Activity of Minocycline and Other Tetracyclines Against Tetracycline-Sensitive and -Resistant Staphylococci (35292)

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Minocycline (7-dimethylamino-6-demethyl-6-deoxytetracycline) is a new tetracycline analog. Its preparation and some of its properties have been described by Martell and Boothe (1). Against experimental infections in mice, it was more effective than tetracycline, both on a dosage and a plasma level basis (2). *In vitro*, it ranked as the most active of seven tetracycline analogs tested against various bacteria isolated from patients (3). It had unexpected activity against staphylococci resistant to tetracycline (1–4). To further evaluate its potential clinical advantage, we compared minocycline with doxycycline,[1] methacycline,[1] demethylchlortetracycline[2] and tetracycline[2] for activity *in vitro* against staphylococci isolated from patients at various clinical centers. We also compared the activity of these drugs in mice against lethal infections produced by a tetracycline-sensitive and a tetracycline-resistant strain of *Staphylococcus aureus*.

Methods. In vitro. The *Staphylococcus aureus* strains were recently isolated from patients and were supplied through the courtesy of the following physicians or medical centers: Dr. J. V. Bennett, Communicable Disease Center, Atlanta, Ga.; Dr. A. Bondi, Hahnemann Medical College, Philadelphia, Pa.; Dr. R. M. Fine, Decatur, Ga.; Dr. Maxwell Finland, Boston City Hospital, Boston, Mass.; Dr. W. J. Holloway, Wilmington Medical Center, Wilmington, Del; Dr. C. D. Graber, Medical College of So. Carolina, Charleston, So. Carolina; Dr. W. C. Grater, Dallas, Texas; Dr. Z. Kaminski, Newark City Hospital, Newark, N.J.; Dr. C. M. Kunin, Univ. of Wisconsin, Madison, Wisc.; Dr. E. Podell, Louisville, Ky.; Dr. C. Thurmond, New Orleans, La.; Wheeler Medical Labs, Kansas City, Mo.; Dr. C. T. Yarington, Omaha, Nebraska; VA Hospital, Washington, D.C.

The activity of the tetracyclines was determined by the agar dilution method. The drugs were incorporated in peptone casein agar (Seed agar, Baltimore Biological Laboratories) in petri plates. The agar surfaces were inoculated with 10^{-3} dilutions of 5-hr Penassay broth cultures by means of the Steers multiple inocula replicator (5). After incubation at 37° for 24 hr, the lowest concentration of drug completely inhibiting growth was recorded as the minimum inhibitory concentration (MIC). The susceptibility of each culture to all 5 analogs was determined simultaneously. Each culture was tested 2 or 3 times and the geometric mean of the MIC's was determined.

In vivo. The methods used have been described (2). Carworth Farms CFl female mice weighing 18 to 22 g each were used. Infections were produced by the intraperitoneal injection of 0.5 ml (2.5×10^8 viable units) of an undiluted 5-hr blood broth culture of the tetracycline-resistant *S. aureus* strain Rose (ATCC 14154), or of 0.5 ml (1×10^7 viable units) of a 10^{-2} dilution in broth of a 5-hr culture of the tetracycline-sensitive *S. aureus* Smith strain (ATCC 13709).

The tetracyclines were suspended in 0.2% aqueous agar, and single oral doses were administered within 1 hr after initiation of in-

[1] The trademark of Chas. Pfizer and Co., Inc. for doxycycline is Vibramycin and for methacycline is Rondomycin.

[2] The trademark of American Cyanamid Co. for demethylchlortetracycline is Declomycin and for tetracycline is Achromycin.

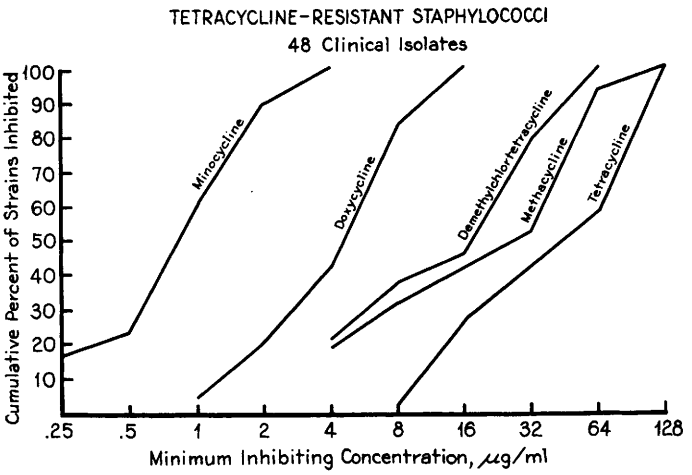

FIG. 1. Susceptibility of tetracycline-resistant staphylococci to minocycline and other tetracycline analogs.

fection. The median effective doses were determined from pooled test results by the method of Litchfield and Wilcoxon (6).

Results and Discussion. The staphylococcal cultures were grouped according to their sensitivity to tetracycline; those requiring more than 4 μg/ml for complete inhibition were classified as resistant.

Minocycline was the most active analog against the 48 tetracycline-resistant cultures. All 48 cultures (100%) were inhibited by 4 μg/ml of minocycline. At this concentration, only 20 strains (40%) were inhibited by doxycycline, 10 strains (21%) by demethylchlortetracycline and 9 strains (19%) by methacycline (Fig. 1).

Against 53 tetracycline-sensitive cultures, minocycline, doxycycline, demethylchlortetracycline, and methacycline were about equivalent and were more active than tetracycline.

In mice, minocycline was effective and potent against the lethal infection produced by the tetracycline-resistant *S. aureus* strain Rose. The median lethal dose (LD_{50}) of minocycline administered to mice in a single oral dose was 3100 mg/kg (2) and the median effective dose (ED_{50}) against the tetracycline-resistant staphylococcal infection was 3.8 mg/kg. Thus, the therapeutic index (LD_{50}/ED_{50}) was 820. The other tetracyclines were ineffective against this infection (Table I).

Against the lethal infection produced by the tetracycline-sensitive *Staphylococcus aureus* strain Smith, minocycline was the most potent, on a dosage basis, of the tetracyclines (Table II). Its therapeutic index (LD_{50}/ED_{50}) was 2000. Doxycycline was next in potency. Our results with doxycycline are consistent with those of English (7) who found doxycycline to be more active than methacycline, demethylchlortetracycline, or tetracycline against an infection produced by a tetracycline-sensitive strain of *S. aureus*.

Minocycline is a unique tetracycline. Our results and those of others (3, 4) show that it has a clinical potential not only for treatment of infections caused by tetracycline-sensitive organisms, but also for treatment of infections caused by many strains of staphylococci resistant to other tetracyclines.

Summary. Minocycline was more active than doxycycline, demethylchlortetracycline, methacycline, and tetracycline *in vitro* against 101 clinical staphylococcal isolates. It was the only analog active against all 48 tetracycline resistant strains at concentrations of 4 μg/ml or less. In mice, minocycline was the most potent analog against a tetracycline-sensitive staphylococcal infection and the only drug effective against a tetracycline-resistant staphylococcal infection. The results show that minocycline has a potential clini-

TABLE I. Effect of Five Tetracycline Analogs on the Tetracycline-Resistant *Staphylococcus aureus* Rose Infection in Mice.[a]

Single oral dose (mg/kg)	Alive/total mice, 14 days after initiation of infection				
	Minocycline	Doxycycline	Methacycline	DMCT[b]	Tetracycline
1024	—	5/20	0/20	0/20	0/20
256	—	3/20	0/20	1/20	1/20
64	—	0/10	0/10	1/10	0/10
16	20/20	0/10	0/10	0/10	0/10
8	18/20	—	—	—	—
4	9/20	—	—	—	—
2	3/20	—	—	—	—
1	0/20	—	—	—	—
Median effective dose (mg/kg)	3.8 (2.6–5.5)[c]	—	—	—	—

[a] Infected nontreated controls: 39/40 mice died within 1 day after initiation of infection. Noninfected nontreated controls: 20/20 mice were alive on the 14th day after initiation of infection.

[b] Demethylchlortetracycline.

[c] Numbers in parentheses are 95% confidence limits.

TABLE II. Effect of Five Tetracycline Analogs on the Tetracycline-Sensitive *S. aureus* Smith Infection in Mice.[a]

Single oral dose (mg/kg)	Alive/total mice, 14 days after initiation of infection				
	Minocycline	Doxycycline	Methacycline	DMCT[b]	Tetracycline
64	—	—	7/10	10/10	15/20
32	—	—	11/20	11/20	3/20
16	—	—	6/20	5/20	2/20
8	20/20	15/20	1/20	1/20	0/20
4	18/20	12/20	1/20	0/20	0/20
2	14/20	3/20	0/20	—	—
1	5/20	1/20	—	—	—
0.5	0/20	0/20	—	—	—
Median effective dose (mg/kg)	1.5 (1.1–2.1)[c]	4.0 (3.1–5.2)	28 (20–38)	28 (20–39)	47 (35–63)

[a] Infected nontreated controls: 40/40 mice died within 1 day after initiation of infection. Noninfected nontreated controls: 20/20 mice were alive on the 14th day after initiation of infection.

[b] Demethylchlortetracycline.

[c] Numbers in parentheses are 95% confidence limits.

cal advantage over other tetracyclines.

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