander Fleming's discovery of Penicillin G, the first β -lactam antibiotic, was a watershed moment in antimicrobial therapy, and since then, several antibiotics have been discovered, many of which are based on the β -lactam scaffold of penicillin.

This type of antibiotic acts on the bacterial cell wall. The bacterial cell wall mainly consists of peptidoglycan, which is formed by a β -1,4-glycosidic bond between two disaccharide units consisting of N-acetylmuramic acid and N-acetylglucosamine. The cross-linked bonds of the peptides vary depending on the types of bacteria.²⁵ It covers the cytoplasmic membrane of the cell to maintain normal bacterial morphology and prevents the invasion of macromolecules. Blumberg and Strominger discovered a high degree of structural similarity between penicillin and the ending of D-Ala-D-Ala peptidoglycan in either gram-positive or gram-negative bacteria (\rightarrow Fig. 3B).²⁶

The similarity of penicillin to D-Ala-D-Ala peptidoglycan is the functional basis of β -lactam antibiotics. Penicillin exerts its bactericidal effects by inhibiting cell wall synthesis

through inhibition of the binding of tetrapeptide side chains and pentapeptide cross-linking bridge. It can compete with the latter for transpeptidases, also called penicillin-binding proteins (PBPs), interfering with the formation of mucopeptides, leading to cell wall defects and ultimately the loss of the bacterial cell wall.²⁷ The destruction of the bacterial cell wall is a mortal attack to eliminate the bacterial stability and lead to its death.

β -Lactamases and Canonical Inhibitors

β -Lactamases have been co-evolving with antibiotics since the first generation of antibiotics was employed in the clinic in the 1940s. To date, approximately four mechanisms of bacterial resistance to β -lactam antibiotics have been identified. The first and foremost is the generation of broad substrate-specific β -lactamases. When β -lactamases break the amide bond of the β -lactam ring, the antibiotic becomes ineffective. The second resistance mechanism is the structural alterations in key target transpeptidase PBPs, such as PBP2a in methicillin-resistant *Staphylococcus aureus* (MRSA). The third and fourth mechanisms are the increased permeability barriers and upregulated efflux of antibiotics employed by gram-negative strains.²⁸ Of all these mechanisms, β -lactamases are the top priority that needs to be addressed.²⁹

The first β -lactamases produced by *Bacillus Escherichia coli* were identified in 1988 by Abraham and Chain.³⁰

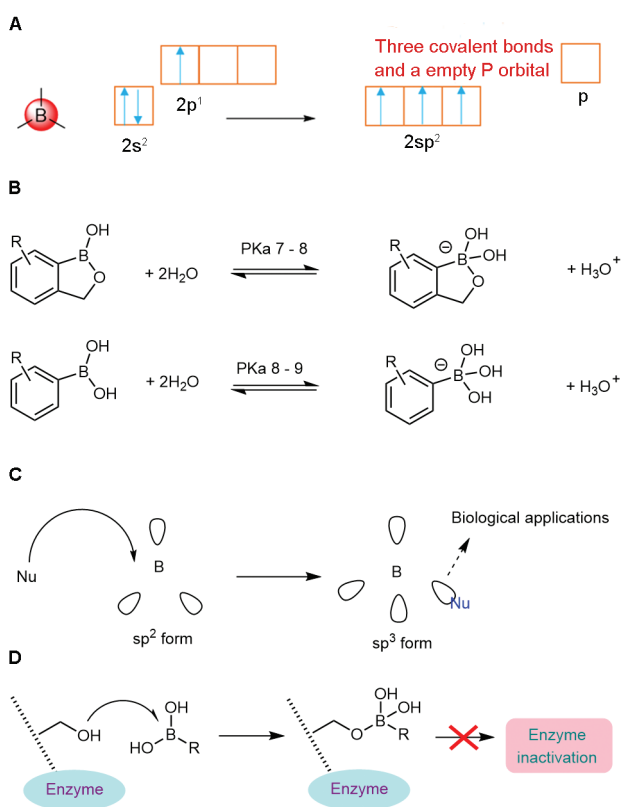


Fig. 2 Chemical characteristics, properties, and inhibitory function of boron atom. (A) Electron orbital description diagram; (B) pKa of benzoxaboroles and phenylboronic acids; (C) configuration modifications of boron; (D) general transition-state mode of boron-containing compounds for protease inhibitors.

Currently, β -lactamases have more than 2,000 unique amino acid sequences, indicating extended portfolio substrate for β -lactamases. The presence of extended-spectrum β -lactamases (ESBLs), penicillinases, AmpC cephalosporinases, and carbapenemases, can lead to antibiotics inactivation, which may be considered a human public safety issue.³¹ Because of their broad spectrum and potency, carbapenem antibiotics are frequently used as a “last resort” for the treatment of serious infections caused by gram-negative bacteria, and inactivation of carbapenem antibiotics will cause a crisis when facing severe gram-negative bacterial infections in the clinic.

β -Lactamases are classified into four categories: A, B, C, and D, based on amino acid sequence homology (Ambler classification).³² All serine- β -lactamases (SBLs) could be categorized into three classes, TEM-, SHV-, CTX-M-, and KPC-type variants (class A), AmpC-type and plasmid-encoded CMY-type cephalosporinases (class C), and OXA-type enzymes (oxacillinases, class D).³³ Class B metallo- β -lactamases (MBLs; including VIM-1, GOB-1, IMP-1, etc.), with extended-broad substrate profiles and potent carbapenemase activity, are zinc-dependent MBLs. Based on the major amino acid sequences, MBLs can be classified into three subclasses, B1, B2, and B3.^{24,34} In addition, another classification approach, called Bush–Jacoby classification, can divide β -lactamases into four classes, based on their

biomedical function, mainly based on substrate specificity.³⁵ This classification method will not be described here in detail.

Currently, ESBLs and carbapenemases can hydrolyze a wide range of β -lactam drugs, including carbapenems and cephalosporins. All SBLs utilize a serine residue as a catalytic site to hydrolyze β -lactam antibiotics in a multi-step process. First, positively charged residues along the chain of the azetidinone ring of the β -lactam antibiotic attract a negatively charged carboxylate or similar group to the active site of β -lactamases, and when the two groups are in the proper position, β -lactamases form a hydrogen bond with the β -lactams.³⁶ Next, β -lactamases act as reactive nucleophiles to complete acylation reactions via serine residues. Finally, activated water molecules deacylate the β -lactam- β -lactamase complex, which opens the antibiotic ring and regenerates β -lactamases. MBLs need at least one (B2 subclass) and commonly two Zn^{2+} ions (B1 and B3 subclasses), which are bridged by a hydroxide ion, and facilitate the attack of the nucleophilic reagent on the carbonyl carbon atom of β -lactam ring, resulting in hydrolysis and inactivation of the antibiotic (**Fig. 4**).³⁷

Combining β -lactam antibiotics with an appropriate β -lactamase inhibitor is currently one of the most successful methods to address β -lactamase-mediated drug resistance. The first-generation β -lactamases inhibitors (BLIs) were created using the penicillin drug scaffold (**Fig. 5A**), which primarily targets class A β -lactamases, yet, has limited inhibitory activity effects against the other three β -lactamases.³⁸

Second-generation BLIs with diazabicyclooctanone (DBO) scaffolds (**Fig. 5B**) displayed improved antimicrobial profiles and also inhibited class C and D BLs.³⁹ Nevertheless, DBOs BLIs did not inhibit class B β -lactamases, and their synthetic routes are challenging, often with over 12 steps of chemical transformation.⁴⁰

Boron-Containing β -Lactamase Inhibitors

Boronic acid-based β -lactamase inhibitors were first reported in 1978,⁴¹ and were previously designed by Waley and his coworkers to fight against class C β -lactamases.⁴² Electrophilic boron atom forms a tetrahedral adduct with the catalytic serine residue that mimics the carbonyl structure of the β -lactam ring, much like the transitional forms of the hydrolysis mechanism of β -lactam antibiotics.⁴³ Boronic acid transition-state inhibitors (BATSIs) are a common name for these compounds.

Benzoxaboroles and aryl boronic acids inhibit the β -lactamases of SBLs or MBLs by forming a sp^3 -hybridized tetrahedral complex, in which boron atom binds to nucleophilic serine residues of SBLs or hydroxide ions of MBLs in the hydrolysis of β -lactam antibiotics to mimic the adduct of SBLs or MBLs.^{44–46}

Most boron-containing inhibitors inhibit SBLs only by binding to serine residues through a covalent bond (**Fig. 6A**), whereas dual-inhibitory boron-containing inhibitors cooperate with the zinc atom of MBLs using an oxygen atom adjacent to the boron, and the carbonyl oxygen on the

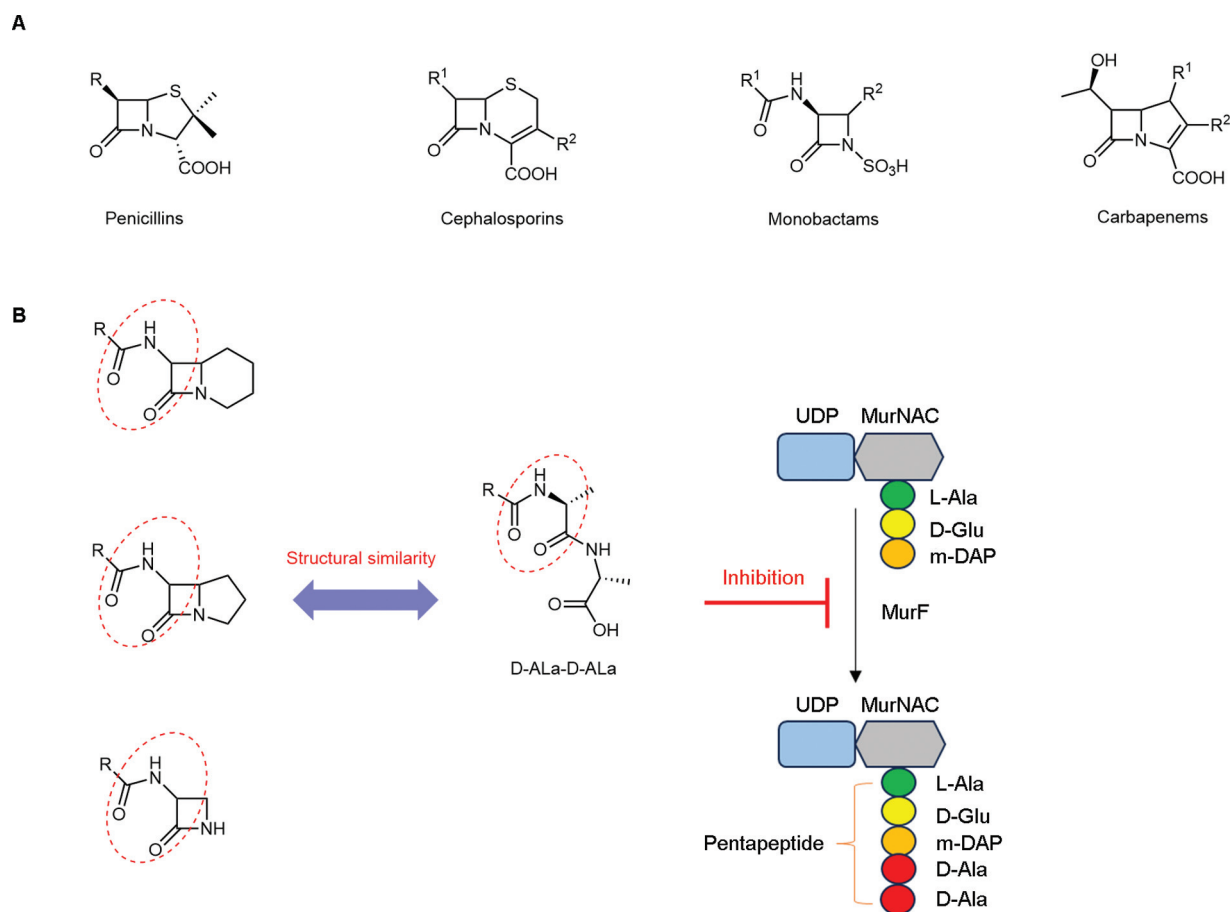


Fig. 3 Chemical structures of common β -lactam drugs. (A) Major classes of β -lactam antibiotics. (B) Structural similarity between azetidinone with different ring numbers and D-Ala-D-Ala.

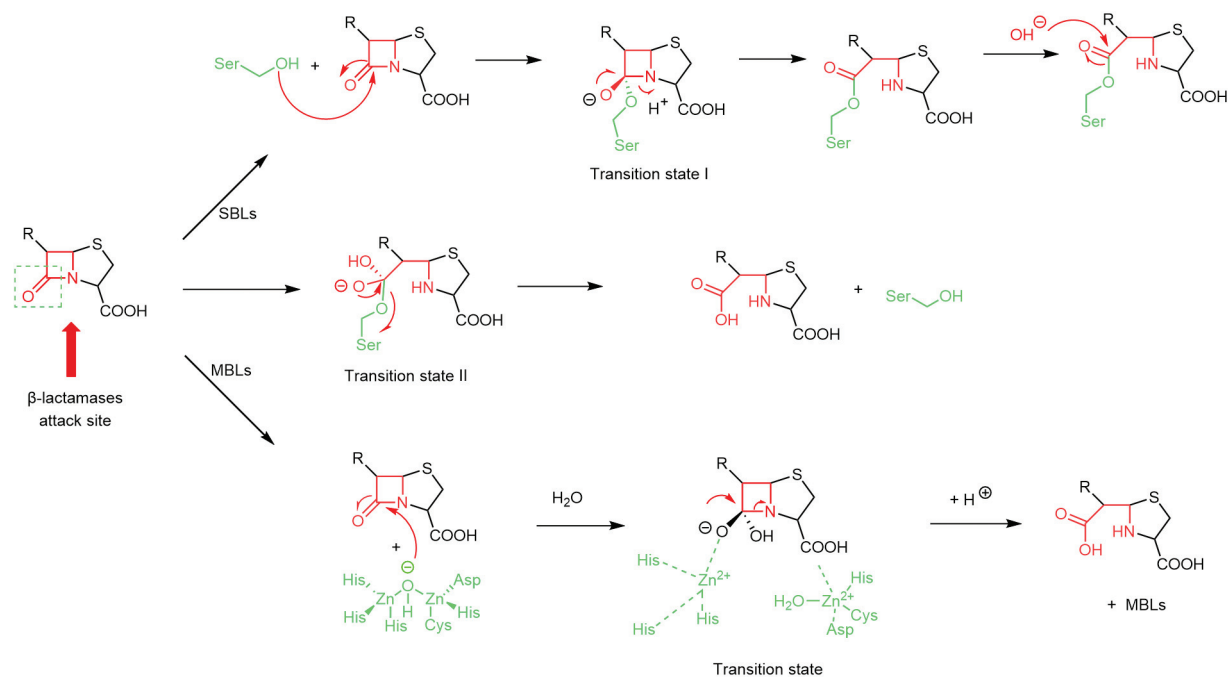


Fig. 4 General process of SBLs and MBLs hydrolyzing β -lactams. SBLs, serine- β -lactamases; MBLs, metallo- β -lactamases.

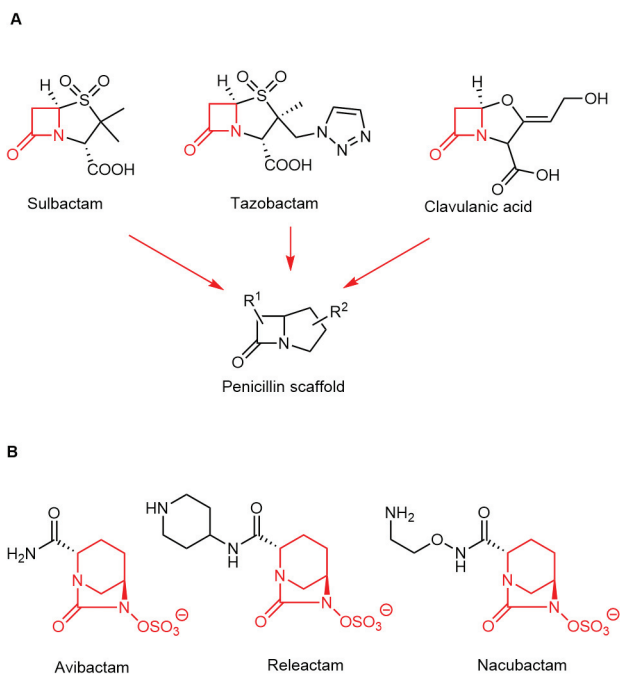


Fig. 5 Structural characteristics of representative (A) first- and (B) second-generation β -lactamase inhibitors using a penicillin scaffold and diazabicyclooctanones (DBOs) scaffolds, respectively.

carboxyl group on the benzene ring (**Fig. 6B**, top). There is also a structural type of boron-containing compound that relies on the thiol group to coordinately interact with two zinc atoms of MBLs and use the boron atom to covalently bind to the serine residue of SBLs to inhibit SBLs (**Fig. 6B**, bottom).

Narrow-Spectrum Boron-Containing β -Lactamase Inhibitors

These compounds act mainly on class A, C, and D SBLs and have little effect on MBLs, and are defined herein as narrow-spectrum BLs. Five of them, vaborbactam, S02030, SM23, CR192, and BA4, will be briefly illustrated in this section (**Fig. 7A**).

Hecker et al.⁴⁷ designed and synthesized vaborbactam, previously known as RPX7009, the first boron-containing compound marketed as a β -lactamase inhibitor for use as an adjuvant to meropenem. Meropenem is a broad-spectrum carbapenem antibiotic that eliminates bacteria by inhibiting the biosynthesis of bacterial cell walls. Vaborbactam is potent against class A (e.g., KPC, TEM, SHV) and class C (P99, MIR, FOX) β -lactamases, but has a mild effect on class B and D β -lactamases.³⁸ The most remarkable feature of vaborbactam is that it can strongly inhibit KPC enzymes and has the potential to combat various carbapenem-resistant strains.^{48,49} Furthermore, vaborbactam shows good inhibition to KPC-2 ($K_i = 69$ nmol/L), CTX-M-15 ($K_i = 44$ nmol/L), SHV-12 ($K_i = 29$ nmol/L), TEM-10 ($K_i = 110$ nmol/L), P99 ($K_i = 53$ nmol/L), and CMY-2 ($K_i = 99$ nmol/L).

Cocrystal analysis of vaborbactam with AmpC or CTX-M-15 shows that the boron atom is covalently bound to serine residue, forming a tetrahedral adduct and several hydrogen bonds with amino acid residues such as Gln 140, Tyr170, and Ser338 in AmpC (**Fig. 7B**, left), or Asn107, Ser105, and Lys209 residues in CTX-M-15 (**Fig. 7B**, right). Remarkably, vaborbactam exhibits minor toxicity in multiple experiments.⁴⁷

Meropenem/vaborbactam is a novel drug combination with potent efficacy against KPC-producing *Enterobacteriales*.^{50,51} The minimum inhibitory concentration (MIC) values of this drug combination against a group of drug-resistant bacteria are shown in **Table 1**.⁵²⁻⁵⁴

S02030 was synthesized by Powers and coworkers to fight against *Acinetobacter*-derived cephalosporinase 7 (ADC-7).⁵⁵ It mimics the structure of cephalothin and binds tightly with ADC-7 ($K_i = 44.5 \pm 2.2$ nmol/L). When S02030 was combined with ceftazidime for the treatment of *E. coli* DH10B bla_{ADC-7} , the MIC values of ceftazidime were reduced from 64 to 8 μ g/mL. Structure-activity relationship (SAR) analysis indicates that the potent S02030 has a carboxylate group, which mimics the C3/C4 carboxyl group in β -lactams and is essential for efficacy. In addition, S02030 strongly interacts with Arg340, a key amino acid residue that distinguishes ADC-7 from other AmpCs. On the other part of this scaffold, the amide

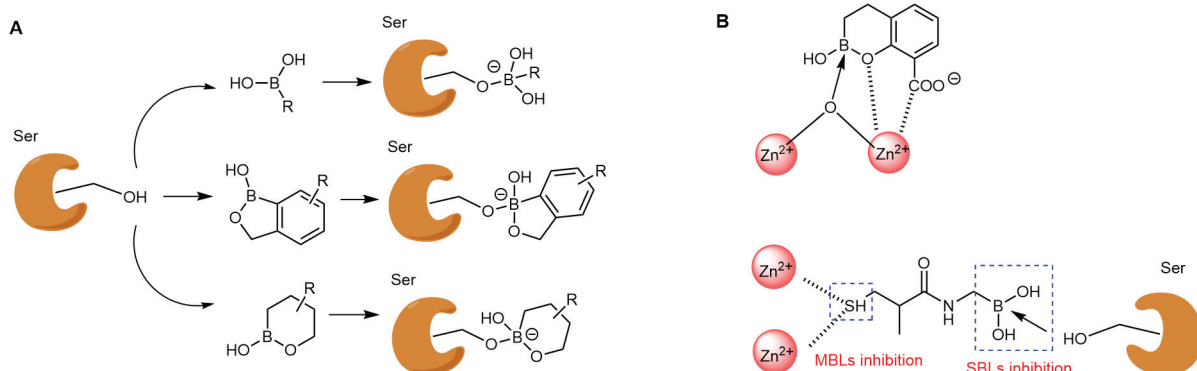


Fig. 6 Mechanisms of BLIs to inhibit SBLs and MBLs. (A) Action mechanism of SBLs inhibitors. (B) Action mechanism of SBL-/MBL-inhibitors to inactivate MBLs (top); mechanism of MBL inhibitors that rely on the sulfur atom to interact with MBLs and rely on boron atom to covalently interact with SBLs, thus achieving the goal of dual-inhibition (bottom). BLIs, β -lactamase inhibitors; SBLs, serine- β -lactamases; MBLs, metallo- β -lactamases.

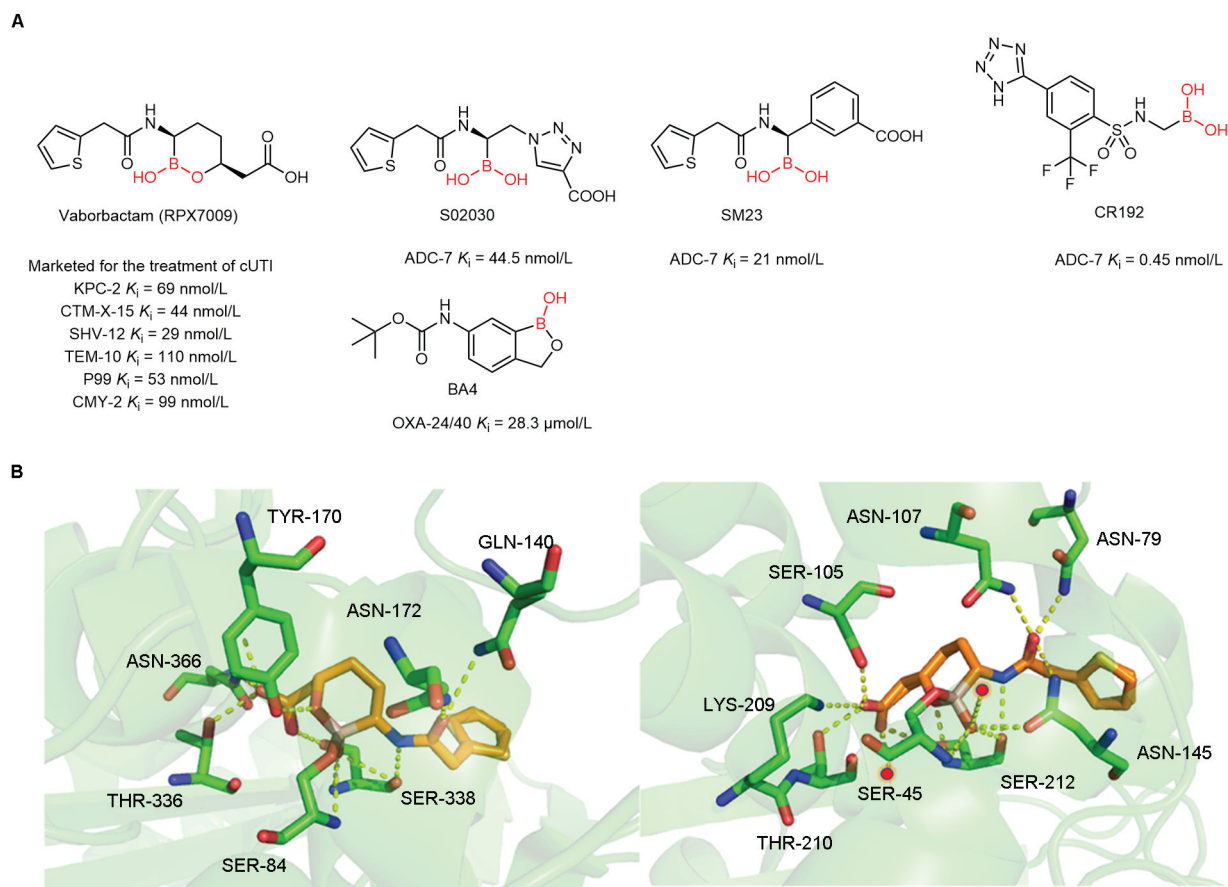


Fig. 7 Chemical structural and action mode diagram of boron-containing SBL inhibitors. (A) Typical boron-containing SBL inhibitors. (B) Co-crystallization analysis of vaborbactam and AmpC (PDB ID 4XUX) (left); co-crystallization analysis of vaborbactam and CTX-M-15 (PDB ID 4XUZ) (right). The red dots represent water molecules. SBLs, serine- β -lactamases.

Table 1 *In vitro* activity of meropenem/vaborbactam against several drug-resistant bacteria

Microorganisms	No. of isolates	MIC range	Meropenem + vaborbactam (8 μ g/mL)		Ref.
			MIC ₅₀	MIC ₉₀	
<i>P. aeruginosa</i>	2,604	≤ 0.015 , > 32	0.5	8	52,53
<i>A. baumannii</i>	84	0.03, > 32	32	64	52,53
<i>Acinetobacter spp.</i>	708	1, > 64	32	> 32	52,53
Enterobacterales (KPC)	991	≤ 0.03 , > 32	0.06	1	52,53
<i>E. coli</i>	6,595	≤ 0.05 , 32	0.015	0.015	52,53
<i>K. pneumoniae</i>	3,247	≤ 0.015 , > 32	0.03	0.12	52,53
<i>S. maltophilia</i>	353	≤ 0.015 , > 32	> 32	> 32	52,53
<i>K. oxytoca</i>	742	≤ 0.015 , 16	0.03	0.03	54
KPC-producing CRE	206	0.015, 32	0.25	1	52,53
Non-KPC-producing CRE	250	0.015, 32	4	> 32	54

Abbreviations: CRE, carbapenem-resistant *Enterobacteriaceae*; KPC, *Klebsiella pneumoniae* carbapenemase; MIC, minimum inhibitory concentration.

group is highly conserved and interacts with Gln120 and Asn152. These interactions between AmpC and BATSIs make S02030 a promising lead compound that can be modified on the carboxylate side to improve its efficacy or chemical properties.⁵⁶

SM23 was synthesized by Morandi et al.⁵⁷ It also effectively inhibits class C β -lactamases, particularly FOX-4 and extended-spectrum cephalosporinases. SM23 had a good binding affinity for AmpC with a K_i value of 21.1 ± 1.9 nmol/L. The MIC values of ceftazidime against *E. coli* DH10B bla_{ADC-7}

were reduced from 64 to 8 µg/mL when combined with SM23.⁵⁵

Bouza et al synthesized CR192,⁵⁸ with the K_i value at the nanomolar level (0.45 nmol/L), indicating that sulfonamide is a successful mimic of the amide moiety. The MIC values of ceftazidime against *E. coli* DH10B bla_{ADC-7} were decreased 32-fold from 64 to 2 µg/mL when combined with CR192.

BA4 was synthesized by Werner et al.⁵⁹ It inhibits class D β-lactamases including OXA-24/40.^{60,61} Unlike the inhibitors mentioned above, BA4 exerts their inhibitory effects mainly on class A and C β-lactamases. SAR analysis showed that its benzoxaborole scaffold is required for its class D β-lactamase inhibitory activity and that the *meta*-substituted derivatives behaved better to accommodate the active site of OXA-24/48.

The five boron-containing inhibitors mentioned above mainly inhibit SBLs of classes A, C, and D but do not inhibit class B β-lactamases. In the next section, broad-spectrum boron-containing BLIs will be discussed.

Broad-Spectrum Boron-Containing β-Lactamase Inhibitors

Narrow-spectrum BLIs, like avibactam or a variety of benzoxaboroles/boronic acids, may be effective against class A, class C, and class D β-lactamases,⁶² but can be hydrolyzed by MBLs. MBLs are more difficult to resolve than SBLs due to

their structural heterogeneity and different hydrolysis mechanisms.⁶³ Bicyclic boron-containing compounds have been reported to bind to SBLs and MBLs in the sp^2 neutral state and subsequently interact with the nucleophilic serine of SBLs or the hydroxide ion of MBLs to form a tight sp^3 adduct, thus expanding their antibacterial profile. This mode of action differs from other BLI scaffolds.

Five typical broad-spectrum boron-containing β-lactamase inhibitors, VNRX-5133, QPX7728, QPX7831 (or in its prodrug form of QPX7728), VNRX-5236 (or in its prodrug form of VNRX-7145), and MS18 (►Fig. 8A), are separately discussed below.

VNRX-5133, also known as taniborbactam, is currently in clinical phase III. It is used in combination with cefepime to treat urinary tract infections and acute pyelonephritis.⁶⁴ VNRX-5133 is distinguished from vaborbactam and avibactam by its ability to inhibit most subclass B1 MBLs (VIM and NDM enzymes),⁶⁵ and in addition, VNRX-5133 maintained good inhibition of class A, C, and D SBLs. According to the structural analysis and biochemical properties, VNRX-5133 competitively inhibits SBLs and MBLs in a substrate-mimicking manner. The co-crystallization figures are given here to vividly demonstrate the interaction of VNRX-5133 with NDM-1 (►Fig. 8B, left) and VIM-2 (►Fig. 8B, right).

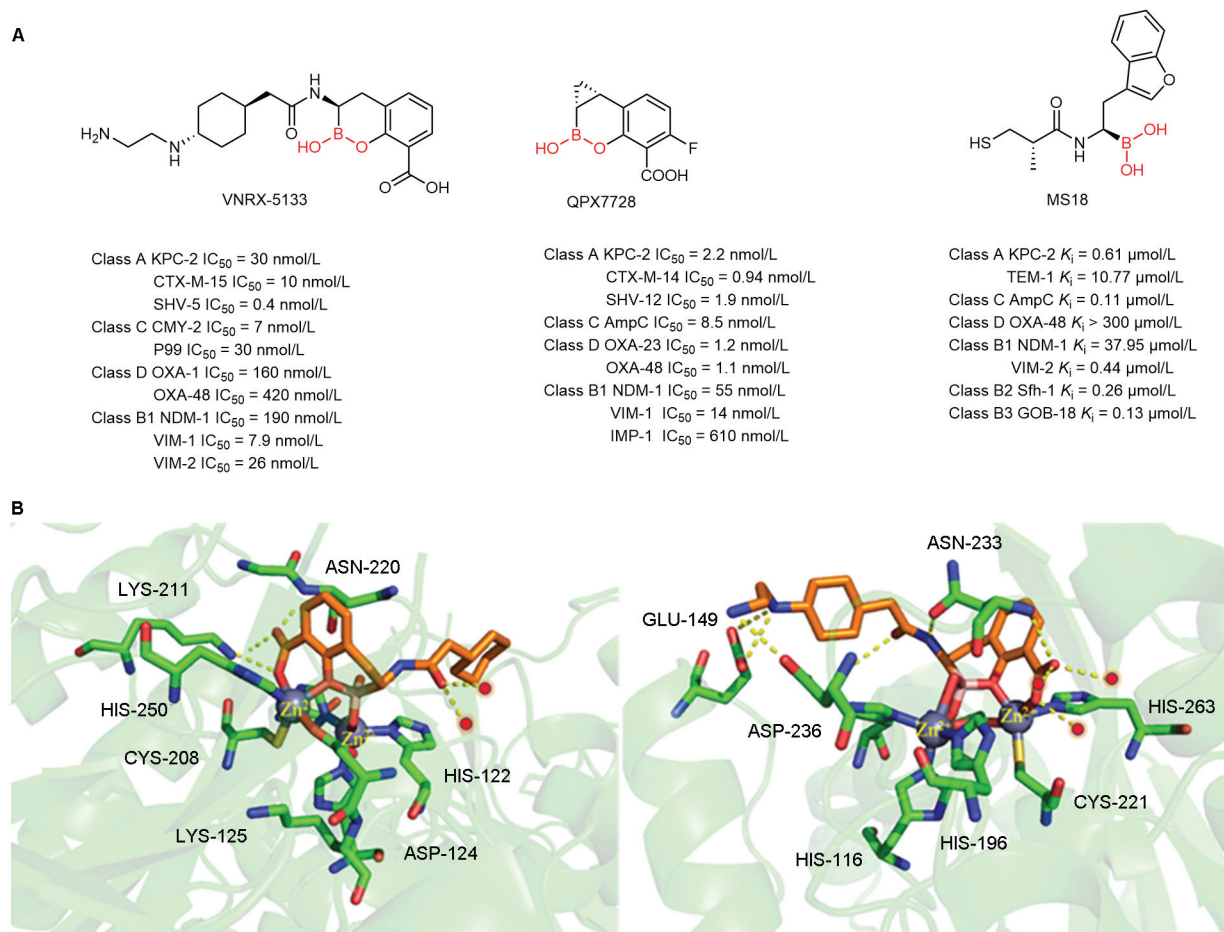


Fig. 8 Chemical structure and action mode diagram of boron-containing MBL inhibitors. (A) Typical boron-containing MBL inhibitors. (B) Co-crystallization analysis of VNRX-5133 with NDM-1 (PDB ID 6RMF) (left) and VIM-2 (PDB ID 6SP7) (right). The red dots represent water molecules.

Hecker et al developed QPX7728, which inhibits β -lactamases on an ultrabroad spectrum and is currently in phase I clinical trial.⁶⁶ QPX7728 showed potent effects on the whole bacterium *Pseudomonas aeruginosa*, which is much better than other BLIs including vaborbactam.⁶⁷ The K_i and IC_{50} values of QPX7728 and vaborbactam against various β -lactamases of class A, class B, and class D are shown in **Table 2**.^{66,68–70}

QPX7831, a prodrug of QPX7728, was synthesized by Reddy et al to boost the oral availability of QPX7728 in human subjects.⁷¹ It is cleaved by esterases in serum in many species. QPX7831 is now in phase I clinical trials.

VNRX-7145, synthesized by Trout et al,⁷² is the prodrug of VNRX-5236, a β -lactamase inhibitor for class A ESBLs, carbapenemases, class C cephalosporinases, and class D oxacillinases. VNRX-7145, along with another structurally similar prodrug QPX-7831, is a potent β -lactamase inhibitor and shows good prospects for the application of the prodrug

concept in addressing microbial resistance. In addition, both the prodrug molecules can be hydrolyzed by esterase (**►Fig. 9**).

Li's group identified a novel MBL inhibitor MS18,⁷³ with great inhibitory activity against MBLs, with K_i values of 0.44, 0.26, and 0.13 $\mu\text{mol/L}$ for VIM-2 (B1 subclass), Sfh-1 (B2 subclass), and GOB-18 (B3 subclass), respectively.

As shown in **►Fig. 6B**, MS18 utilizes the boronic acid moiety to covalently bind to SBLs and the sulfur atom of the thiol group to coordinately interact with MBLs. Through this action mode, the compound achieves dual-inhibitory activity against SBLs and MBLs.

In summary, BATSIs have been proven to be a practical approach to diminish microbial resistance induced by β -lactamases, and several BLIs have entered the market or clinical trials, and therefore more efforts should be devoted to finding boron-containing compounds to fight against drug-resistant MBLs.

Table 2 K_i and IC_{50} values of vaborbactam and QPX7728 against different β -lactamases

Enzyme	Class	K_i value (in $\mu\text{mol/L}$)		IC_{50} (in $\mu\text{mol/L}$)		Ref.
		Vaborbactam	QPX7728	Vaborbactam	QPX7728	
TEM-10	A	0.11	0.00066	0.47	0.0022	66,68
KPC-2	A	0.069	0.0019	0.11	0.0029	66,68
SHV-12	A	0.029	0.00074	0.056	0.0019	66,68
CTX-M-14	A	0.033	0.00029	0.11	0.00094	66,69
AmpC	C	0.035	0.0085	5	0.0085	66,68
OXA-23	D	>40	0.00074	120	0.0012	66,70
OXA-48	D	14	0.00028	6.9	0.0011	66,70
VIM-1	B1	>40	0.008	>160	0.014	69,70
NDM-1	B1	>40	0.032	>160	0.055	69,70
IMP-1	B1	>40	0.22	>160	0.61	69,70

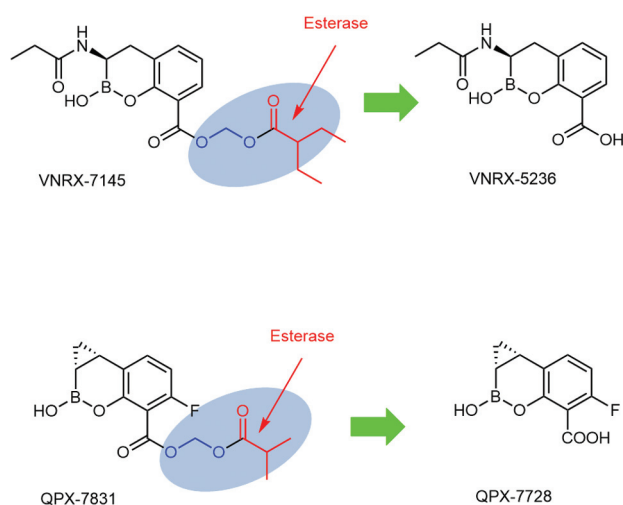


Fig. 9 VNRX-7145 and QPX-7831 as two prodrugs of β -lactamase inhibitors with similar chemical structures and hydrolyzing mechanisms.

Leucyl-tRNA Synthetase Inhibitor

Introduction and Mechanism of Leucyl-tRNA Synthetase

Amino acyl-tRNA synthetases (aaRSs) are an ancient family of proteins that widely exist in all pathogens. They are responsible for catalyzing the covalent binding of amino acids to their corresponding transfer RNA (tRNA), providing raw materials for protein synthesis on the ribosome. Hence, aaRSs are regarded as a key factor in the protein biosynthesis process. In a few cases, aaRSs are structurally completely distinct from their eukaryotic counterparts, thus allowing selective targeting.^{74,75} In addition, aaRSs have a low rate of drug resistance because they are less likely to have mutations in their genes, making aaRSs promising targets for the treatment of bacterial infection.⁷⁶ Currently, aminoacyl-tRNA synthetases have been clinically validated as antibacterial biotargets for mupirocin (brand name Bactroban), a marketed aaRS inhibitor targeting iso-leucyl-tRNA synthetase (IleRS), which can be used as an antibacterial agent for clinical trauma infections.

Among the 20 kinds of aaRSs, LeuRS is characterized by having a unique synthesis and editing site.⁷⁷ LeuRS is a class I aaRS with two active synthesis sites at a distance of 30 Å. One of them can aminoacylate tRNA^{Leu}, and the other one is an editing site that assures the accuracy of the translation process through proofreading.⁷⁸ LeuRS is an important bacterial enzyme that catalyzes the coupling of the amino acid leucine at its corresponding position to form tRNA^{Leu}, which can be used by the ribosome for protein synthesis. Moreover, LeuRS has been relatively more thoroughly studied in biochemical and structural aspects compared with other kinds of aaRSs. The inhibition of LeuRS blocks protein synthesis and stops the growth of the bacteria, making LeuRS an intriguing target to fight against bacteria.⁷⁹ Recent studies in structural biology and biochemistry have shown that the structure of LeuRS in prokaryotes is highly conserved, which provides the basis for the development of novel and selective antibiotics targeting LeuRS.

Considering the successful story of tavaborole and the feasibility of using LeuRS as an antimicrobial target, medicinal chemists are focusing on the development of boron-containing compounds to exploit the boron atoms' unique transformative ability to interfere with protein biosynthesis and cause bacterial death.

Boron-Containing Compounds Used as Leucyl-tRNA Synthetase Inhibitors

Benzoxaboroles are newly developed antibiotics that show prominent antimicrobial activity against gram-negative bacteria by inhibiting the editing site of the LeuRS enzyme.^{80,81} For example, mechanism studies of AN2690, also known as tavaborole, whose chemical structure is shown in ►Fig. 1, indicate that the compound specifically binds to the editing domain of leucyl-tRNA synthase.⁸²

In general, benzoxaboroles play a role in the inhibition of leucyl-tRNA through forming a tetrahedral adduct between the oxaborole moiety and ribose of terminal adenosine phosphate of tRNA in the transition state, i.e., boron atom bonds to the *cis*-diols of the 3'-terminal adenosine nucleotide Ade76 of tRNA^{Leu}, referred to as oxaborole tRNA-trapping (►Fig. 10).⁸³

AN2679 (►Fig. 11A) is a preclinical compound synthesized by Palencia et al.⁸⁴ As a lead compound, it provides a

classic scaffold for later modifications and SAR experiments. SAR studies have shown that the 3-aminomethyl group is essential for inhibitory activity, and this functional group can also be found in compounds GSK2251052, GSK656, and DS86760016.

GSK2251052 is a novel extended-spectrum antibiotic boron-containing compound that selectively inhibits LeuRS.⁸⁵ It has been designed and synthesized to combat infections caused by multidrug-resistant (MDR) gram-negative pathogens. This compound inhibits LeuRS function ($IC_{50} = 0.31 \mu\text{mol/L}$) by binding to the 3'-terminus of tRNA^{Leu} located in the editing active site of LeuRS to form a tetrahedral complex and ultimately lead to bacterial death. The hydroxyl group of the compound contributes to the interaction with the terminus adenosine ribose (A76) of LeuRS.⁸⁶

GSK2251052 has been used in phase II clinical trials for the treatment of complicated urinary tract infections and inner-abdominal infections. Unfortunately, the clinical trial was terminated due to the rapid emergence of microbial resistance.⁸⁷ Subsequent studies showed that GSK2251052, which is used to treat bacterial infections, causes mutations in amino acid residues at several sites in the editing center of bacterial LeuRS.^{88,89} These mutations weaken the binding of GSK2251052 to LeuRS, preventing the drug from forming a stable complex with the enzyme and tRNA.

GSK656, synthesized and evaluated by Li et al.,⁹⁰ is currently the only leucyl-tRNA inhibitor in clinical trials (phase II) for the treatment of tuberculosis. GSK656 is structurally similar to GSK2251052 but with one less carbon atom on the aliphatic chain and the introduction of a chlorine atom at position 4 of the benzene ring. The hydroxy group on the aliphatic chain (C-7 position) enhances the hydrophilicity, which significantly improves the selectivity for *Mycobacterium tuberculosis* LeuRS (*Mtb*LeuRS) for human mitochondrial LeuRS ($IC_{50} = 300 \mu\text{mol/L}$) and human cytoplasmic LeuRS ($IC_{50} = 132 \mu\text{mol/L}$).

DS86760016 is a newly developed LeuRS inhibitor effective against MDR gram-negative bacteria. It was synthesized at Daiichi Sankyo Indian Pharma Pvt. Ltd. and is currently undergoing a preclinical trial. DS86760016 has a favorable pharmacokinetic profile against extended-spectrum drug-resistant *P. aeruginosa* and other MDR gram-negative bacteria (e.g., *E. coli*, *K. pneumoniae*).⁸⁷ The MIC values of DS86760016 against gram-negative bacteria were 0.25 to 2 $\mu\text{g/mL}$, which is close to the inhibitory efficacy of GSK2251052. In contrast, DS86760016 is more stable in the human body and therefore interacts with bacterial pathogens for a longer period of time. The good antimicrobial capacity and stability of DS86760016 make it a potential candidate for clinical trials.

Zhou's group screened benzoxaborole group analogs against *S. pneumoniae* LeuRS (*Sp*LeuRS). As a result, ZCL039 and compound 1 were discovered and synthesized as benzhydrol-oxaborole boron-containing inhibitors against *Sp*LeuRS (►Fig. 11A).^{91,92}

ZCL039 is a promising inhibitor against *S. pneumoniae* (MIC = 5 $\mu\text{g/mL}$) but has less activity against *E. coli* (MIC = 60 $\mu\text{g/mL}$). The IC_{50} values of ZCL039 against

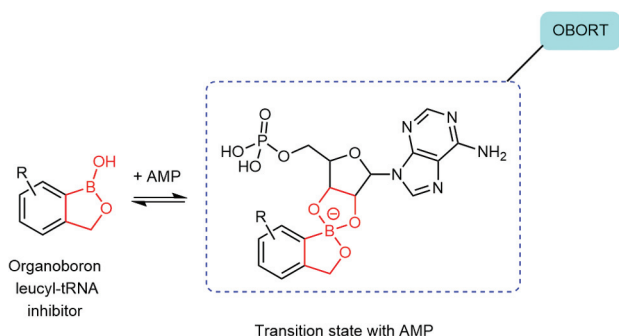


Fig. 10 General mechanism of benzoxaborole analogues that exert its unique chemical property to combine with AMP. AMP, adenosine monophosphate; OBORT, oxaborole tRNA-trapping.

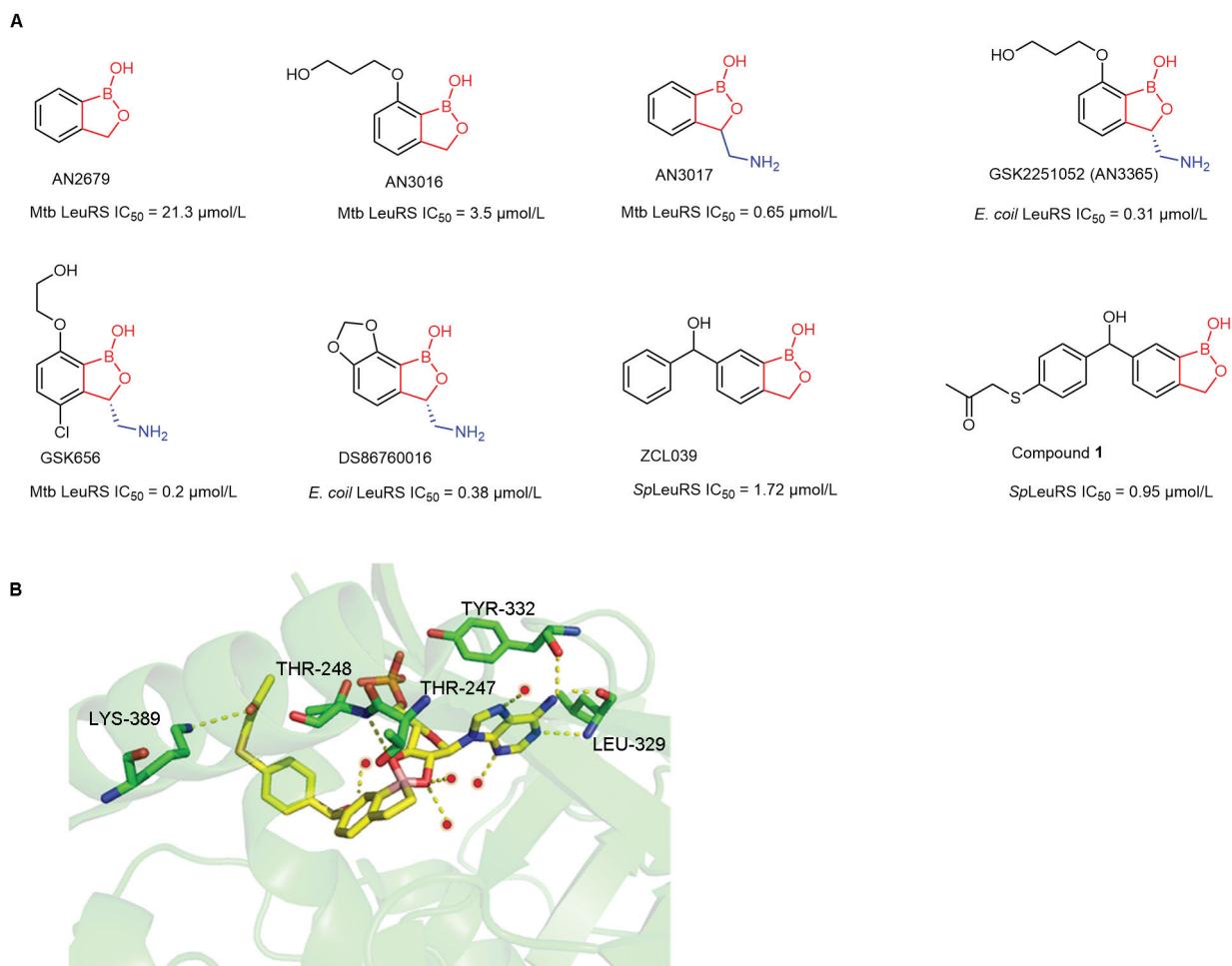


Fig. 11 Classic scaffold of LeuRS inhibitors and their action mode. (A) Typical LeuRS boron-containing inhibitors; (B) Binding mode of compound 1 with AMP in *SpLeuRS* (PDB ID 7BZJ). AMP, adenosine monophosphate; LeuRS, leucyl-tRNA synthetase.

S. pneumoniae and *E. coli* were 1.72 and 8.25 μmol/L, respectively. The IC₅₀ values of ZCL039 for LeuRS in both human mitochondria and cytoplasm were higher than 250 μmol/L, indicating that ZCL039 has good selectivity. The efficacy of ZCL039 depends on the presence of tRNA.⁹² The researchers hypothesized that ZCL039 targets two mutation sites, T252R and Y332D, in the structural domain of CP1, which may affect the binding between ZCL039 and tRNA^{Leu}. In addition, Thr252 residue is a structural component of the binding pocket of benzoxaborole compound,⁸³ and Tyr332 may contribute to tRNA 3'-terminus binding.⁹³ This facilitates the latter modifications based on existing compounds.

Compared with ZCL039, a carbonyl group was introduced to the aliphatic chain of compound **1**. Co-crystallization analysis of compound **1** with *SpLeuRS* (► Fig. 11B) revealed that the carbonyl group could form a hydrogen bond with Lysine 389, thus enhancing the interactions between organoboron molecules and *SpLeuRS*. Compound **1** inhibited *SpLeuRS* with an IC₅₀ value of 0.95 μmol/L, and a MIC value of 6 μg/mL (anti-pneumococcal activity). From the perspective of protein-molecular interactions, compound **1** has a better binding ability than ZCL039 due to an additional hydrogen bond.

An increasing number of boron-containing leucyl-tRNA inhibitors have been reported in recent years, but only a few typical inhibitors are listed and discussed here. Although AN2690 is marketed for the treatment of fungal infections, the development of organoboron compounds by inhibiting leucyl-tRNA may have a prosperous future for addressing drug-resistant issues with these newly developed inhibitors.

Inhibitors of LexA Self-Cleavage

SOS and LexA Self-Cleavage

The bacteria SOS response system is indispensable in maintaining bacterial genomes. The coordinated cellular response to bacterial DNA damage was first described in *E. coli* in detail and named the SOS response by Miroslav Radman in the 1970s.⁹⁴ Bacteria use the coordinated cellular response to recover from DNA damage, a process controlled by the RecA and LexA proteins.⁹⁵ In growing and unstressed *E. coli* cells, the SOS system is repressed by LexA of approximately 50 promoters, which control the expression of the SOS regulon.^{96,97} The RecA/LexA axis of the bacterial SOS response system is currently a promising drug biotarget that can be used to overcome drug resistance.⁹⁸

The SOS response system is caused by bacterial DNA damage and depends on DNA replication to produce an SOS signal, i.e., accumulation of single-strand DNA (ssDNA). RecA connects with ssDNA to form filaments, which promotes proteolytic cleavage of the LexA repressor. More than 40 of these genetic products can be expressed to help with DNA repair.⁹⁹ The accumulation of ssDNA caused by replication of damaged DNA is seen as a signal to ignite the SOS response system.¹⁰⁰ In addition, approximately 80% of LexA is DNA-bound, and the rest is free, which is the target of RecA* (the activated RecA).⁹⁹

LexA contains 12 serine residues, five of which are located in the C-terminal domain (CTD) and seven in the N-terminal domain (NTD). The CTD has protease activity, while the NTD has DNA-binding activity.¹⁰¹ Around the landing site, the LexA repressor is thought to permeate and slide along the nonspecific DNA rotational coupling, which helps to find specific binding sites.¹⁰²

The SOS system relies on the interplay of two components, which are key regulatory proteins, repressors and inducers, similar to a pair of switches to control the alternation between on and off states.¹⁰³ LexA is a repressor and downregulates its expression during normal bacterial growth. RecA is an inducer that binds to ssDNA to form a filament when bacterial DNA is damaged. The filaments stimulate self-cleavage of the scissile peptide bond between Ala84 and Gly85, causing a large conformational change of LexA in its CTD.¹⁰¹ In the end, LexA dissociates from its binding site (SOS boxes) and causes activation of bacterial SOS regulons.⁹⁵ During self-cleavage of LexA, the CTD undergoes a dramatic conformational change between the active and inactive states, allowing the cleavage ring to move around the active site.^{101,104}

LexA's affinity for its sequence allows the fine regulation of gene expression. Thus, the inactivation of LexA

prevents bacteria from developing a drug-resistance phenotype.^{105,106} Fig. 12 shows this process and how LexA inhibitors play a role in this process.

Boron-Containing Compounds Used to Inhibit LexA Self-Cleavage

Compounds targeting RecA have been reported to have many mammalian homologs that may cause side effects, making such compounds unsuitable for drug design.¹⁰⁷ In contrast, LexA autoproteolysis is unique to the prokaryotic SOS response. Therefore, targeting LexA autoproteolysis is more likely to obtain high selectivity between human cells and bacteria.

The formation of a tetrahedral intermediate with a boron atom is a novel idea for the design of LexA self-cleavage inhibitors. In this article, four boron-containing compounds are outlined (Fig. 13A), which were found to have the potential to form covalent interactions with Ser119 through simulations with virtual reaction software (Fig. 13B).

The intramolecular reaction is known as hydrolysis of the amide bond between Ala84 and Gly85, which is mediated by a transient tetrahedral intermediate formed by catalytic Ser119 with the peptide bond between Ala84 and Gly85. The reactivity of boronic acid compounds is due to their Lewis acidic nature, which allows the formation of a tetrahedral adduct with nucleophiles through covalent bonding. In addition, the presence of Lys156 helps to polarize the hydroxy group of serine, increasing its nucleophilicity and enhancing the covalent binding ability of serine to the boron atom. Moreover, hydrogen bonds within Ser-116, Val-153, and compound 2 enhanced the interaction between the inhibitor and LexA. Thus, the use of phenylboronic acid derivatives to inhibit Ser119 activity in LexA has shown promising applications through theoretical and docking validation (Fig. 13B).

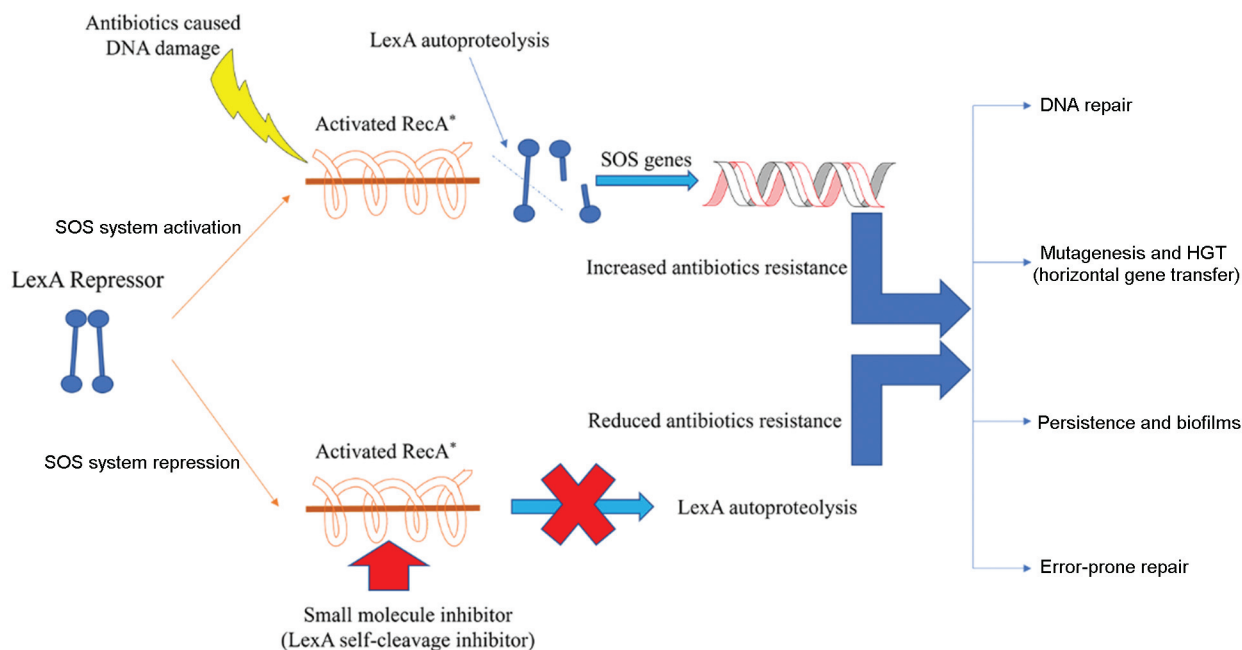


Fig. 12 LexA inhibitors targeting self-cleavage to inhibit bacterial SOS response system.

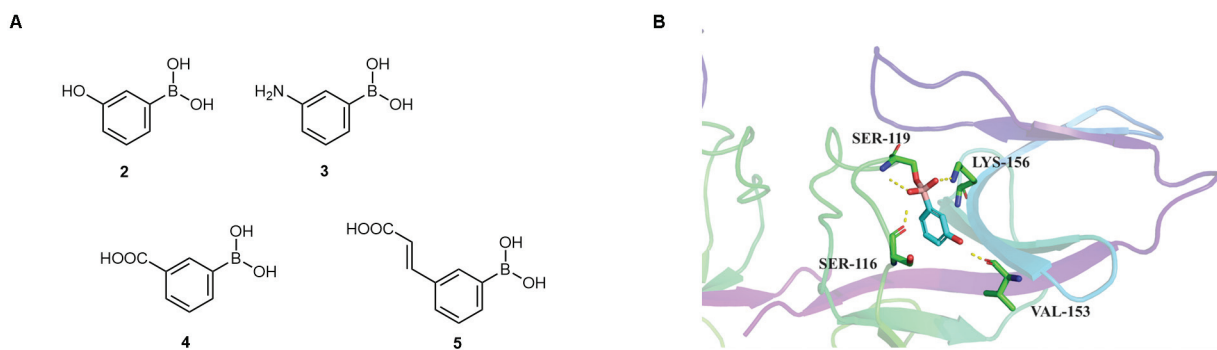


Fig. 13 Boron-containing LexA inhibitors and binding mode with LexA. (A) Chemical structure. (B) Docking model of compound 2 in LexA-binding site (PDB ID 1JHF). The docking was operated by the LibDock portion of Discovery Studio 3.1.

To date, the main drawbacks of boron-containing compounds for targeting LexA are the lack of sufficient inhibitors, detailed enzyme activity, and *in vitro* and *in vivo* antibacterial data. Despite the limitations of the current work, LexA offers potential as a new biological target for the use of boron-containing inhibitors to overcome microbial drug resistance.

Efflux Pump Inhibitor

Efflux Pump in Drug Resistance

Among several mechanisms by which MDR bacteria respond to antibiotics, the efflux pump is one of the major mechanisms by which bacteria confront drug treatment. The efflux pump is initially identified in mammalian cancer cells, and this kind of antibiotic transport system was reported in the bacterial domain in the early 1980s as a cause of tetracycline resistance.¹⁰⁸ The survival of pathogens depends not only on the production of enzymes that can inactivate drugs but also on the efflux pumps that export drugs.^{109,110}

Efflux pumps are located on the bacterial cell membrane and consist of an inner-membrane pump, periplasmic adaptor proteins, and an outer membrane channel that regulates the extrusion of toxic substances from the cell interior to the external cell circumstance. Efflux pumps occur in almost all species of bacteria, and the genes coding them are either plasmid-mediated or found in the bacterial genome.¹¹¹ Many substrates, such as antibiotics, toxins, or detergents, are exported via efflux pumps, which are substrate-specific or act against a range of structurally different materials.¹¹² MDR-related efflux pumps can be divided into five classes: ATP-binding cassette (ABC), small MDR, multidrug and toxin extrusion, resistance-nodulation division (RND), and major facilitator superfamily (MFS).^{113–117} Except for the ABC, which utilizes energy produced by ATP hydrolysis, the other kinds of efflux pumps utilize energy generated by the proton-driven electrochemical gradient across the membrane.¹¹⁸ The three kinds of efflux pumps in gram-negative bacteria are the RND (AcrB), ABC (MacB), and MFS (EmrB) families (►Fig. 14).

Understanding how efflux pumps exclude antibiotics and the different categories of efflux pumps in various bacteria is essential for designing effective efflux pump inhibitors

(EPIs). An overview of efflux pumps in specific microorganisms is listed in ►Table 3.^{119–132}

Nevertheless, bacteria often achieve resistance through various and synergistic pathways. For example, resistance to aminoglycoside antibiotics in *Pseudomonas aeruginosa* is mainly due to the synergistic effect of aminoglycoside-modifying enzymes, efflux pumps, and RNA methylases. The same occurred with β -lactam antibiotics, where resistance is synergistically mediated by β -lactamases and efflux pumps that exclude the antibiotics.¹³³

Efflux Pump Inhibitor

EPIs increase the concentrations of antibiotics in bacterial cells, allowing the drugs to kill microorganisms more effectively. Many inhibitors have been discovered in the past two decades due to the prospect of developing EPIs.¹³⁴ Disruption of the MexB gene-coding in *P. aeruginosa* led to reduced MIC values for a variety of antibiotics (e.g., aminoglycosides); therefore, *P. aeruginosa* mutants were used for EPI screening.¹³⁵ In addition, based on previously reported structural data, several amino acid residues may be involved in interactions with transport substrates.¹³⁶

Initial efforts to create novel EPIs focused not only on directly inhibiting the function of efflux pumps but also on inhibiting the transcription of genes encoding efflux pumps, or inhibiting efflux pumps by altering the structure of EPIs to mimic the conformation of a specific substrate. Designing EPIs to block the activity of these pumps could be a promising method to restore the function of antibiotics as substrates for efflux pumps. For example, peptidomimetic compounds (PA β N) have been used as efflux inhibitors to eradicate infections caused by *P. aeruginosa*, which is responsible for a high percentage of nosocomial infections. PA β N is a competitive inhibitor that acts as a substrate for efflux pumps, thereby rejuvenating antibiotics. Unfortunately, PA β N cannot be used in the clinic because it is toxic to eukaryotic cells,¹³⁵ therefore, medicinal chemists should create other small-molecule drugs to better inhibit efflux pumps.

Boron-Containing Compounds Used as Efflux Pump Inhibitors

MRSA is a gram-positive bacterium that has caused serious community- and hospital-acquired pathogenic

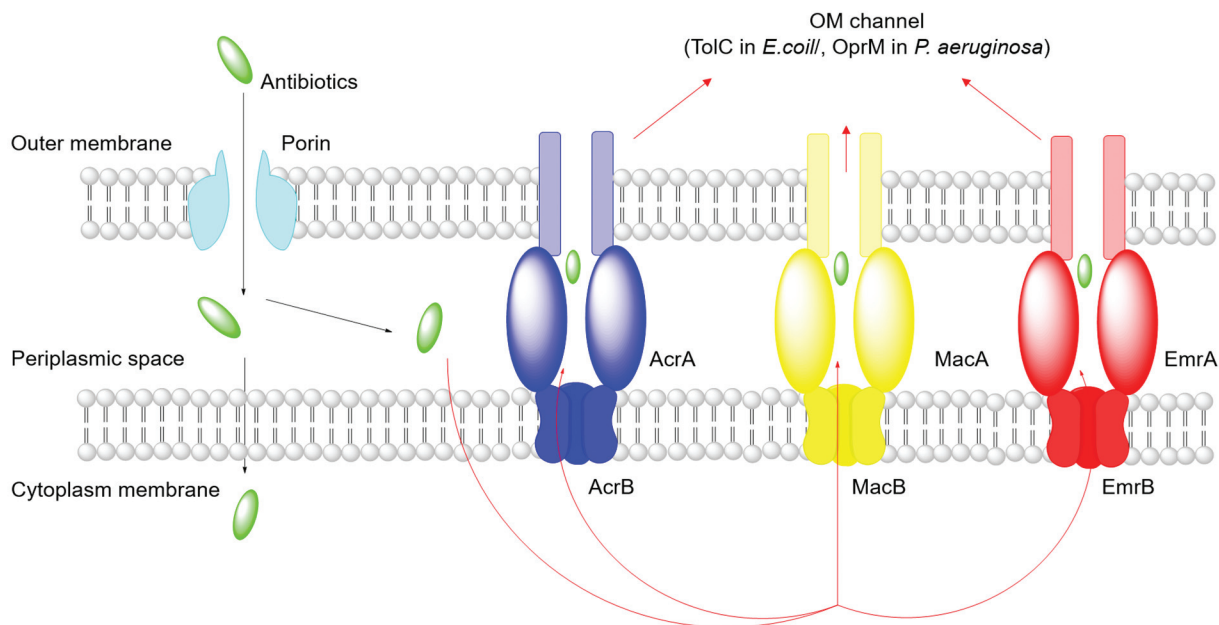


Fig. 14 Schematic presentation of efflux pump superfamily located at gram-negative bacteria and attached with specific antibiotics extruded by corresponding efflux pump. AcrB belongs to the RND family and effluxes a wide range of drugs. MacB belongs to the ABC family and mainly extrudes macrolide antibiotics. EmrB belongs to the MFS family and effluxes nalidixic and novobiocin.

Table 3 Efflux pumps found in specific bacteria, along with those belonging to superfamily, and likely substrates

Bacteria strain	Main efflux pump	Superfamily	Likely substrates	Ref.
Gram-negative bacteria				
<i>A. baumannii</i>	AdeABC	RND	Aminoglycosides, fluoroquinolones, tetracyclines	119
<i>E. coli</i>	AcrAB-TolC	RND	β -Lactams, ciprofloxacin, chloramphenicol, fluoroquinolones, tetracycline	120,121
<i>P. aeruginosa</i>	MexAB-OprM	RND	β -Lactams, macrolides, fluoroquinolones, tetracycline, novobiocin, chloramphenicol	122
<i>S. enterica</i>	AcrAB-TolC	RND	Nalidixic acid, tetracycline, erythromycin, norfloxacin, fluoroquinolones, rifampicin	123,124
<i>K. pneumoniae</i>	AcrAB-TolC	RND	Cefepime, tetracycline, levofloxacin, chloramphenicol, ciprofloxacin	125
Gram-positive bacteria				
<i>S. aureus</i>	NorA	MFS	Fluoroquinolones, norfloxacin, ciprofloxacin	126,127
<i>S. pneumoniae</i>	PmrA	MFS	Fluoroquinolones, ciprofloxacin	128
<i>B. subtilis</i>	BmrA	ABC	Chloramphenicol, fluoroquinolones, doxorubicin	129,130
Mycobacteria				
<i>M. tuberculosis</i>	DrrABC	ABC	Daunorubicin, doxorubicin, tetracycline	131
<i>M. smegmatis</i>	LfrA	MFS	Fluoroquinolones, ethidium bromide	132

infections.^{137,138} NorA, present in all MRSA strains, is a multidrug efflux pump belonging to the core genome of *S. aureus* and encoded by the chromosome.¹³⁹ Through an experiment using the *S. aureus* 1199B strain that overexpresses the NorA efflux pump, medicinal chemists discovered several boron-containing compounds (–Fig. 15) that can restore the activity of ciprofloxacin.^{140,141} When these compounds were combined with antibiotics to inhibit *S. aureus*, the minimum modulatory concentration (MMC) values dropped dramatically and were much lower than the MMC values of inhibitor analogs in combination with antibiotics against *S. aureus*.

Further, SAR experiments showed that boron is indispensable for the antiresistance activity and identified pyridine-3-boronic or pyridine-4-boronic compounds with better efficacy. Statistics showed that 11 compounds belonging to the pyridine-3-boronic acid derivatives could enhance the efficacy of ciprofloxacin, suggesting that the pyridine-3-boronic acid scaffold could be used for further studies. In addition, the MIC values of boron-containing EPIs typically exceed 64 µg/mL, which means that they have little or even no antibacterial activity when used alone.

The above example provides an extra approach to using boron-containing compounds as EPIs to restore the activity of antibiotics including ciprofloxacin.

Conclusion and Future Perspectives

Bacterial pathogens have developed a considerable number of resistance mechanisms to the existing antimicrobials, posing a growing threat to human beings. Due to the unique property of the boron atom that can covalently bind to nucleophiles, boron-containing compounds have been used as antibacterial adjuvants or antimicrobials to reduce drug resistance.

This review summarizes the research progress of boron-containing compounds to overcome bacterial resistance. Among these types of inhibitors, boron-containing β-lactamase inhibitors in combination with marketed β-lactam antibiotics for the treatment of drug-resistant bacteria

have reached the clinical stages. Other inhibitors, such as LexA self-cleavage inhibitors and EPIs, still lack sufficient *in vivo* assays to demonstrate their efficacy and safety. In terms of mode of action, boron-containing inhibitors have made concrete progress in inhibiting β-lactamases and leucyl-tRNA. However, other mechanisms involving LexA self-cleavage and efflux pumps are similar to a theoretical concept, and the specific inhibitory effects need to be confirmed by further experimental studies.

To better address the problems encountered with the use of boron-containing compounds for bacterial resistance, scientists have devoted more efforts to the following issues during their studies. The first is bioavailability. Taking QPX7728, which has poor oral bioavailability, as an example, researchers developed the prodrug QPX7831 and confirmed its enhanced oral bioavailability in animal models *in vivo*. Therefore, the prodrug concept is an alternative strategy to improve the bioavailability of compounds rather than a structural modification methodology. The second is how to extend the inhibition spectrum of boron-containing compounds. The mentioned inhibitors of MBLs, except MS18, which has inhibitory activity against B2 or B3 MBLs, mainly target B1 MBLs but have slight or even no inhibitory activity against B2 or B3 MBLs. These deficiencies of existing MBL inhibitors require further structural modifications to improve their inhibitory efficacy against B2 and B3 MBLs, and the discovery of pan-spectrum boron-containing BLIs is desired. The third is the emerging drug resistance. Organoboron compounds were synthesized to cope with drug resistance, but bacteria rapidly developed resistance toward these compounds after clinical trials. Elucidation of the mechanism of GSK2251052-induced resistance will contribute to the further discovery of potent and selective benzoxaborole LeuRS inhibitors. The fourth is the toxicity of boron-containing compounds. Although listed boron-containing drugs such as bortezomib can cause serious toxic reactions, the toxicity of bortezomib is primarily due to its mechanism of action rather than the presence of boron fragments. In addition, several studies have shown that trace intake of boron is beneficial for brain function and bone growth.¹⁴²

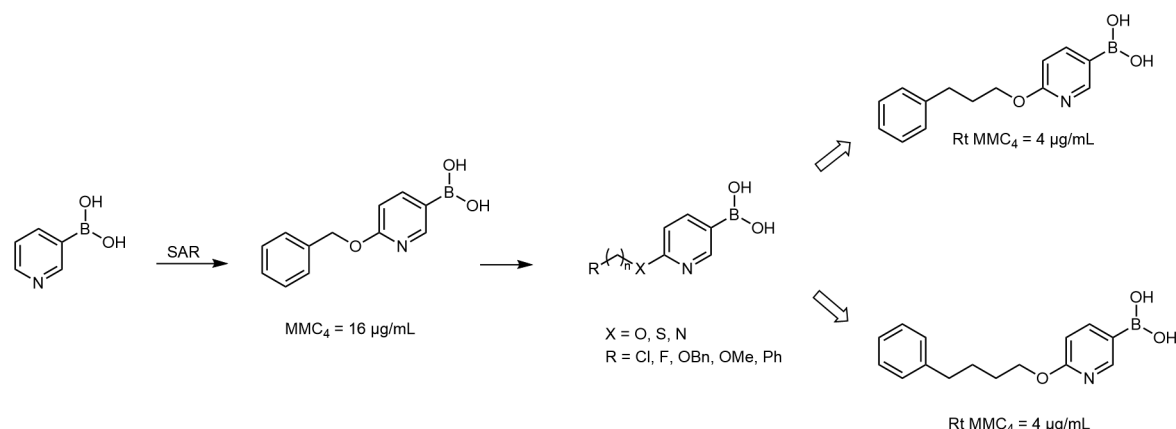


Fig. 15 Structural modification of novel heterocyclic scaffold, and corresponding MMC₄ values for promising compounds, MMC: minimum modulatory concentration, which was measured combined with ciprofloxacin (at subinhibitory concentration MIC/4, 2 µg/mL); Rt: resistant *S. aureus* 1199B (NorA).

Thus, the toxicity of boron-containing compounds can be circumvented by rational drug design in the development of antimicrobial compounds. The fifth is the stability of boron-containing compounds. In the case of boronic acid compounds, for example, they degrade easily before binding to the target compared with electrophilic compounds such as acrylates and aldehydes,¹⁵ which causes problems such as off-targeting and reduced efficacy. The sixth is the synthesis of organic boric acid compounds. The preparation of boric acid is both difficult and expensive.^{143,144} It needs to purchase expensive borate esters and hydrolyze them under hydrochloric acid conditions to obtain the target product. Therefore, it is necessary to develop economically feasible synthetic methods to advance the synthetic preparation of boric acid compounds in the future. The seventh is the depth of clinical application of boron-containing compounds in the antimicrobial field. Currently, boric acid is mainly used to interact with serine residues, while the interaction with other nucleophiles, e.g., arginine, tyrosine, and threonine, has been poorly studied. Therefore, by expanding the application of boric acid to other nucleophiles and discovering antimicrobial mechanisms based on other nucleophiles, boron-containing drugs will uncover a huge space for clinical applications of antimicrobials.

In the future, medicinal chemists could build a library of boron-containing compounds to screen the activity against drug-resistant bacteria or as a probe to find new therapeutic targets. Designing a novel class of antibiotics by merging the scaffold of existing antibiotics and boron-containing groups may be another direction, and the potential advantage of this idea is that an antibiotic could possess original antibacterial activity and further kill certain kinds of drug-resistant bacterial pathogens. In conclusion, the use of boron-containing inhibitors to address bacterial resistance will be important for the treatment of clinical bacterial infections and deserves further exploration.

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Conflict of Interest

None declared.

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