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In Vitro Activity of LY146032 against Staphylococci, Streptococci, and Enterococci

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The in vitro activities of LY146032 and seven comparative antimicrobial agents against 14 species of staphylococci, streptococci, and enterococci were studied. MICs of LY146032 were ≤0.5 μg/ml for all staphylococci, including oxacillin-resistant strains; ≤0.25 μg/ml for all streptococci (except viridans group streptococci); and ≤4 μg/ml for all viridans group streptococci and enterococci. MICs were minimally affected by variations in inoculum size, and LY146032 was bactericidal against all species tested.

LY146032 is a new antibiotic that was synthesized from A21978C, a complex of acidic lipopeptide antibiotics containing the same peptide core (F. T. Counter, P. J. Baker, L. D. Boeck, M. DeBono, P. W. Ensminger, R. L. Hamill, V. M. Krupinski, R. M. Molloy, and J. L. Ott, Program Abstr. 24th Intersci. Conf. Antimicrob. Agents Chemother., abstr. no. 1078, 1984). It inhibits peptidoglycan synthesis in gram-positive bacteria (1). In this study, the in vitro activities of LY146032 and seven comparative antimicrobial agents against staphylococci, streptococci, and enterococci were studied.

The organisms studied included 316 strains, representing 14 species of staphylococci, streptococci, and enterococci. LY146032, cephalothin, and vancomycin were obtained from Lilly Research Laboratories, Indianapolis, Ind.; oxacillin from Bristol Laboratories, Syracuse, N.Y.; ampicillin from Wyeth Laboratories, Philadelphia, Pa.; imipenem from Merck Sharp & Dohme, West Point, Pa.; ciprofloxacin from Miles Pharmaceuticals, West Haven, Conn.; rifampin from Merrell Dow Pharmaceuticals, Cincinnati, Ohio; and gentamicin from Pfizer Inc., New York, N.Y. Laboratory standards were diluted according to the recommendations of the manufacturer and dispensed into microdilution plates with a dispensing machine (MIC-2000 Plus; Dynatech Laboratories, Alexandria, Va.) in $\log_2$ dilution steps within the range of 0.016 to 32 μg/ml. Plates were stored at −70°C until used.

MICs were determined by a standardized microdilution method (2) in 0.1-ml volumes of cation-supplemented Mueller-Hinton broth (Difco Laboratories, Detroit, Mich.). For supplementation, 45 mg of calcium per liter and 25 mg of magnesium per liter were added to Mueller-Hinton broth; physiologic concentrations were confirmed by testing the activity of gentamicin against *Pseudomonas aeruginosa* ATCC 27853 (2). For *Streptococcus pyogenes* and *Streptococcus pneumoniae*, the medium was supplemented with 1% heat-inactivated horse serum. Microdilution plates were inoculated with disposable inoculators (Dynatech) which deliver 10 μl per well so that the inoculum was 5×10^5 to 1×10^6 CFU/ml. To distinguish oxacillin-susceptible from oxacillin-resistant staphylococci, a modification (4) of the microdilution test was used; criteria were that MICs ≤2 μg/ml were considered susceptible and MICs ≥16 μg/ml were considered resistant for *Staphylococcus aureus* and *Staphylococcus haemolyticus*, and MICs ≤0.25 μg/ml were considered susceptible and ≥16 μg/ml were considered resistant for *Staphylococcus epidermidis*.

The MICs of the study drugs for the 316 strains are shown in Table 1. MICs of LY146032 were ≤0.5 μg/ml for all staphylococci, including oxacillin-resistant strains; ≤0.25 μg/ml for all streptococci (except viridans group streptococci); and ≤4 μg/ml for all viridans group streptococci and enterococci. There was no correlation of relative susceptibility or resistance between LY146032 and other drugs against any species.

For 85 of the study strains (5 strains each of the organisms listed in Table 1), the effects of inoculum size variation on MICs were determined by simultaneously testing the standard inoculum and inocula 10-fold higher and 10-fold lower. Variations of 100-fold had little effect (≤2 dilution steps) with any drug, with the following exceptions: ampicillin against penicillinase-producing and oxacillin-resistant staphylococci; oxacillin, cephalothin, and imipenem against oxacillin-resistant staphylococci; gentamicin against viridans group streptococci; and rifampin against occasional enterococci.

To determine MBCs for the same 85 strains, the wells from MIC plates containing an initial inoculum of 5×10^5 to 1×10^6 CFU/ml were subcultured (10 μl) to 0.1-ml volumes of antimicrobial-free broth to detect a ≥99.9% reduction in CFU/ml (3). With LY146032, MBCs were ≤2 dilution steps higher than respective MICs for all species. With other drugs, MBCs were usually ≤2 dilution steps higher than respective MICs for susceptible strains of staphylococci and streptococci, but enterococci were often resistant or had higher MBC/MIC ratios.

The results of this and another (1) study indicate that LY146032 has a favorable profile of antibacterial activity against staphylococci, streptococci, and enterococci.

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TABLE 1. In vitro activities of LY146032 and comparative antimicrobial agents against staphylococci, streptococci, and enterococci

Organism (no.)	Drug	MIC (μg/ml)[a]		
		Range	50%	90%
Staphylococcus aureus, oxacillin susceptible (20)	LY146032	0.13–0.25	0.25	0.25
	Cephalothin	0.5–1	0.5	1
	Imipenem	0.03–0.06	0.03	0.03
	Vancomycin	0.5–1	0.5	1
	Ciprofloxacin	0.13–1	0.25	0.5
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin[b]	0.13–32	0.25	0.25
Staphylococcus aureus, oxacillin resistant (20)	LY146032	0.13–0.5	0.25	0.25
	Cephalothin[c]	1–>32	8	32
	Imipenem[c]	0.13–16	0.25	2
	Vancomycin	0.25–2	0.5	1
	Ciprofloxacin	0.25–1	0.5	0.5
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin[b]	0.25–>32	0.5	32
Staphylococcus epidermidis, oxacillin susceptible (20)	LY146032	0.13–0.25	0.25	0.25
	Cephalothin	0.13–0.25	0.25	0.25
	Imipenem	≤0.016–0.03	0.03	0.03
	Vancomycin	1–2	1	1
	Ciprofloxacin	0.13–0.5	0.25	0.25
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin	≤0.03–0.06	≤0.03	0.06
Staphylococcus epidermidis, oxacillin resistant (20)	LY146032	0.13–0.5	0.25	0.25
	Cephalothin[c]	0.25–2	1	1
	Imipenem[c]	0.06–32	0.5	4
	Vancomycin	0.5–2	2	2
	Ciprofloxacin	0.25–0.5	0.25	0.5
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin[b]	≤0.03–32	4	16
Staphylococcus haemolyticus, oxacillin susceptible (20)	LY146032	0.13–0.25	0.25	0.25
	Cephalothin	0.25–0.5	0.25	0.5
	Imipenem	≤0.016–0.06	0.03	0.06
	Vancomycin	0.5–1	1	1
	Ciprofloxacin	0.13–0.25	0.25	0.25
	Rifampin	≤0.016–0.03	≤0.016	≤0.016
	Gentamicin	≤0.03–0.06	≤0.03	≤0.03
Staphylococcus haemolyticus, oxacillin resistant (20)	LY146032	0.25–0.5	0.25	0.5
	Cephalothin[c]	2–>32	>32	>32
	Imipenem[c]	0.13–>32	>32	>32
	Vancomycin	1–4	2	2
	Ciprofloxacin	0.25	0.25	0.25
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin[b]	≤0.03–>32	16	32
Staphylococcus saprophyticus (20)	LY146032	0.06–0.5	0.25	0.5
	Ampicillin	0.13–0.5	0.25	0.5
	Cephalothin	0.25–2	1	1
	Imipenem	0.03–0.13	0.06	0.06
	Vancomycin	0.5–4	1	2
	Ciprofloxacin	0.25–0.5	0.5	0.5
	Rifampin	≤0.016–0.03	0.03	0.03
	Gentamicin	≤0.03	≤0.03	≤0.03
Staphylococcus hominis (20)	LY146032	0.13–0.5	0.25	0.25
	Cephalothin	0.13–0.5	0.13	0.5
	Imipenem	≤0.016–0.06	0.03	0.06
	Vancomycin	0.5–2	1	1
	Ciprofloxacin	0.13–0.5	0.13	0.25
	Rifampin	≤0.016	≤0.016	≤0.016
	Gentamicin	≤0.03–0.13	≤0.03	0.06
Streptococcus pyogenes (20)	LY146032	0.03–0.13	0.06	0.06
	Ampicillin	≤0.016–0.03	0.03	0.03
	Cephalothin	0.03–0.13	0.13	0.13
	Imipenem	≤0.016	≤0.016	≤0.016
	Vancomycin	0.25–0.5	0.25	0.5
	Ciprofloxacin	0.25–2	0.5	1
	Rifampin	≤0.016–0.13	0.06	0.06
	Gentamicin	4–16	16	16
Streptococcus agalactiae (20)	LY146032	0.06–0.13	0.13	0.13
	Ampicillin	≤0.016–0.06	0.06	0.06

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TABLE 1—*Continued*

Organism (no.)	Drug	MIC (μg/ml)[a]		
		Range	50%	90%
	Cephalothin	0.06–0.13	0.13	0.13
	Imipenem	≤0.016	≤0.016	≤0.016
	Vancomycin	0.25–0.5	0.5	0.5
	Ciprofloxacin	0.5–1	0.5	1
	Rifampin	0.13–0.25	0.25	0.25
	Gentamicin	2–4	2	4
Streptococcus pneumoniae (20)	LY146032	0.13–0.25	0.13	0.13
	Ampicillin	≤0.016–0.03	≤0.016	0.03
	Cephalothin	0.06–0.13	0.13	0.13
	Imipenem	≤0.016	≤0.016	≤0.016
	Vancomycin	0.25–0.5	0.25	0.25
	Ciprofloxacin	1–2	1	2
	Rifampin	≤0.016–0.13	0.03	0.06
	Gentamicin	8–16	16	16
Viridans group streptococci (20)	LY146032	≤0.016–2	0.25	0.5
	Ampicillin	≤0.016–1	0.03	0.5
	Cephalothin	≤0.016–16	0.13	4
	Imipenem	≤0.016–1	0.03	0.5
	Vancomycin	0.06–4	0.5	0.5
	Ciprofloxacin	0.03–8	1	2
	Rifampin[b]	≤0.016–8	0.03	0.06
	Gentamicin	0.13–32	4	16
Streptococcus bovis (12)	LY146032	≤0.016–0.13	0.06	0.06
	Ampicillin	0.03–0.13	0.06	0.13
	Cephalothin	0.06–0.5	0.25	0.25
	Imipenem	≤0.016	≤0.016	≤0.016
	Vancomycin	0.25–0.5	0.5	0.5
	Ciprofloxacin	0.5–4	2	4
	Rifampin	0.06–0.25	0.13	0.25
	Gentamicin	2–16	4	8
Enterococcus faecalis (20)	LY146032	0.25–2	0.5	0.5
	Ampicillin	1–4	2	2
	Cephalothin	32–>32	32	32
	Imipenem	1–4	1	2
	Vancomycin	0.5–4	1	4
	Ciprofloxacin	0.5–2	1	1
	Rifampin	0.25–16	2	8
	Gentamicin	4–16	8	8
Enterococcus faecium (20)	LY146032	0.5–4	1	2
	Ampicillin	0.13–8	2	8
	Cephalothin	4–>32	>32	>32
	Imipenem	0.25–32	4	32
	Vancomycin	0.5–4	1	4
	Ciprofloxacin	0.5–8	2	4
	Rifampin[b]	≤0.016–32	2	32
	Gentamicin	2–16	4	8
Enterococcus durans (12)	LY146032	0.5–4	1	4
	Ampicillin	2–8	4	8
	Cephalothin	16–>32	>32	>32
	Imipenem	2–>32	8	32
	Vancomycin	0.5–4	1	2
	Ciprofloxacin	1–16	2	8
	Rifampin[b]	≤0.016–32	8	32
	Gentamicin	4–32	8	16
Enterococcus avium (12)	LY146032	0.5–1	0.5	1
	Ampicillin	1–4	1	2
	Cephalothin	8–32	8	16
	Imipenem	1–8	1	2
	Vancomycin	0.5–1	1	1
	Ciprofloxacin	1–2	1	2
	Rifampin	1–4	2	4
	Gentamicin	1–2	2	2

[a] The values of 50 and 90% MICs are those MICs required to inhibit 50% and 90% of isolates, respectively.
[b] Bimodal distribution of MICs.
[c] Cephalothin and imipenem MICs for oxacillin-resistant staphylococci may not be relevant; all strains may be resistant based on the heteroresistant phenotype.

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