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Report from the 2005 Zyvox® Annual Appraisal of Potency and Spectrum (ZAAPS)

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ABSTRACT

Background: Linezolid (LZD) activity among Gram-positive pathogens on 5 continents (16 countries) was determined using a central laboratory and reference CLSI methods. The worldwide ZAAPS program has accumulated 4 years of LZD surveillance results directed at serious infections caused by MRSA and other resistant (R) organisms (VRE, multidrug-resistant [MDR] *S. pneumoniae* [SPN]).

Methods: Over 4,000 Gram-positive strains were collected in 2005 from 39 medical centers located in North America (5 sites; Canada [CAN] only), South America (10), Europe (14), and the Asia-Pacific (12). Sites within each nation contributed isolates toward a total of 200/nation (except CAN; 1,000 isolates from 5 sites) for reference broth microdilution MIC susceptibility (S) testing. Major organism groups tested were: *S. aureus* (SA; 1,416), coagulase-negative staphylococci (CoNS; 634), enterococci (718), SPN (853), viridans group streptococci (218) and ß-haemolytic streptococci (BHS; 370). Thirteen to 15 comparator agents were tested and current CLSI interpretative criteria were applied.

Results: No LZD-R strains were detected from the monitored countries, consistent with 2002-2004 results. Prior ZAAPS reports observed LZD-R strains only in the USA (0.1%; 2002-2003) and one referral isolate in 2004 from Greece. LZD remained highly active against all SA as shown in the table.

Table: LZD versus SA isolates.

Region (nations/sites/strains)	MRSA % (range)	LZD $MIC_{50/90}$ µg/ml (% S)
Canada (1/5/267)	16.9	1/2 (100.0)
South America (4/10/355)	46.0 (31.6 – 61.1)	2/2 (100.0)
Europe (6/14/465)	23.4 (3.3 – 41.7)	1/2 (100.0)
Asia-Pacific (5/12/329)	47.8 (15.0 – 78.8)	2/2 (100.0)

VRE rates ranged from a high in Korea (33%) to nil in 7 other nations. The overall erythromycin (ERY) and penicillin (PEN) R rates among SPN were 35 and 23%, respectively. SPN ERY-R rates were very high in the Asia-Pacific region (Australia 22% to Taiwan 94%). However, LZD remained highly active with $MIC_{50/90}$ values of 1/1 µg/ml. BHS remained S to PEN but monitored nations averaged 16% ERY-R with the highest rate in Taiwan (48%).

Conclusions: LZD remains highly active against all Gram-positive strains sampled from the 16 nations (not USA), including MRSA strains. As MRSA, VRE, and MDR Gram-positive organisms increase in prevalence, continued surveillance of LZD is a prudent action as this oxazolidinone becomes more widely prescribed worldwide.

INTRODUCTION

The Zyvox Annual Appraisal of Potency and Spectrum (ZAAPS) Program has completed its fourth year (2005) of resistance surveillance for linezolid, the first oxazolidinone class agent to be licensed for use in clinical practice. Linezolid has been used primarily to treat multidrug-resistant Gram-positive pathogens in complicated skin and soft tissue infections (cSSTI) and nosocomial pneumonias, after its release in 2000.

The linezolid mechanism of action has been described as selective binding to the 50S ribosomal subunit of the 23S rRNA molecule with resultant inhibition of protein synthesis. Among the rare cases of linezolid resistance reported to date in staphylococci and enterococci, G2576U or T2500A target site mutations have been the dominant resistance mechanism.

Linezolid has emerged as a viable, therapeutic alternative for infections caused by Gram-positive organisms that are resistant to conventional drugs, such as methicillin-resistant *Staphylococcus aureus* (MRSA), drug-resistant *Streptococcus pneumoniae* (DRSP) and vancomycin-resistant enterococci (VRE). Therefore, it is important to continuously monitor the potency and potential for emerging resistance to linezolid as the use of this agent increases, in volume and in geographic distribution.

MATERIALS AND METHODS

Organism collection

A total of 4,209 isolates were forwarded to the central monitoring site (JMI Laboratories, North Liberty, IA, USA) from 16 different nations for the 2005 ZAAPS Program. Each participating site (39 total) or country forwarded a target total of 200 consecutive, non-duplicate patient isolates from infections of the bloodstream, respiratory tract, urinary tract, or wounds/skin and soft tissue. The countries/sites that participated in 2005 included: Latin America (4 nations/10 sites), Europe (6 nations/14 sites), the Far East (5 nations/12 sites), and North America (Canada only/5 sites; see Table 1).

Susceptibility testing

Antimicrobial susceptibility testing was performed using validated, dry-form microdilution panels with cation-adjusted Mueller-Hinton broth (2-5% lysed horse blood added for testing streptococci) prepared by TREK Diagnostics (Cleveland, OH, USA). The categorical interpretations of MIC results produced are described in the Clinical and Laboratory Standards Institute (CLSI) document M100-S16. Quality control organism (*S. aureus* ATCC 29213, *E. faecalis* ATCC 29212, and *S. pneumoniae* ATCC 49619) results were within acceptable ranges as published by the CLSI.

All isolates were tested against antimicrobial agents active against Gram-positive organisms including: linezolid, chloramphenicol, ciprofloxacin, erythromycin, levofloxacin, penicillin, quinupristin/dalfopristin, rifampin, teicoplanin, and vancomycin. Other drugs tested against selected pathogen subgroups were: ampicillin, ceftriaxone, clindamycin, doxycycline, gentamicin (high-level resistance screen), piperacillin/tazobactam, streptomycin (high-level resistance screen only), tetracycline, and trimethoprim/sulfamethoxazole (Table 2).

RESULTS

- The MIC results for linezolid versus *S. aureus* showed 98.6% of values at 1 or 2 µg/ml. This did not change with oxacillin-resistant *S. aureus* as the $MIC_{50/90}$ remained at 1 and 2 µg/ml, respectively. None of the *S. aureus* isolates had a linezolid MIC above 2 µg/ml (Table 2).
- Linezolid MIC values were generally two-fold lower for CoNS when compared to *S. aureus* MIC results. The CoNS linezolid $MIC_{50/90}$ were both 1 µg/ml (range of 0.25-2 µg/ml), unchanged from 2004. Only isolates from Argentina and France demonstrated a two-fold higher MIC_{90} value at 2 µg/ml.
- The vast majority of enterococci tested (94.2%) were *E. faecalis* (502 strains) and *E. faecium* (174 strains) with a narrow linezolid MIC range of 0.25-2 µg/ml (Table 2). Over 90% of linezolid MIC results were either 1 or 2 µg/ml with no MIC values occurring at 4 µg/ml (intermediate category). Resistance to linezolid among enterococci remained absent in the monitored countries (2002-2005).
- The overall VRE rate for non-USA areas, including the intermediate category, was 5.7%; a 0.4% increase over 2004.
- The overall erythromycin resistance rate among *S. pneumoniae* isolates increased slightly from 22.6% in 2004 to 24.0% in 2005 (Table 2); but the penicillin resistance (MIC, ≥ 2 µg/ml) rate remained the same at 41.0%. Nation-specific penicillin resistance rates among pneumococcal strains greatly differed from a high in Korea (71.8%), Taiwan (69.7%) and Hong Kong (57.1%) to the lowest rates in Europe (1-5%, except 32.0% for France), Canada (7.9%), Argentina (8.3%) and Mexico (7.0%).
- Linezolid remained highly active against all streptococci (*S. pneumoniae*, beta-haemolytic streptococci and viridans group streptococci) with $MIC_{50/90}$ results at only 1 µg/ml. (Table 3).

Table 1. Distribution of identified organisms for the 2005 ZAAPS sample indexed by nation of origin (4,209 strains).

Nation (number of medical centers)	Number of strains						
	S. aureus	CoNS	Enterococci	*S. pneumoniae*	Vir. gr. streptococci	ß-streptococci	Total
Canada[a] (5)	267	199	202	127	96	101	992
Argentina (2)	92	47	20	12	3	23	197
Brazil (4)	78	61	50	31	3	17	240
Chile (2)	95	41	45	18	4	15	218
Mexico (2)	90	39	35	57	3	1	225
France (3)	65	28	30	50	7	30	210
Germany (2)	90	36	50	4	9	13	202
Italy (3)	79	30	30	60	1	10	210
Spain (2)	99	22	33	29	12	16	211
Sweden (1)	60	10	40	60	3	30	203
United Kingdom (1)	72	14	21	71	8	17	203
Australia (5)	100	40	24	80	32	42	318
Hong Kong (1)	26	0	9	35	2	9	81
Japan (2)	80	30	40	75	14	10	249
Korea (2)	71	18	50	78	6	9	232
Taiwan (2)	52	19	39	66	15	27	218
TOTAL (39)	1,416	634	718	853	218	370	4,209

a. Only nation where the target sample size was 200 organisms per medical center (200 x 5 sites = 1000 strains).

Table 2. Comparative activity of linezolid tested against 4,209 Gram-positive cocci from 16 nations in the ZAAPS Program (2005).

Organism (no. tested)/ antimicrobial agent	MIC (µg/ml) 50%	90%	Range	% by category[a] Susceptible/Resistant
S. aureus (1,416)				
Linezolid	1	2	0.25-2	100.0/-
Ceftriaxone	4	>32	0.5->32	67.5/25.9
Chloramphenicol	8	8	≤2->16	93.6/4.9
Ciprofloxacin	0.25	>4	0.06->4	66.0/33.1
Clindamycin	≤0.25	>2	≤0.25->2	74.2/25.4
Erythromycin	0.25	>8	≤0.06->8	58.7/36.7
Gentamicin	≤2	>8	≤2->8	80.3/18.4
Levofloxacin	≤0.5	>4	≤0.5->4	66.7/32.4
Oxacillin[b]	0.5	>2	0.25->2	65.8/34.2
Penicillin	8	>32	≤0.016->32	12.0/88.0
Quinupristin/dalfopristin	≤0.25	0.5	≤0.25->2	99.9/0.1
Rifampin	≤0.5	≤0.5	≤0.5->2	94.9/2.9
Teicoplanin	≤2	≤2	≤2-4	100.0/0.0
Tetracycline	≤2	>8	≤2->8	86.1/13.7
Trimethoprim/sulfamethoxazole	≤0.5	≤0.5	≤0.5->2	93.1/6.9
Vancomycin	1	1	0.25-2	100.0/0.0
CoNS (634)				
Linezolid	1	1	0.25-2	100.0/-
Ceftriaxone	8	>32	≤0.25->32	54.2/15.8
Chloramphenicol	4	>16	≤2->16	84.7/13.3
Ciprofloxacin	2	>4	≤0.03->4	49.8/47.8
Clindamycin	≤0.25	>2	≤0.25->2	61.8/36.9
Erythromycin	>8	>8	≤0.06->8	31.5/54.6
Gentamicin	≤2	>8	≤2->8	58.4/29.2
Levofloxacin	1	>4	0.06->4	50.7/43.6
Oxacillina	>2	>2	≤0.25->2	25.4/74.6
Penicillin	4	>32	≤0.016->32	12.9/87.1
Quinupristin/dalfopristin	≤0.25	0.5	≤0.25->2	99.2/0.3
Rifampin	≤0.25	>2	≤0.25->2	86.7/10.4
Teicoplanin	≤2	8	≤2->16	96.4/0.9
Tetracycline	≤2	>8	≤2->8	88.0/11.4
Trimethoprim/sulfamethoxazole	≤0.5	>2	≤0.5->2	59.5/40.5
Vancomycin	1	2	0.25-4	100.0/0.0
Enterococcus spp. (718)				
Linezolid	1	2	0.25-2	100.0/0.0
Ampicillin	≤1	>16	≤1->16	77.6/22.4
Chloramphenicol	8	>16	≤2->16	79.3/19.0
Ciprofloxacin	1	>4	0.25->4	51.5/44.6
Doxycycline	8	>8	≤1->8	46.1/15.2
Erythromycin	>8	>8	≤0.06->8	11.3/51.1
Gentamicin (HL)[b]	≤500	>1000	≤500->1000	66.4/33.6
Levofloxacin	2	>4	≤0.5->4	55.9/42.3
Penicillin	4	>32	0.12->32	75.1/24.9
Piperacillin/tazobactam	4	>64	1->64	77.6/22.4
Quinupristin/dalfopristin	>2	>2	≤0.25->2	18.2/74.2
Rifampin	2	>2	≤0.25->2	29.5/48.1
Streptomycin (HL)[b]	≤1000	>2000	≤1000->2000	67.7/32.3
Teicoplanin	≤2	≤2	≤2->16	96.5/3.2
Vancomycin	1	2	0.5->16	94.3/4.3
S. pneumoniae (853)				
Linezolid	1	1	≤0.06-2	100.0/-
Amoxicillin/clavulanic acid	≤1	2	≤1-16	91.0/5.0
Ceftriaxone	≤0.25	1	≤0.25-8	93.3/0.7
Clindamycin	≤0.25	>2	≤0.25->2	72.6/26.8
Erythromycin	≤0.25	>32	≤0.25->32	58.6/41.0
Levofloxacin	1	1	≤0.5->4	98.7/1.2
Penicillin	≤0.03	4	≤0.03-8	61.5/24.0
Quinupristin/dalfopristin	0.5	1	≤0.25-1	100.0/0.0
Rifampin	≤0.5	≤0.5	≤0.5->2	99.1/0.9
Tetracycline	≤2	>8	≤2->8	63.3/36.0
Trimethoprim/sulfamethoxazole	≤0.5	4	≤0.5->4	61.7/26.9
Vancomycin	0.25	0.5	≤0.12-1	100.0/-
Viridans group streptococci (218)				
Linezolid	0.5	1	0.12-2	100.0/-
Amoxicillin/clavulanic acid	≤1	≤1	≤1-16	76.1/3.2
Ceftriaxone	≤0.25	0.5	≤0.25-8	95.9/2.3
Clindamycin	≤0.25	≤0.25	≤0.25->8	90.4/9.2
Erythromycin	≤0.25	4	≤0.25->32	58.7/37.2
Levofloxacin	1	1	0.25->4	96.8/2.8
Penicillin	0.03	1	≤0.016-8	76.1/3.2
Quinupristin/dalfopristin	0.5	1	≤0.25-2	97.7/0.0
Tetracycline	≤2	>8	≤2->8	58.7/39.9
Vancomycin	0.5	1	≤0.12-1	100.0/-
ß-haemolytic streptococci (370)				
Linezolid	1	1	0.25-2	100.0/-
Amoxicillin/clavulanic acid	≤1	≤1	≤1	100.0/-
Ceftriaxone	≤0.25	≤0.25	≤0.25-0.5	100.0/-
Clindamycin	≤0.25	≤0.25	≤0.25->8	93.5/6.2
Erythromycin	≤0.06	>2	≤0.06->2	83.5/16.5
Levofloxacin	≤0.5	1	≤0.5->4	98.4/1.6
Penicillin	≤0.016	0.06	≤0.016-0.12	100.0/-
Quinupristin/dalfopristin	≤0.25	≤0.25	≤0.25-1	100.0/0.0
Tetracycline	≤2	>8	≤2->8	51.6/47.3
Vancomycin	0.5	0.5	≤0.12-1	100.0/-

a. Criteria as published by the CLSI ß-lactam susceptibility for staphylococci should be directed by the oxacillin test results; "-" = no interpretive criteria have been published by the CLSI; HL= high-level resistance screen.

Table 3. Cumulative % inhibited results at each linezolid MIC when testing six different groups of Gram-positive cocci isolated on five continents (ZAAPS, 2005)[a].

Organism group (no. tested)	Cum. % inhibited at linezolid MIC (µg/ml) ≤0.12	0.25	0.5	1	2	4
Viridans group streptococci (218)	1.8	6.4	52.7	99.5	100.0[b]	-
S. pneumoniae (853)	0.5	3.2	24.3	98.5	100.0[b]	-
ß-haemolytic streptococci (370)	0.0	0.3	1.1	29.5	99.8	100.0[b]
CoNS (634)	0.0	0.9	23.8	97.0	100.0	-[b]
Enterococci (718)	0.0	0.3	5.3	68.5	100.0[b]	-
S. aureus (1,416)	0.0	0.1	1.3	55.9	100.0	-[b]

a. Organism groups were ranked in decreasing order of susceptibility to the oxazolidinone.
b. Susceptible CLSI (M100-S16) breakpoint concentrations.

CONCLUSIONS

- No linezolid resistance was discovered in the 2005 non-USA ZAAPS Program and there was no trend toward greater linezolid resistance (MIC creep), in fact, the modal MIC (1 µg/ml) was lower for *S. aureus* compared to results from the 2002-2004 period.
- Linezolid continues to show potent activity in all six organism subsets analyzed (Table 2). Of particular interest, linezolid exhibited excellent activity against DRSP, *S. pyogenes*, *S. agalactiae*, and viridans group streptococci.
- False resistance may be reported by laboratories due to the difficulty in reading linezolid broth microdilution MIC endpoints. A trailing effect is frequently reported in enterococci and may require a second method to confirm resistance.
- As the use of linezolid steadily increases in the USA and the rest of the world, associated evolution toward resistance must be monitored closely by organism, nation, year and mechanism type, when they occur.

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