Antigenic Types of "Large Colony" Human Genital Mycoplasmas

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Received for publication 29 September 1965

Reports from Europe (Nicol and Edwards, Brit. J. Venereal Diseases 29:141, 1953) have previously indicated that the common human genital mycoplasma which produces "large colonies" on agar is Mycoplasma hominis type 1. "Large colony" refers to the cultural characteristic that differentiates these strains from the T strains of Shepard (J. Bacteriol. 71:362, 1956). North American studies (Norman, Saslaw, and Kuhn, Proc. Soc. Exptl. Biol. Med. 75:718, 1950) suggested that a different antigenic type of mycoplasma existed in the genital tract, and this was subsequently called "M. hominis type 2," the prototype being strain Campo. Recently, the nature of M. hominis type 2 has been questioned because of serological identity to M. arthritidis (Lerncke, J. Hyg. 62:199, 1964). In view of this uncertainty, and also because the frequency with which M. fermentans (Edward and Freundt, J. Gen. Microbiol. 14:197, 1956) occurs in the human genital tract has not been evaluated, a study was made of the antigenic types of "large colony" human genital mycoplasmas.

Vaginal swabs were taken from women admitted to the city jail. Urethral swabs and scrapings were obtained from both normal males and patients with nongonococcal urethritis. These swabs and scrapings were plated on PPLO Agar (Difco) supplemented with 10% horse serum, 1% Oxoid yeast extract, and 1,000 units of penicillin per ml. The cultures were incubated by Fortner's method. "Large colony" mycoplasmas were subcultured in similar broth, and antigenic typing was performed by the growth-inhibition method of Clyde (J. Immunol. 92:958, 1964). Stock strains of the oral and genital human mycoplasmas were obtained from R. M. Chanock of the National Institute of Infectious Diseases, Bethesda, Md.; M. pneumoniae was obtained from G. E. Kenny, Department of Preventive Medicine, University of Washington, Seattle. Rabbit antisera were obtained by the intravenous injection of saline-washed deposits of horse serum broth cultures. The injections were given three times weekly for 2 weeks, followed by a booster injection at the end of the third week. The rabbit antisera were tested by the growth-inhibition test of Edward and Fitzgerald (J. Pathol. Bacteriol. 68:23, 1954), by use of serial dilutions of the sera in agar. At a dilution of 1:20, complete inhibition of growth was obtained; this inhibition was absolutely strain-specific. No cross-inhibition occurred with any serum or strain. Partial inhibition was demonstrable with all sera in dilutions varying from 1:100 to 1:800. For the antigenic typing of unknown strains, the disc inhibition method of Clyde was found to be simple, specific, and easily read. A broth culture of the unknown strain was evenly spread over the surface of an agar plate, 0.1 ml for a plate 100 mm in diameter. After drying for 1 hr, six blotting-paper discs (6 mm in diameter) were placed equidistantly on the agar, each one having been previously moistened with approximately 0.02 ml of the respective typing antiserum. After 5 days of incubation at 37 C, the plates were examined for zones of inhibition around the discs.

Of the 100 strains isolated from women, 94 were inhibited by M. hominis type 1 antiserum; none was inhibited by M. hominis type 2 antiserum. Six strains were inhibited by none of the antisera employed; each of these strains fermented glucose with the production of acid. They were considered to be M. fermentans-like strains, and were forwarded to R. M. Chanock for further antigenic study. All of 17 strains isolated from men were inhibited by M. hominis type 1 antiserum.

It is concluded that M. hominis type 2 is not present in the genital tract of individuals in this city. It is also concluded that M. hominis type 1 is the usual "large colony" mycoplasma in the human genital tract, but that about 5% of "large colony" human genital strains are closely related to M. fermentans.

This investigation was supported by the Canadian Arthritis and Rheumatism Society and by Federal Health Grant 609-7-40.