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Comparative In Vitro Susceptibilities of 396 Unusual Anaerobic Strains to Tigecycline and Eight Other Antimicrobial Agents▽

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Tigecycline was tested against 396 strains of lesser-known anaerobic species encountered in human infections. It was active against all gram-positive strains and 228 of 232 gram-negative anaerobes at ≤1 μg/ml. One strain of *Prevotella oralis* was nonsusceptible at 8 μg/ml.

Tigecycline, a new glycylcycline antimicrobial agent, has recently been approved by the FDA for the treatment of complicated intra-abdominal infections (IAIs) and skin and soft tissue infections (SSTIs). It has shown activity against a variety of aerobic and anaerobic bacteria (1, 2–5, 7, 8, 11, 14). While premarket testing of new antimicrobial compounds is often extensive, it focuses on typical anaerobic pathogens such as the members of the *Bacteroides fragilis* group, *Clostridium perfringens*, and *Clostridium difficile* (1, 2, 3, 4), yet the bacteriology of IAIs and SSTIs is complex. In IAIs, for example, *B. fragilis* and *B. fragilis* group species account for 44% of anaerobic isolates, whereas less commonly clinically identified anaerobes account for the remaining 56% (6). Clostridia account for an additional 17% of IAI isolates, with *C. clostridioforme* and *C. innocuum* comprising 30% and 23%, respectively, of *Clostridium* species. Complicated SSTIs, such as diabetic foot and animal and human bite wound infections are also polymicrobial, involving multiple anaerobic species, including peptostreptococci, *Porphyromonas* spp., and fusobacteria (6, 10, 15). These organisms are recognized pathogens yet are often not isolated or identified by routine clinical laboratories due to cost and time considerations or lack of technical expertise. Since most clinical laboratories also do not perform susceptibility testing on anaerobes, most therapy is empirical, with clinicians relying on published studies to help guide therapy choices. We therefore determined the activity of tigecycline against a large variety of these less common anaerobic species, most of which have not been tested in previously reported studies.

The strains used in this study were previously isolated from human clinical specimens from a variety of sources and were identified by standard criteria or, for atypical strains, by 16S RNA gene sequencing (9, 12). The numbers and species of isolates tested are given in Table 1. Fifty-seven of the strains were tested in a previous study of bite wound infections (5) but with different comparator antimicrobial agents.

Frozen cultures were transferred twice onto brucella agar supplemented with hemin, vitamin K_1, and 5% sheep blood to assure purity and good growth. Susceptibility testing was performed according to CLSI standards (M11-A6) (13). Antimicrobial agents were reconstituted according to the manufacturers' instructions, and serial twofold dilutions of antimicrobial agents were prepared on the day of the test and added to the media at various concentrations. The agar plates were inoculated with a Steers replicator (Craft Machine Inc., Chester, Pa.), using an inoculum of 10^5 CFU/spot. Control plates without antimicrobial agents were inoculated before and after each set of drug-containing plates. Plates were incubated at 37 C° for 48 h in an anaerobic chamber (Anaerobe Systems, Morgan Hills, Calif.). The control strains tested included *Bacteroides fragilis* ATCC 25285 and *Bacteroides thetaiotaomicron* ATCC 29741. The MIC was defined as the lowest concentration of an agent that yielded no growth or a marked change in the appearance of growth compared to that of the control plates.

Standard laboratory powders were supplied as follows: tigecycline and piperacillin-tazobactam, Wyeth Ayerst Pharmaceuticals, Pearl River, N.Y.; metronidazole, Searle Research & Development, Skokie, Ill.; imipenem, Merck & Co., West Point, Pa.; meropenem, Astra Zeneca, Wilmington, Del.; moxifloxacin, Bayer Pharmaceuticals, West Haven, Conn.; clindamycin, Voigt Global Distribution, Kansas City, Mo.; and doxycycline and penicillin G, Sigma Chemical Co., St. Louis, Mo.

The MICs are shown in Table 1. The MICs of tigecycline and their frequency of occurrence for the quality control strains were as follows: *B. fragilis* ATCC 25285, 0.125, once, and 0.25, 7 times; *B. thetaiotaomicron* ATCC 29841, 0.25, once, 0.5, 3 times; and 1, 3 times.

Against gram-positive anaerobes (*Actinomyces* spp., clostridia, lactobacilli, and peptostreptococci), tigecycline compared favorably to the other agents tested. In our study, it showed excellent activity against *C. difficile*, as was also noted in several other studies (2, 3, 14), with all isolates susceptible to ≤0.06 μg/ml. Tigecycline was also very active at ≤0.25 μg/ml against 82 of 85 strains from the eight *Clostridium* species tested. Only one strain each of *C. bifermentans* and *C. butyricum* required 1 μg/ml for inhibition. Tigecycline was the most active agent tested against *Lactobacillus* species, which included multidrug-resistant strains of *L. casei*. Isolates from three infrequently studied *Actinomyces* species, including

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TABLE 1. Organisms and agents tested in this study

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Anaerobiospirillum spp. (17)[a]			
Tigecycline	0.06–0.25	0.06	0.125
Imipenem	0.06–0.125	0.06	0.125
Meropenem	≤0.015–0.03	0.03	0.03
Piperacillin-tazobactam	≤0.03–4	0.125	4
Penicillin G	≤0.015–0.5	0.125	0.25
Clindamycin	2–32	8	16
Metronidazole	2–8	4	8
Moxifloxacin	0.06–1	0.125	0.5
Doxycycline	0.125–0.5	0.25	0.5
Alistipes putredinis (previously *Bacteroides*) (10)			
Tigecycline	0.03–0.5	0.25	0.25
Imipenem	≤0.015–1	0.5	1
Meropenem	≤0.015–0.5	0.25	0.5
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	0.25–8	4	8
Clindamycin	≤0.03–>32	≤0.03	0.5
Metronidazole	0.06–0.25	0.125	0.25
Moxifloxacin	0.125–0.5	0.125	0.5
Doxycycline	0.06–16	4	16
Bacteroides tectus (15)			
Tigecycline	0.06–0.25	0.125	0.25
Imipenem	≤0.015–0.125	0.06	0.125
Meropenem	≤0.015–0.125	0.03	0.125
Piperacillin-tazobactam	≤0.03–0.25	≤0.03	0.125
Penicillin G	≤0.015–4	0.03	0.125
Clindamycin	≤0.03–2	≤0.03	0.06
Metronidazole	0.125–1	0.5	1
Moxifloxacin	≤0.03–0.25	0.06	0.125
Doxycycline	0.06–8	0.125	1.25
Bacteroides ureolyticus-Campylobacter gracilis group (19)[b]			
Tigecycline	0.06–0.5	0.06	0.25
Imipenem	≤0.015–0.125	0.03	0.125
Meropenem	≤0.015–0.06	≤0.015	0.03
Piperacillin-tazobactam	≤0.03–8	≤0.03	4
Penicillin G	≤0.015–0.5	0.06	0.5
Clindamycin	0.06–>32	0.125	1
Metronidazole	0.125–4	1	4
Moxifloxacin	≤0.03–>8	0.06	0.5
Doxycycline	0.06–8	0.125	1
Fusobacterium mortiferum (11)			
Tigecycline	0.125–0.25	0.25	0.25
Imipenem	0.06–0.5	0.5	0.5
Meropenem	≤0.03–0.5	0.25	0.5
Piperacillin-tazobactam	0.125–0.5	0.25	0.5
Penicillin G	0.25–1	0.25	1
Clindamycin	0.06–4	0.06	0.125
Metronidazole	0.25–1	0.5	0.5
Moxifloxacin	0.25–2	0.5	0.5
Doxycycline	0.06–8	0.25	0.5
Fusobacterium naviforme (10)			
Tigecycline	0.03–0.125	0.06	0.125
Imipenem	≤0.015	≤0.015	≤0.015
Meropenem	≤0.015–0.03	≤0.015	≤0.015
Piperacillin-tazobactam	≤0.03–0.5	≤0.03	0.125
Penicillin G	≤0.015–0.06	≤0.015	≤0.015
Clindamycin	≤0.03–0.06	≤0.03	0.06
Metronidazole	≤0.03–0.5	0.06	0.5
Moxifloxacin	0.125–4	0.25	1
Doxycycline	0.06–1	0.25	0.5

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TABLE 1—*Continued*

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Fusobacterium russii (12)			
Tigecycline	≤0.03–0.25	0.06	0.125
Imipenem	≤0.015–0.06	0.03	0.06
Meropenem	≤0.015	≤0.015	≤0.015
Piperacillin-tazobactam	≤0.03–0.125	≤0.03	≤0.03
Penicillin G	≤0.015	≤0.015	≤0.015
Clindamycin	≤0.03–0.06	0.06	0.06
Metronidazole	0.06–1	0.25	0.5
Moxifloxacin	0.5–8	4	8
Doxycycline	0.06–0.5	0.125	0.5
Fusobacterium varium (13)			
Tigecycline	0.06–0.25	0.25	0.25
Imipenem	0.125–1	0.5	1
Meropenem	≤0.015–0.125	0.06	0.125
Piperacillin-tazobactam	0.25–4	2	2
Penicillin G	0.25–1	0.25	0.5
Clindamycin	0.06–32	4	32
Metronidazole	0.125–1	0.5	0.5
Moxifloxacin	0.25–>8	2	>8
Doxycycline	0.125–0.5	0.25	0.5
Porphyromonas canoris (10)			
Tigecycline	≤0.015–0.06	0.03	0.06
Imipenem	≤0.015–0.06	0.03	0.06
Meropenem	≤0.015–0.03	≤0.015	≤0.015
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	≤0.015–0.25	≤0.015	≤0.015
Clindamycin	≤0.03	≤0.03	≤0.03
Metronidazole	≤0.03–1	0.5	0.5
Moxifloxacin	≤0.03–0.25	0.25	0.25
Doxycycline	0.06–0.125	0.06	0.125
Porphyromonas gingivalis (12)			
Tigecycline	≤0.015–0.03	≤0.015	0.03
Imipenem	≤0.015	≤0.015	≤0.015
Meropenem	≤0.015	≤0.015	≤0.015
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	≤0.015–0.03	≤0.015	0.03
Clindamycin	≤0.03	≤0.03	≤0.03
Metronidazole	≤0.03–0.25	≤0.03	0.06
Moxifloxacin	≤0.03–0.25	≤0.03	0.06
Doxycycline	0.06–0.125	0.125	0.125
Porphyromonas somerae (previously *levii*) (10)			
Tigecycline	0.06–0.5	0.125	0.25
Imipenem	≤0.015–0.03	≤0.015	0.0156
Meropenem	≤0.015–0.06	≤0.015	0.03
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	≤0.015–8	1	2
Clindamycin	≤0.03–>32	≤0.03	≤0.03
Metronidazole	0.125–1	0.25	1
Moxifloxacin	0.5–1	0.5	0.5
Doxycycline	0.25–8	8	8
Porphyromonas salivosa (14)			
Tigecycline	≤0.015–0.125	0.03	0.06
Imipenem	≤0.015–0.125	0.03	0.125
Meropenem	≤0.015–0.03	≤0.015	0.03
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	0.03–0.5	0.125	0.5
Clindamycin	≤0.03	≤0.03	≤0.03
Metronidazole	≤0.03–0.5	0.25	0.25
Moxifloxacin	≤0.03–0.125	0.125	0.125
Doxycycline	0.06–16	0.125	0.125

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TABLE 1—*Continued*

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Prevotella buccae (10)			
Tigecycline	0.06–2	0.06	0.5
Imipenem	0.03–0.25	0.03	0.125
Meropenem	0.06–0.5	0.06	0.25
Piperacillin-tazobactam	≤0.03–2	≤0.03	0.125
Penicillin G	0.125–>32	0.125	16
Clindamycin	≤0.03–>32	≤0.03	>32
Metronidazole	0.5–2	1	2
Moxifloxacin	0.25–1	0.5	1
Doxycycline	0.125–8	0.125	4
Prevotella disiens (10)			
Tigecycline	0.06–4	0.25	1
Imipenem	≤0.015–0.06	0.03	0.06
Meropenem	≤0.015–0.06	0.03	0.06
Piperacillin-tazobactam	≤0.03	≤0.03	≤0.03
Penicillin G	≤0.015–32	2	16
Clindamycin	≤0.03	≤0.03	≤0.03
Metronidazole	0.25–2	1	2
Moxifloxacin	0.125–1	0.5	0.5
Doxycycline	0.125–>16	4	16
Prevotella heparinolytica (13)			
Tigecycline	0.06–0.125	0.06	0.125
Imipenem	0.06–0.125	0.125	0.125
Meropenem	0.06–0.125	0.125	0.125
Piperacillin-tazobactam	≤0.03–0.25	0.125	0.25
Penicillin G	0.125–32	0.125	0.25
Clindamycin	≤0.03	≤0.03	≤0.03
Metronidazole	0.125–1	0.25	0.5
Moxifloxacin	0.125–0.25	0.25	0.25
Doxycycline	0.125–4	0.125	4
Prevotella melaninogenica (11)			
Tigecycline	0.06–4	0.25	1
Imipenem	≤0.015–1	0.03	0.03
Meropenem	≤0.015–0.125	0.06	0.06
Piperacillin-tazobactam	≤0.03–2	≤0.03	≤0.03
Penicillin G	0.03–8	4	8
Clindamycin	≤0.03–>32	≤0.03	32
Metronidazole	0.25–1	0.5	1
Moxifloxacin	0.25–>8	0.5	>8
Doxycycline	0.06–16	0.125	8
Prevotella oralis (10)			
Tigecycline	0.06–8	0.125	1
Imipenem	≤0.015–0.25	0.125	0.25
Meropenem	0.03–0.25	0.125	0.25
Piperacillin-tazobactam	≤0.03–8	≤0.03	2
Penicillin G	0.06–>32	8	>32
Clindamycin	≤0.03–>32	≤0.03	2
Metronidazole	0.25–2	1	2
Moxifloxacin	0.125–8	1	2
Doxycycline	0.06–>16	2	16
Prevotella oris (10)			
Tigecycline	≤0.015–0.5	0.125	0.25
Imipenem	≤0.015–0.25	0.03	0.125
Meropenem	≤0.015–0.125	0.06	0.125
Piperacillin-tazobactam	0.03–0.25	≤0.03	0.25
Penicillin G	≤0.015–>32	0.06	16
Clindamycin	0.03–0.5	≤0.03	0.25
Metronidazole	0.25–2	1	2
Moxifloxacin	0.125–1	0.25	0.5
Doxycycline	0.03–16	0.125	8

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TABLE 1—*Continued*

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Veillonella spp. (15)			
Tigecycline	0.5–1	1	1
Imipenem	0.03–0.5	0.125	0.5
Meropenem	≤0.015–0.125	0.03	0.125
Piperacillin-tazobactam	0.5–>64	8	64
Penicillin G	0.125–4	2	4
Clindamycin	0.06–1	0.125	0.25
Metronidazole	2–8	4	4
Moxifloxacin	0.125–8	2	8
Doxycycline	0.5–>16	2	16
Actinomyces spp. (17)[c]			
Tigecycline	0.03–1	0.125	0.5
Imipenem	≤0.015–0.5	0.06	0.125
Meropenem	≤0.015–0.5	0.06	0.25
Piperacillin-tazobactam	≤0.03–2	0.25	1
Penicillin G	≤0.015–0.5	0.06	0.35
Clindamycin	≤0.03–>32	0.06	0.25
Metronidazole	1–>16	>16	>16
Moxifloxacin	0.25–2	1	2
Doxycycline	0.06–16	0.5	16
Actinomyces odontolyticus (15)			
Tigecycline	0.06–0.25	0.125	0.25
Imipenem	0.06–0.125	0.06	0.125
Meropenem	0.06–0.25	0.125	0.25
Piperacillin-tazobactam	0.125–2	0.5	1
Penicillin G	0.03–0.125	0.06	0.125
Clindamycin	≤0.03–0.5	0.125	0.25
Metronidazole	16–>16	>16	>16
Moxifloxacin	1–2	1	2
Doxycycline	0.25–1	0.25	1
Actinomyces viscosus (12)			
Tigecycline	0.06–1	0.125	0.25
Imipenem	≤0.015–0.06	0.06	0.06
Meropenem	≤0.015–0.25	0.03	0.125
Piperacillin-tazobactam	≤0.03–8	0.5	0.5
Penicillin G	≤0.015–0.5	0.06	0.125
Clindamycin	≤0.03–>32	0.25	0.5
Metronidazole	16–>16	>16	>16
Moxifloxacin	0.125–1	0.5	1
Doxycycline	0.25–2	0.25	0.5
Clostridium bifermentans (11)			
Tigecycline	0.06–1	0.06	0.5
Imipenem	0.06–0.25	0.125	0.25
Meropenem	0.06–0.125	0.06	0.125
Piperacillin-tazobactam	0.125–0.5	0.5	0.5
Penicillin G	≤0.015–0.25	0.125	0.25
Clindamycin	≤0.03–16	≤0.03	0.25
Metronidazole	0.25–8	1	2
Moxifloxacin	0.5–0.5	0.5	0.5
Doxycycline	≤0.03–8	0.06	4
Clostridium butyricum (11)			
Tigecycline	0.06–1	0.06	0.25
Imipenem	0.125–1	0.25	1
Meropenem	0.06–1	0.125	0.5
Piperacillin-tazobactam	0.25–2	0.25	2
Penicillin G	0.25–>32	0.25	32
Clindamycin	≤0.03–0.5	0.25	0.25
Metronidazole	1–2	1	2
Moxifloxacin	0.5–1	0.5	0.5
Doxycycline	≤0.03–8	≤0.03	4

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TABLE 1—*Continued*

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Clostridium cadaveris (10)			
Tigecycline	0.06–0.125	0.06	0.06
Imipenem	0.06–0.125	0.06	0.06
Meropenem	≤0.015–0.03	≤0.015	≤0.015
Piperacillin-tazobactam	≤0.03–0.25	≤0.03	≤0.03
Penicillin G	0.125–0.125	0.125	0.125
Clindamycin	≤0.03–2	≤0.03	0.25
Metronidazole	0.06–0.125	0.125	0.125
Moxifloxacin	0.25–0.25	0.25	0.25
Doxycycline	0.125–8	0.125	8
Clostridium clostridioforme group (10)[d]			
Tigecycline	≤0.015–0.06	0.03	0.06
Imipenem	0.25–2	1	2
Meropenem	0.125–2	1	2
Piperacillin-tazobactam	0.125–64	8	8
Penicillin G	0.25–>32	1	16
Clindamycin	≤0.03–1	0.5	1
Metronidazole	≤0.03–1	0.125	0.25
Moxifloxacin	2–8	4	8
Doxycycline	2–16	4	16
Clostridium difficile (12)			
Tigecycline	0.06	0.06	0.06
Imipenem	4–8	4	8
Meropenem	1–4	1	2
Piperacillin-tazobactam	4–16	8	16
Penicillin G	0.5–4	1	4
Clindamycin	2–>32	2	>32
Metronidazole	0.25–1	0.5	1
Moxifloxacin	0.5–>8	1	>8
Doxycycline	0.06–16	0.125	8
Clostridium innocuum (11)			
Tigecycline	0.06–0.125	0.06	0.125
Imipenem	1–4	2	4
Meropenem	1–2	1	2
Piperacillin-tazobactam	0.06–2	1	2
Penicillin G	0.125–1	0.5	0.5
Clindamycin	0.25–>32	0.5	>32
Metronidazole	0.5–2	0.5	2
Moxifloxacin	0.125–>8	1	>8
Doxycycline	0.25–>16	2	16
Clostridium ramosum (10)			
Tigecycline	0.125	0.125	0.125
Imipenem	0.125–0.5	0.25	0.5
Meropenem	1–1	1	1
Piperacillin-tazobactam	0.06–1	0.125	1
Penicillin G	0.03–1	0.125	0.5
Clindamycin	1–>32	2	>32
Metronidazole	0.5–2	1	2
Moxifloxacin	1–2	2	2
Doxycycline	0.125–16	4	16
Clostridium tertium (10)			
Tigecycline	0.06	0.06	0.06
Imipenem	0.125–0.25	0.25	0.25
Meropenem	0.25	0.25	0.25
Piperacillin-tazobactam	4–16	16	16
Penicillin G	0.5–2	1	2
Clindamycin	1–8	8	8
Metronidazole	0.5–4	1.5	2.2
Moxifloxacin	0.25–0.5	0.25	0.5
Doxycycline	0.03–0.125	0.06	0.06–>64

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TABLE 1—*Continued*

Organism (no. of isolates) and agent	MIC (μg/ml)		
	Range	50%	90%
Lactobacillus spp. (15)[e]			
Tigecycline	0.06–1	0.25	0.5
Imipenem	≤0.015–8	0.125	8
Meropenem	0.03–>16	0.25	>16
Piperacillin-tazobactam	≤0.03–16	1	16
Penicillin G	≤0.015–16	0.5	4
Clindamycin	≤0.03–>32	0.125	16
Metronidazole	1–>16	>16	>16
Moxifloxacin	0.125–>8	1	2
Doxycycline	0.125–>16	4	8
Anaerobic gram-positive cocci (20)[f]			
Tigecycline	≤0.015–0.25	0.06	0.25
Imipenem	≤0.015–0.5	0.03	0.06
Meropenem	≤0.015–0.5	0.03	0.125
Piperacillin-tazobactam	≤0.03–0.5	≤0.03	0.25
Penicillin G	≤0.015–1	1	1
Clindamycin	≤0.03–32	0.125	1
Metronidazole	0.06–8	0.5	1
Moxifloxacin	0.06–>8	0.25	2
Doxycycline	0.06–>16	1	16

[a] *A. succiniciproducens* (3), *A. thomasii* (14).
[b] *Bacteroides ureolyticus* (10), *Campylobacter gracilis* (9).
[c] *A. israellii* (7), *A. meyeri* (3), *A. naeslundii* (6), and *A. neuii* (1).
[d] *C. bolteae* (3), *C. hathewayi* (3), and *C. clostridioforme* (4).
[e] *L. acidophilus* (1), *L. brevis* (2), *L. casei* (4), *L. catenaforme* (3), *L. delbruekii* (1), *L. gasseri* (1), *L. jensenii* (1), and *L. lactis* (2).
[f] *Peptoniphilus asaccharolyticus* (6), *Finegoldia magna* (5), *Peptostreptococcus micros* (5), and *Anaerococcus prevotii* (4).

several doxycycline-resistant strains, were all susceptible to ≤1 μg/ml of tigecycline.

The gram-negative anaerobes were also very susceptible to tigecycline, with MICs of ≤1μg/ml for 228 of 232 strains. Isolates requiring >1 μg/ml for inhibition included one strain each of *Prevotella buccae* (MIC, 2 μg/ml; also penicillin and clindamycin resistant), *Prevotella melaninogenica* (MIC, 4 μg/ml; also penicillin, moxifloxacin, and doxycycline resistant), and 1 strain each of *Prevotella oralis* and *Prevotella disiens* (MIC, 8 and 4 μg/ml, respectively; both penicillin and doxycycline resistant). *Veillonella* species generally required 1 μg/ml of tigecycline for inhibition.

Overall, tigecycline had excellent in vitro activity against both gram-positive and gram-negative unusual anaerobes, with 395 of 396 (99.7%) of strains susceptible at ≤4 μg/ml, the FDA-assigned breakpoint.

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