Drugs of the Future 1999, 24(8): 839-846

CCC: 0377-8282/99

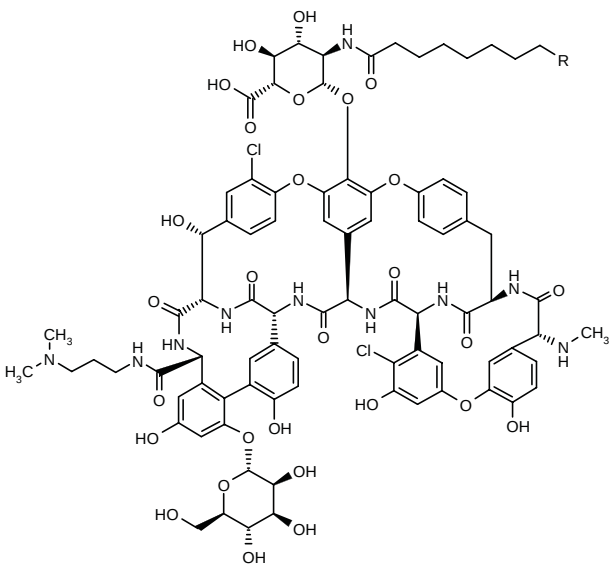
BI 397

Glycopeptide Antibiotic

Semisynthetic glycopeptide antibiotic complex of six main related homologues, namely A_0, A_1, B_0, B_1, C_0, C_1, which only differ in the structure of the *N*-acyl side chain of the *N*-acylaminoglucuronic acid at position 56; factor B_0 is the main component, with >75% of the whole complex

BI 397 complex

EN: 279026



Factor A_0: R = $CH(CH_3)_2$

EN: 279027

Factor A_1: R = $CH_2CH_2CH_3$

EN: 279028

Factor B_0: R = $CH_2CH(CH_3)_2$

EN: 264176

Factor B_1: R = $CH_2CH_2CH_2CH_3$

EN: 279030

Factor C_0: R = $CH_2CH_2CH(CH_3)_2$

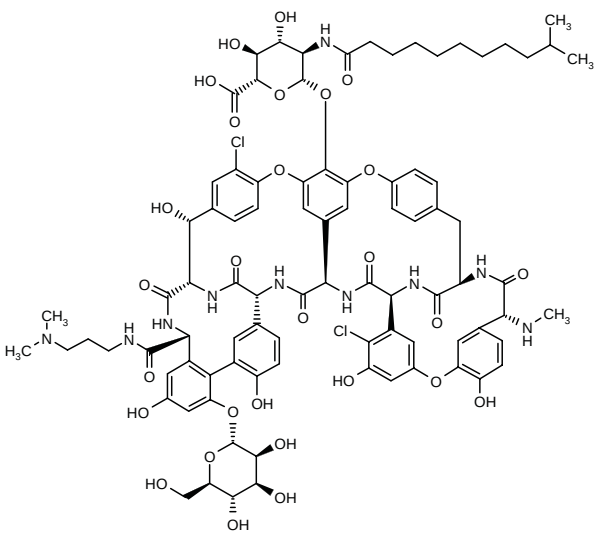
EN: 279031

Factor C_1: R = $(CH_2)_4CH_3$

EN: 279032

BI 397 factor B_0

(3*S*,15*R*,18*R*,34*R*,35*S*,48*S*,50a*R*)-5,31-Dichloro-56-[2-deoxy-2-(10-methyl-1-oxoundecylamino)-6-carboxy]-β-D-glucopyranosyloxy]-*N*-[3-(dimethylamino)propyl]-6,11,34,40,44-pentahydroxy-42-(α-D-mannopyranosyloxy)-15-(methylamino)-2,16,36,50,51,59-hexaoxo-2,3,16,17,18,19,35,36,37,38,48,49,50,50a-tetradecahydro-1*H*,15*H*,34*H*-20,23:30,33-dietheno-3,18:35,48-bis(iminomethano)-4,8:10,14:25,28:43,47-tetramethenodioxadiazacyclooctacosino[4,5-*m*][10,2,16]benzoxadiazacyclotetracosine-38-carboxamide



$C_{88}H_{100}Cl_2N_{10}O_{28}$ Mol wt: 1816.7060

CAS: 171500-79-1

EN: 264176

Synthesis

BI 397 (1) is the 3-(dimethylamino)-1-propylamide at the peptide-carboxy group of the natural glycopeptide

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A-40,926 (2, 3). BI 397 is prepared from A-40,926 following a three-step procedure (Scheme 1) which consists of: 1) selective methyl esterification of the *N*-acylaminoglucuronic acid function, 2) amidation of the peptide-carboxy group and 3) saponification of the sugar methyl ester. Steps 2 and 3 are performed in one-pot.

Step 1): Reaction of A-40,926 (A) (I) in methanol in the presence of H_2SO_4 at 0-5 °C for 24 h gives the monomethyl ester (MA) (II) at the sugar-carboxy group (80% yield).

Step 2): Amidation at the peptide-carboxy group is carried out by reaction of MA (II) with 3-(dimethylamino)-1-propylamine (DMEPA) (III) in DMSO in the presence of PyBOP to obtain the methyl ester (MA-A-1) (IV) of BI 397; this compound is not isolated.

Step 3): Hydrolysis of MA-A-1 (IV) in the above DMSO solution with 15% NAOH gives BI 397 (A-A-1, 63% yield from MA).

Introduction

The commercially available glycopeptides vancomycin and teicoplanin are a very important part of the clinician's armamentarium against multiresistant Gram-positive pathogens. However, the clinical usefulness of these antibiotics is limited by some drawbacks. Vancomycin must be given slowly by intravenous administration because of its possible hypotensive effects related to histamine release; it has ototoxic and nephrotoxic potential and frequent administrations are necessary because of its relatively short half-life; in addition, VanC and VanB enterococci are poorly susceptible or resistant to vancomycin (4, 5). Teicoplanin, which is not registered for human use in the U.S., is poorly active against some coagulase-negative staphylococci (CoNS), particularly isolates of *Staphylococcus haemolyticus* which are intermediate or resistant to teicoplanin (6).

The need for a new potent, safe and easy-to-use glycopeptide is justified by the increasing incidence of serious nosocomial infections caused by major Gram-positive pathogens, particularly methicillin-resistant *Staphylococcus aureus* (MRSA) and CoNS which are often resistant to almost all other first-line antibiotics. Moreover, high-level penicillin resistance in *Streptococcus pneumoniae* is becoming widespread and increasing resistance to macrolides is limiting currently available therapeutic options. A further challenge for new glycopeptides is the activity against highly glycopeptide-resistant VanA enterococci which are also highly resistant to virtually all other classes of antibiotics (7, 8).

The mechanism of action of the glycopeptide antibiotics is by inhibition of the biosynthesis of the bacterial cell wall: they act by selectively binding to terminal D-alanyl-D-alanine-containing precursors of peptidoglycan, thus preventing the reactions of transglycosylation and transpeptidation. Resistance to glycopeptides in enterococci is due to the replacement of the target peptide by a D-Ala-D-lactate depsipeptide which prevents efficient binding and thus allows bacterial growth. While enterococci of the VanA phenotype are generally resistant to both vancomycin and teicoplanin, VanB enterococci remain susceptible to teicoplanin.

In recent years, much effort has focused on the search for new glycopeptide derivatives with enhanced efficacy against clinical isolates of MRSA and CoNS. Encouraging results have been obtained from a number of chemical derivatives of naturally occurring glycopeptides (9). Structure-activity relationships of some modified teicoplanin- and vancomycin-type compounds also indicated the possibility of achieving activity against highly glycopeptide-resistant enterococci. One of these derivatives, LY-333328 (Lilly) is currently undergoing phase II clinical trials for its promising activity against VanA enterococci.

BI 397 is a novel semisynthetic glycopeptide antibiotic, structurally related to but generally more potent than teicoplanin against teicoplanin-susceptible organisms. It has the additional advantage of providing coverage of all CoNS, including methicillin-resistant strains. BI 397 is especially indicated for the treatment of serious Gram-positive infections caused by MRSA, CoNS and streptococci, including penicillin-resistant *S. pneumoniae*.

Based on antibacterial activity data both *in vitro*, against a wide range of staphylococci, streptococci and enterococci, and *in vivo*, against strains of these species in animal models of infection predictable of efficacy in humans, BI 397 represents a significant improvement over vancomycin and teicoplanin, in terms of activity against glycopeptide-susceptible Gram-positive pathogens. BI 397 is more potent than vancomycin, teicoplanin and LY-333328 against MRSA and CoNS and is expected to be better than the other two glycopeptides as it shares the positive characteristics of each one without having their drawbacks. For example, like vancomycin, BI 397 is highly active on all CoNS species, including *S. haemolyticus* isolates poorly susceptible to teicoplanin, and on MRSA, including strains with reduced susceptibility to teicoplanin. And like teicoplanin, BI 397 is highly active on all streptococcal and non-VanA enterococcal species, including VanB and VanC phenotypes poorly susceptible or resistant to vancomycin. BI 397 shows high and prolonged plasma levels, suggesting that a once-daily administration may be possible; however, like vancomycin and teicoplanin, it is poorly active against most VanA enterococci.

Preclinical toxicology and safety pharmacology studies in rats and dogs have shown that BI 397 is well tolerated upon intravenous bolus administration at doses several times higher than the expected therapeutic dose, without causing side effects typical of bolus i.v. vancomycin, namely, skin changes (red-man syndrome) and hypotension.

These preclinical results suggest that BI 397 may be the ideal candidate for a second-generation glycopeptide, combining the excellent *in vivo* efficacy, safety profile and ease of administration of teicoplanin with high activity against MRSA and all CoNS species.

Scheme 1: Synthesis of BI 297 (Factor B_o)

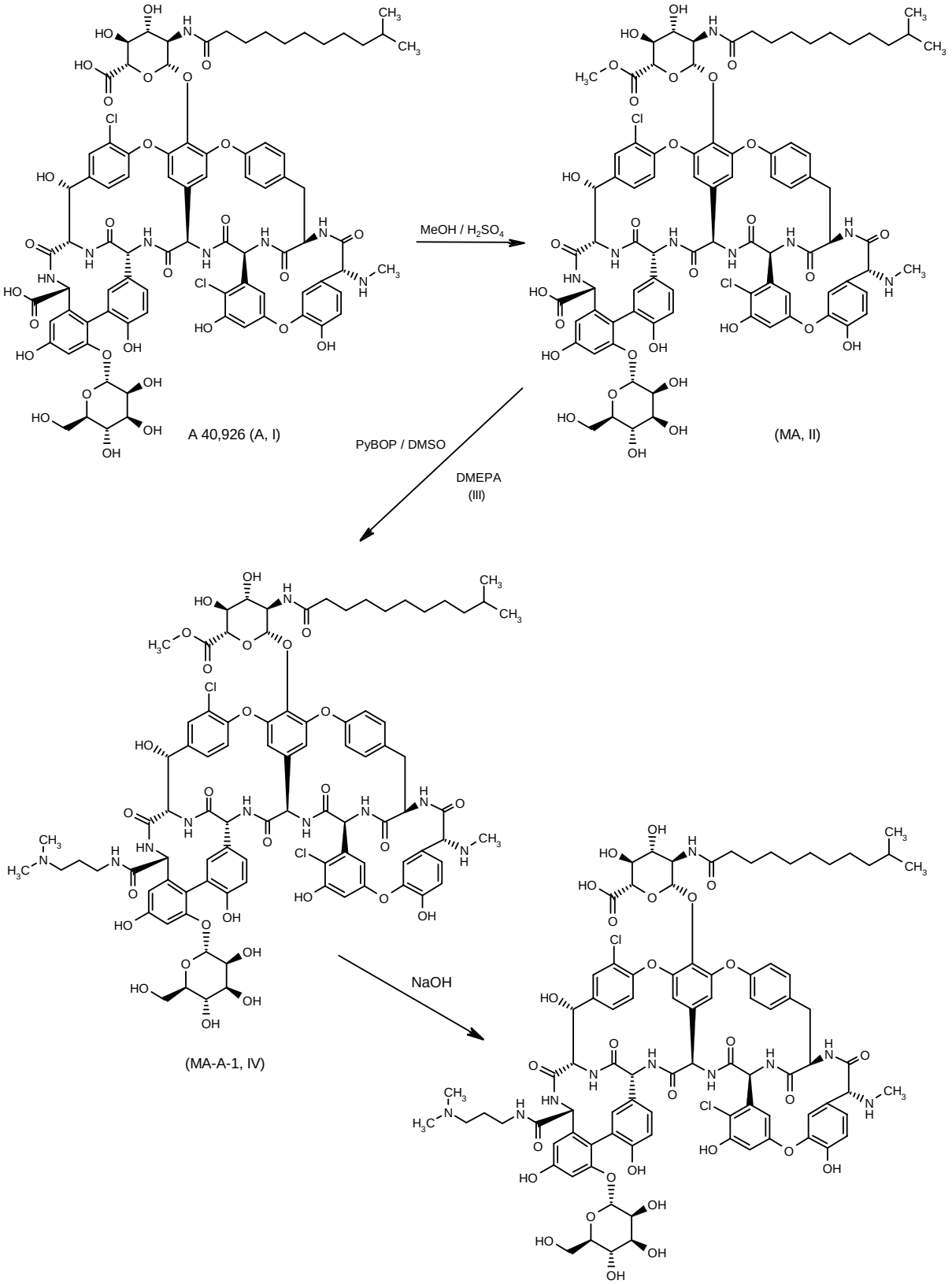

Table I: In vitro *antistaphylococcal activity (MIC, µg/ml).*

Organism (No. strains)	Antibiotic	MIC range	MIC_{90}
S. aureus			
Meth-S (10)	BI 397	≤0.03-0.5	0.13
	Teicoplanin	0.25-8	4
	Vancomycin	0.13-1	1
	LY-333328	0.13-1	1
Meth-R (23)	BI 397	0.06-1	0.25
	Teicoplanin	0.13-8	8
	Vancomycin	0.5-4	4
	LY-333328	0.13-4	2
S. epidermidis			
Meth-S (13)	BI 397	≤0.03-0.25	0.25
	Teicoplanin	0.25-16	8
	Vancomycin	0.13-1	1
	LY-333328	0.25-1	2
Meth-R (12)	BI 397	≤0.03-1	0.25
	Teicoplanin	1-16	16
	Vancomycin	1-4	4
	LY-333328	0.25-4	1
S. haemolyticus			
Meth-S (10)	BI 397	≤0.03-0.25	0.13
	Teicoplanin	1-32	32
	Vancomycin	1-4	2
	LY-333328	0.06-1	1
Meth-R (12)	BI 397	≤0.03-4	0.5
	Teicoplanin	2-128	32
	Vancomycin	0.5-8	4
	LY-333328	0.13-1	1
Other CoNS			
Meth-S (10)	BI 397	≤0.03-0.13	0.13
	Teicoplanin	0.13-4	2
	Vancomycin	0.5-2	2
	LY-333328	0.06-0.5	0.5
Meth-R (12)	BI 397	≤0.03-0.13	0.13
	Teicoplanin	0.06-32	2
	Vancomycin	0.5-4	2
	LY-33328	≤0.03-0.5	0.25

Table II: In vitro *activity (MIC, µg/ml) against streptococci and enterococci.*

Organism (No. strains)	Antibiotic	MIC range	(MIC_{50}) MIC_{90}
S. pyogenes			
(5)	BI 397	≤0.002-0.06	(≤0.002)
	Teicoplanin	0.008-0.06	(0.016)
	Vancomycin	0.5-0.5	(0.5)
	LY-333328	0.016-0.13	(0.016)
S. pneumoniae			
Pen-S (12)	BI 397	0.016-0.13	0.06
	Teicoplanin	0.008-0.06	0.06
	Vancomycin	0.13-0.5	0.5
	LY-333328	≤0.002-0.06	0.008
Pen-R (5)	BI 397	0.008-0.13	(0.03)
	Teicoplanin	0.016-0.13	(0.03)
	Vancomycin	0.25-2	(0.25)
	LY-333328	≤0.002-0.06	(≤0.002)
Enterococcus spp.			
Van-S (6)	BI 397	0.06-0.13	(0.13)
	Teicoplanin	0.13-0.5	(0.13)
	Vancomycin	0.25-4	(0.5)
	LY-333328	0.06-0.25	(0.06)
VanB (10)	BI 397	0.02-2	1
	Teicoplanin	0.13-8	2
	Vancomycin	8-128	128
	LY-333328	≤0.03-0.13	0.13
VanA (21)	BI 397	0.5->128	>128 (32)
	Teicoplanin	64->128	>128 (>128)
	Vancomycin	>128	>128 (>128)
	LY-333328	0.06-1	1 (0.25)

Antibacterial Activity (10,11)

Like other known glycopeptides, BI 397 inhibits the biosynthesis of the bacterial cell wall by binding to D-alanyl-D-alanine-terminating precursors of peptidoglycan.

In vitro *activity*

BI 397 is more active than other known glycopeptide antibiotics against several groups of Gram-positive bacteria. In particular, it has excellent activity against coagulase-positive and coagulase-negative staphylococci, including methicillin-resistant (Meth-R) strains of *S. aureus*, *S. epidermidis* and *S. haemolyticus*. It also has excellent activity against a number of streptococci, including strains of penicillin-resistant (Pen-R) *S. pneumoniae*, and against vancomycin-susceptible (Van-S) and vancomycin-resistant (Van-R) enterococcal isolates of the VanB phenotype. BI 397 is poorly active or inactive against most VanA enterococci that are highly resistant to vancomycin and teicoplanin but susceptible to the new glycopeptide LY-333328.

The *in vitro* activities of BI 397, teicoplanin, vancomycin and LY-333328 against *Staphylococcus* isolates, including methicillin-susceptible (Meth-S) and Meth-R strains, are shown in Table I. *In vitro* activities of the four compounds against streptococci and enterococci, including penicillin-susceptible (Pen-S) and Pen-R strains of *S. pneumoniae*, as well as Van-S, VanA and VanB strains of *Enterococcus* species, are shown in Table II.

Bactericidal activity

The bactericidal activity of BI 397 was tested against two different strains of *S. aureus* (L561 and L1524) and *S. epidermidis* (L537 and L1679). Strains L1524 and L537 were chosen because they were the isolates used in experimental endocarditis infections.

Time-kill experiments performed with *S. aureus* showed that BI 397 was bactericidal (>99.9% of microorganism killed within 24 h) against strain L1524 at a

Table III: In vivo *activity in experimental septicemia in immunocompetent mice.*

Organism	Antibiotic	MIC (μg/ml)	ED_{50} (mg/kg s.c.)
S. aureus Smith	BI 397	0.06	0.08
	Teicoplanin	0.5	0.20
	Vancomycin	0.5	1.12
S. pyogenes L49	BI 397	0.06	0.06
	Teicoplanin	0.03	0.18
	Vancomycin	0.25	Not tested
S. pneumoniae L44	BI 397	0.03	0.56
	Teicoplanin	0.06	0.40
	Vancomycin	0.5	0.79[1]

[1]BI 397 and teicoplanin were administered subcutaneously once after infection, while vancomycin was given twice (the first after infection, the second 5 h later).

Table IV: In vivo *activity in experimental septicemia in neutropenic mice.*

Organism	Antibiotic	MIC (μg/ml)	ED_{50} (mg/kg s.c.)
S. epidermidis L1480	BI 397	0.13	0.38
	Teicoplanin	4	10.7
	Vancomycin	2	7.1[1]
E. faecalis L1139	BI 397	0.06	1.53
	Teicoplanin	≤0.13	0.53
	Vancomycin	0.5	Not tested

[1]BI 397 and teicoplanin were administered subcutaneously once after infection, while vancomycin was given twice (the first after infection, the second 5 h later).

concentration of 8 μg/ml (ca. 64-fold the MIC value), while teicoplanin and vancomycin were poorly active. Against strain L561 (teicoplanin-resistant), only vancomycin showed bactericidal activity at 32 and 64 μg/ml (8- and 16-fold the MIC value).

In time-kill experiments performed with *S. epidermidis*, BI 397 was bactericidal at concentrations of 1 and 2 μg/ml (4- and 8-fold the MIC value) against strain L537 and at 4 μg/ml (8-fold the MIC value) against strain L1679. Teicoplanin and vancomycin had moderate or no bactericidal effect, except for vancomycin at a concentration of 16 μg/ml (8-fold the MIC value) against strain L537.

In vivo *efficacy*

The *in vivo* efficacy of BI 397 was determined in several animal models of infection, including experimental septicemia in immunocompetent and neutropenic mice, endocarditis in rats and pneumococcal pneumonia in immunocompetent and neutropenic rats. As shown in Table III, BI 397 was very active in protecting mice from Gram-positive septicemia. The ED_{50} values (doses at which 50% of the mice survived) ranged from 0.06-0.56 mg/kg for different bacterial species.

As shown in Table IV, BI 397 was clearly superior to teicoplanin and vancomycin against acute *S. epidermidis* septicemia in neutropenic mice, while it was slightly less effective than teicoplanin against *Enterococcus faecalis*.

In experimental endocarditis in rats, BI 397 was very effective in reducing heart bacterial loads in animals infected with methicillin-resistant *S. aureus* (Table V) or *S. epidermidis* (Table VI).

In MRSA LI524 endocarditis in rats, a once-daily regimen of 1.25 mg/kg i.v. of BI 397 was equivalent to twice-daily teicoplanin (20 mg/kg), and a once-daily regimen of 10 mg/kg i.v. of BI 397 was as effective as twice-daily vancomycin (100 mg/kg i.m.). The *S. aureus* infection was particularly difficult to eradicate in this experimental model.

In the *S. epidermidis* endocarditis experiment, BI 397 administered once daily (total dose of 7.5 mg/kg over 5 days) was at least as effective as twice-daily teicoplanin (total dose 220 mg/kg) and twice-daily vancomycin (total dose 1000 mg/kg). As reported in Table VI, all treatment regimens significantly reduced (p <0.05) heart bacterial load as compared to untreated controls. The efficacy of the three compounds was also reflected in the number of animals with sterile vegetations.

In a series of experiments of lobar pneumonia in rats caused by Pen-S or Pen-R *S. pneumoniae,* a single i.v. dose of BI 397 was very effective in reducing lung bacterial load compared to untreated controls.

Against Pen-S *S. pneumoniae* pneumonia in immunocompetent rats, all treatments were effective in significantly reducing lung bacterial load. However, a single dose of BI 397 (4 mg/kg) was more effective than procaine Pen-G (6 mg/kg) given i.m. twice-daily for 3 days. The lungs of all animals were sterilized by a single 10-mg/kg i.v. dose of BI 397; even at the lowest dose of 1.6 mg/kg, BI 397 was somewhat effective.

Table V: In vivo *activity in 5-day therapy of* S. aureus *L1524 endocarditis in rats.*

Regimen	MIC (μg/ml)	Dose (mg/kg)	No. of survivors vs. no. of animals	No. of sterile samples vs. no. of animals	Bacterial load (log_{10} CFU/g of heart ± SD)
Control			0/12	0/12	9.7 ± 0.3
BI 397	0.13	10 i.v. qd	11/11	4/11	3.7 ± 2.2[a,b]
BI 397		1.25 i.v. qd	8/12	0/12	6.8 ± 1.6[a]
Teicoplanin	0.25	20 i.v. bid	8/10	2/10	6.1 ± 2.2[a]
Vancomycin	1.00	100 i.m. bid	8/10	2/10	4.1 ± 2.2[a,b]
Pretreatment			6/6	0/6	7.3 ± 1.0

[a]p < 0.05 compared with control. [b]p < 0.05 compared with pretreatment.

Table VI: In vivo *activity in 5-day therapy of* S. epidermidis *L537 endocarditis in rats.*

Regimen	MIC (μg/ml)	Dose (mg/kg)	No. of survivors vs. no. of animals	No. of sterile samples vs. no. of animals	Bacterial load (log_{10} CFU/g of heart ± SD)
Control			4/11	0/11	6.3 ± 0.7
BI 397	0.13	2.5 i.v. qd	11/13	5/11	3.5 ± 1.8[a]
BI 397		1.25 i.v. qd	7/11	6/11	2.8 ± 1.2[a]
Teicoplanin	8.00	20 i.v. bid	10/12	6/12	3.4 ± 1.7[a]
Vancomycin	2.00	100 i.m. bid	10/11	9/11	2.4 ± 1.2[a]
Pretreatment			6/6	0/6	5.6 ± 0.5

[a] $p < 0.05$ compared with control.

Against Pen-R *S. pneumoniae* pneumonia in immunocompetent rats, the lowest single dose (1.6 mg/kg) of BI 397 was significantly more active than 48 mg/kg i.m.of procaine Pen-G administered twice-daily for 3 days.

Against Pen-R *S. pneumoniae* pneumonia in neutropenic rats, all single doses of BI 397 (4, 10 and 25 mg/kg) were effective both in reducing lung bacterial load (*vs.* pretreatment load) and in preventing mortality (100% survived animals). The result is especially important considering the characteristics of this difficult-to-treat infection in immunocompromised animals, as shown by the high (100%) mortality among controls and rats treated with procaine Pen-G (6 x 60 mg/kg).

Dose-response effects were observed with BI 397 in all of the above experiments.

Pharmacokineties and Pharmacodynamics (10, 11)

The pharmacokinetics of intravenous BI 397 were investigated in rats, rabbits and dogs. In all species, BI 397 showed high and prolonged plasma levels, indicating that once-daily administration may be feasible in humans.

In rats and rabbits, after a single dose of 20 mg/kg, 24-h plasma levels of BI 397 (equal to or more than 10 mg/l) were higher than MIC values for all tested Gram-positive pathogens except most VanA enterococci. Plasma concentrations (equal to or more than 5 mg/l) at least 2-fold higher than MIC values for susceptible organisms were also found in dogs 48 h after administration of a 5-mg/kg dose of the drug.

Tissue distribution studies performed in rats using [^{3}H]-radiolabeled BI 397 showed that radioactivity was distributed into almost all organs and tissues. At present no information is available on the metabolic fate of BI 397 in rats. Studies to identify possible metabolites will be carried out during phase I clinical trials in humans.

Kinetics in rats

Following administration of BI 397 (20 mg/kg i.v.), high and prolonged plasma concentrations were found in rats: plasma concentration averaged 333.1 ± 32.1 mg/l 3 min after administration and 0.43 ± 0.03 mg/l at 120 h, after which the compound was no longer detectable. A two-compartment model with elimination from the central compartment was fitted to drug concentration-time profiles. The half-lives of the initial and terminal disposition phases were 0.48 and 14 h. The fraction of the dose eliminated in the second phase (area under the concentration-time curve in the second phase) was 94%. The initial distribution volume was 0.06 l/kg, and the steady-state volume of distribution was 0.13 l/kg. Total clearance from plasma was 8.8 ml/h/kg. The plasma concentration-time profile of BI 397 after administration of 20 mg/kg i.v. in rats infected with *S. aureus* or *S. epidermidis* endocarditis was similar to that observed in healthy rats. Experimental data obtained for regimens of 10 and 2.5 mg/kg/day for 5 days show linear pharmacokinetics. As expected from its long half-life, BI 397 produced plasma concentrations higher than teicoplanin at lower doses and longer treatment intervals.

Kinetics in rabbits

After administration of BI 397 (20 mg/kg i.v.), high and prolonged plasma concentrations were found in rabbits: initial plasma concentration averaged 372.1 ± 46.7 mg/l 3 min after administration and declined rapidly in the first 2-4 h, followed by a slower elimination phase with a half-life of 15.68 h; clearance was very low. Generally, the larger the animal the slower the pharmacokinetics of a drug, which appears to be the case for BI 397. However, a comparison between plasma concentration-time profiles in rabbits and rats, normalized for dose and body weights, shows that disposition of BI 397 in rabbits is different from that in rats, with a more rapid decline in plasma occurring in rabbits.

Kinetics in dogs

Following administration of BI 397 (5 mg/kg i.v.), high and prolonged plasma concentrations were found in dogs: plasma concentration averaged 66.3 ± 18.5 mg/l 5 min after administration, with a plateau between 1 and 4 h (30-40 mg/l), which suggests the presence of an

enterohepatic recycle. Elimination half-life was 24.8 ± 5.7 h, total AUC was 1192.6 ± 209.7 h.mg/l, total body clearance was 3.8 ± 0.7 ml/h/kg – much lower than the glomerular filtration rate (about 100 ml/h/kg) – and the volume of distribution was 136.3 ± 42.8 ml/kg, corresponding to about 14 ± 4% of body volume, *i.e.*, similar to extracellular water volume.

Tissue distribution in rats

Whole-body autoradiography studies in rats after single i.v. administration of 20 mg/kg of [^{3}H]-radiolabeled BI 397 indicated that radioactivity was distributed into almost all organs and tissues. This might be due to metabolism and does not necessarily demonstrate distribution of unchanged drug. However, the fact that high levels were observed in liver and kidneys up to the last sampling time of 120 h posttreatment suggests that the processes of metabolism and subsequent excretion are slow. Furthermore, concentrations in lymph nodes throughout the body and in testicles demonstrated targeting to these organs.

Toxicology (12)

Safety pharmacology and mutagenicity studies

No statistically significant effects on the autonomic nervous system have been observed in rats. In particular, BI 397 administered to conscious rats at doses of 5, 10 and 20 mg/kg i.v. did not induce any statistically significant effects on mean arterial pressure, was devoid of alpha-adrenolytic properties and did not interact with 5-HT_3 or 5-HT_{1d} receptor-mediated cardiovascular effects induced by serotonin. These results suggest that BI 397 has no sympatholitic, β-blocking, sympathomimetic, parasympatholytic, parasympathomimetic or ganglioplegic properties or antagonistic properties on serotonin-induced cardiovascular effects in conscious rats.

No statistically significant changes in blood pressure and other cardiovascular parameters were observed in anesthetized dogs after administration of escalating doses of 5, 10 and 20 mg/kg i.v. of BI 397 (total cumulative dose of 35 mg/kg/dog).

Further safety studies showed that BI 397 had no mutagenic potential, was not cytotoxic and had no effect on platelet aggregation or bleeding time. Gene mutation studies in V79 Chinese hamster lung cells showed that were no significant effects with or without metabolic activation. Results of an *in vitro* cytogenic assay in Chinese hamster ovary cells demonstrated that BI 397 did not induce a statistically significant increase in cells with chromosome aberrations up to a concentration of 2000 μg/ml, both in the absence and presence of metabolic activation. Evaluation of the drug's effect on platelet aggregation on rabbit platelets showed no statistically significant effect on arachidonic acid-induced aggregation and a dose-dependent effect on collagen-induced aggregation. BI 397 administered to rats at doses of 5, 10 and 20 mg/kg i.v. had no statistically significant effect on bleeding time.

Other tolerability tests, such as the Irwin behavioral test in mice, showed that BI 397 (5, 10 and 20 mg/kg i.v.) had no statistically significant effect on body temperature and no neurobehavioral, neurovegetative, psycotropic or neurotoxic effects. Furthermore, BI 397 was not skin-sensitizing in guinea pigs, was nonirritant for the eye or skin in rabbits and was safe in mice and rats up to the maximum tested dose of 2000 mg/kg p.o.

Systemic toxicity

In 1-month studies in dogs, BI 397 (5 and 10 mg/kg/day i.v. bolus) did not induce significant changes indicating systemic involvement. At a dose of 20 mg/kg/day, minor reversible hepatic and renal changes were observed in the blood chemistry and urinalysis determinations, along with minimal inflammatory changes in the liver and slight tubular necrosis in the kidneys of 1 male dog. A dose of 40 mg/kg/day induced marked toxic changes in the liver and kidneys, including necrosis of hepatocytes and renal tubules, in the lymphoid tissues and bone marrow. Most of these effects were reversible after a 1-month recovery period.

In 1-month studies in rats, BI 397 was well tolerated at doses of 5 and 10 mg/kg/day i.v. bolus since only negligible increases in serum GOT activity and blood in urine, as well as negligible body weight gain retardation were observed, compared to controls. At a 10 mg/kg/day dose, slight pigment deposition in the renal tubules and slight vacuolation in the exocrine pancreas were the only morphological changes seen at histology. These morphological signs were more pronounced and dose-related with the 20- and 40- mg/kg doses, where renal pigmentation was associated with degenerative changes in the tubular epithelium, and vacuolation of histiocytes in the liver and lymph nodes was also observed. All morphological changes associated with the 40 mg/kg dose were reversible after 1-month recovery period.

Manufacturer

Biosearch Italia S.p.A. (IT). Versicor, Inc. (US) has acquired North American marketing rights (13).

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