APPLIED MICROBIOLOGY, July 1969, p. 21-23
Vol. 18, No. 1

In Vitro Susceptibility of *Neisseria gonorrhoeae* to Nine Antimicrobial Agents

JOHN E. MARTIN, JR., ARZELL LESTER, DOUGLAS S. KELLOGG, JR., AND JAMES D. THAYER[1]

Venereal Disease Research Laboratory, State and Community Services Division, National Communicable Disease Center, Health Services and Mental Health Administration, Public Health Service, Atlanta, Georgia 30333

Received for publication 18 March 1969

The in vitro action of nine antibiotics was tested by the agar streak method against 45 gonococcal strains isolated from penicillin-therapy failures. The penicillin susceptibility range of these strains was 0.003 to 1.32 μg/ml, and the tetracycline susceptibility range was 0.125 to 2.0 μg/ml. Minimal inhibitory concentrations of minocycline and doxycycline paralleled the activity of tetracycline and ranged from 0.125 to 1.0 μg/ml and 0.125 to 2.0 μg/ml, respectively. Rifampicin, with a narrow range of 0.5 to 1.0 μg/ml, inhibited 75% of the strains at 0.5 μg/ml. The range for cephaloridine and cephaloglycine was 0.5 to 20.0 μg/ml, but another cephalosporium derivative, cephalexin, exhibited greater activity in its range of 0.25 to 20.0 μg/ml. A semisynthetic penicillin, carbenicillin, with a range of 0.025 to 0.75 μg/ml, displayed more activity against the lower susceptible penicillin G gonococcal strains.

The true incidence of gonorrhea is unknown, but it is conservatively estimated that 1.5 million new cases occur annually in the United States (4). Although penicillin G is the therapeutic agent of choice in the treatment of this disease, it is doubtful if it will remain so indefinitely because of the steadily rising resistance of gonococci to this drug (5), necessitating an ever-increasing dosage. Furthermore, approximately 5 to 10% of the population is now, to some degree, allergic to penicillin; and these patients must be treated with an alternate, and often less effective, antibiotic (1). Therefore, the need to study newer antimicrobial agents is apparent. Desirable characteristics of a new therapeutic agent are that: (i) it should be nonallergenic, (ii) it should be highly active against the penicillin-resistant and penicillin-sensitive gonococcal strains, and (iii) it should be injectable so as to aid in the public health control of the disease.

In this report, a comparison is made of the in vitro activity of penicillin G and tetracycline with the in vitro activity of seven newer antibiotics against isolates of gonococci. These isolates were from patients with gonorrhea who reportedly failed to respond to penicillin therapy. The seven antibiotics tested were cephaloridine, cephaloglycine, cephalexin (Eli Lilly & Co., Indianapolis, Ind.), rifampicin (Pitman-Moore Company), minocycline (Lederle Laboratories, Pearl River, N.Y.), doxycycline, and carbenicillin (Chas. Pfizer & Co., Inc., Brooklyn, N.Y.).

[1] Deceased July 18, 1968.

MATERIALS AND METHODS

The method for performing susceptibility tests of the antimicrobials has been previously described (6). We added the desired concentrations of the antimicrobial agent to conventional chocolate medium (Difco) and streaked the agar surface with a standardized inoculum of the gonococcal strain under test. After 24 and 48 hr of incubation, the plates were observed for growth. The drug concentration preventing growth was taken as the minimal inhibitory concentration (MIC) or susceptibility of the strain.

The gonococcal isolates used in this study were received by this laboratory during the past 3 years for penicillin sensitivity testing. They were from presumed penicillin-therapy failure patients.

The control drugs, sodium penicillin G and tetracycline, were obtained from the Antibiotics Division of the Food and Drug Administration.

RESULTS

The susceptibility range and the MIC average of the antibiotics are given in Table 1 and shown in Fig. 1 and 2 for 45 gonococcal strains.

The activity range for penicillin G was 0.003 to 1.32 μg/ml, and the mean MIC was 0.33 μg/ml (0.5 IU/ml); for tetracycline the range was 0.125 to 2.0 μg/ml, and the mean MIC was 0.75 μg/ml.

Carbenicillin, an injectable semisynthetic penicillin, exhibited a range of 0.025 to 0.75 μg/ml with its mean of 0.31 μg/ml.

TABLE 1. *Susceptibility of 45 gonococcal strains to nine antibiotics*

Antimicrobial agents	Susceptibility (MIC μg/ml)	
	Range	Mean
Penicillin G	0.003–1.32	0.33
Carbenicillin	0.025–0.75	0.31
Tetracycline	0.125–2.0	0.75
Minocycline	0.125–1.0	0.52
Doxycycline	0.125–2.0	0.83
Rifampicin	0.5–1.0	0.62
Cephalexin	0.25–20.0	3.16
Cephaloglycine	0.5–20.0	5.65
Cephaloridine	0.5–20.0	4.94

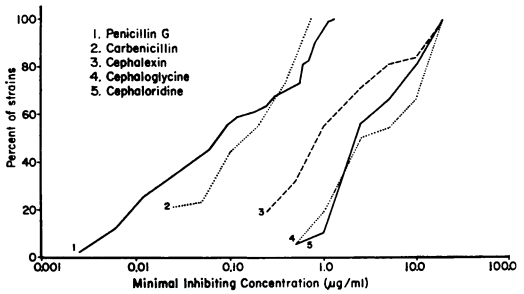

FIG. 1. *In vitro susceptibility of N. gonorrhoeae to specified antimicrobial agents.*

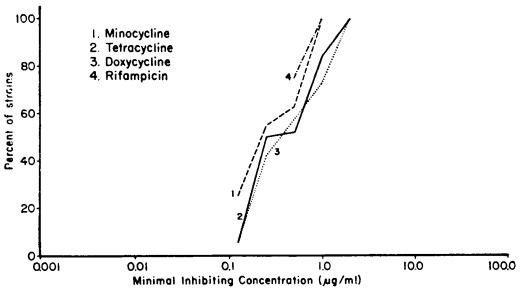

FIG. 2. *In vitro susceptibility of N. gonorrhoeae to specified antimicrobial agents.*

The three derivatives of cephalosporium C paralleled one another with respective ranges, with all strains inhibited at 20 μg/ml.

When minocycline was compared with tetracycline and doxycycline, complete inhibition was observed at 1.0 μg/ml against all cultures that had required 2.0 μg/ml of tetracycline and doxycycline for inhibition.

Rifampicin exhibited a very narrow susceptibility range of 0.5 to 1.0 μg/ml.

DISCUSSION

Effective drugs for the treatment of gonorrhea cannot be predicted by their in vitro sensitivity alone, but this knowledge, coupled with other factors such as degree of absorption, maximal blood concentrations, duration of effective blood concentrations of unbound drug, and excretion give some basis for selection.

In 1961 gonococcal strains from penicillin-therapy failure patients were tested for tetracycline sensitivity, and they showed a susceptibility range of 0.06 to 0.25 μg/ml with a mean of 0.13 μg/ml. In this study, 50% of the strains tested were susceptible to concentrations between 0.5 and 2.0 μg/ml with a mean of 0.75 μg/ml. This decrease in susceptibility may indicate a trend toward tetracycline-resistant gonococci.

Although the activity of carbenicillin was similar to penicillin G, it was observed that gonococcal isolates requiring greater than 0.3 μg/ml of carbenicillin for complete inhibition were less susceptible to an equivalent concentration of penicillin G. In an earlier report of in vitro activity of nine semisynthetic penicillins, none showed a greater potency than penicillin G for resistant gonococci (3).

In a clinical trial (2), cephaloridine was shown to be an effective antibiotic in the treatment of gonorrhea. When compared to that drug, the effective action of cephaloglycine and cephalexin for the gonococcal isolates suggests that both drugs may be suitable agents for therapeutic trials in the treatment of gonorrhea.

It will be noted that 25% of the gonococcal isolates from therapy-failure patients were susceptible to 0.012 μg/ml of penicillin. This result was somewhat surprising since the recommended dosage of penicillin for gonorrhea would seem to assure a blood level much greater than this amount. However, since very little history accompanied these strains, the possibility of reinfection or inadequate dosage must also be considered.

Our results in this study, when considered with other knowledge about the antibiotics studied, indicate that there may be several alternate antibiotics for therapy of gonorrhea.

LITERATURE CITED

1. Lucas, J. B., J. D. Thayer, P. M. Utley, T. E. Billings, and J. F. Hackney. 1965. Cephalothin therapy in male gonorrhea. Antimicrobial Agents and Chemotherapy—1965, p. 686–688.

2. Lucas, J. B., J. D. Thayer, P. M. Utley, T. E. Billings, and J. F. Hackney. 1966. Treatment of gonorrhea in males with cephaloridine. J. Amer. Med. Ass. 199:919–921.
3. Thayer, J. D., and N. W. Axnick. 1963. Susceptibility of gonococci to ten penicillins. Antimicrobial Agents and Chemotherapy—1963, p. 427–429.
4. Thayer, J. D., and M. B. Moore, Jr. 1964. Gonorrhea: Present knowledge and control efforts. Med. Clin. N. Amer. 48: 755–765.
5. Thayer, J. D., and W. Garson. 1965. The gonococcus, p. 451–467. *In* R. Dubos and J. Hirsch (ed.), Bacterial and mycotic infections in man, 4th ed. J. B. Lippincott Co., Philadelphia.
6. U.S. Department of Health, Education, and Welfare. 1962. Gonococcus—procedures for isolation and identification, (Rev.) U.S. Public Health Serv. Publ. 499.