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Full Length Research Paper

Antibiotic sensitivity of human genital mycoplasmas

Jafar Khan[1]*, Robeena Farzand[1] and Pir Bux Ghumro[2]

[1]Department of Microbiology, Kohat University of Science and Technology Kohat, 26000, NWFP, Pakistan.
[2]Department of Microbiology, Quaid-i-Azam University, Islamabad 45320, Pakistan.

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The therapeutic failure of antimycoplasma drugs, subject to resistance is vital for specific microbiological investigation to overcome urogenital diseases among women. The susceptibility of 5 *Mycoplasma hominis* strains and 11 strains of *Ureaplasma urealyticum* were studied against six antibiotics namely: minocycline, lincomycin, erythromycin, tetracycline, doxycycline and rosaramicin. Both *M. hominis* and *Ureaplasmas* strains were highly resistant to tetracycline. It was observed that both rosaramicin and minocycline were active growth inhibitors for Ureaplasmas. However, erythromycin and lincomycin showed differential growth inhibitory patterns for *M. hominis* and *Ureaplasma* strains. In comparison, the minocycline and doxycycline exhibited similar antibiotic activity against *Ureaplasma* and *M. hominis.* The *in vitro* antibiotic sensitivity analysis for rosaramicin and erythromycin was also noted to be at the same level for all the isolates.

Key words: *Mycoplasma hominis, Ureaplasma urealyticum,* antibiotic sensitivity, drug resistance.

INTRODUCTION

Mycoplasmas belong to the class Mollicutes which also contain Ureaplasmas, Acholeplasmas, Spiroplasmas, the newly classified Haemoplasmas and other wall-less bacteria. They are characterized by small size (500 - 1100 kbp), lack of cell wall, extremely fastidious for an *in vitro* environment and tendency to form centered colonies on solid medium (Baron et al., 1998). While all species live as parasitic existence colonizing the mucosal epithelium and relying on the host to provide most nutritional requirements, the majority are commensals, though occasionally opportunistic invading lung tissue following bacterial or viral infections (Ruth and Sharon, 2006). Mycoplasmas are inherently resistant to those antibiotics that target the cell wall; so agents like β-lactams are completely inactive, since mycoplasmas and ureaplasmas do not have a cell wall. In addition, they have higher mutation rates than conventional bacteria which mean that they can rapidly develop resistance to other drugs including the oxytetracyclines and tylosin as has been seen in Europe recently (Ayling et al., 2000; Thomas et al., 2003).

*Corresponding author. E-mail: jafarkhan1@yahoo.com. Tel: +92-300-5314922.

Since culturing the organism is difficult, limited information has been available regarding its antimicrobial drug susceptibility. *In vitro* studies suggest that it is susceptible to tetracyclines, macrolides and fluoroquinolones (Hannan and Woodnutt, 2000; Yasuda et al., 2005). Although reduced susceptibility to tetracyclines and specifically to fluoroquinolones has also been reported (Hannan, 1998; Yasuda et al., 2005; Hamasuna et al., 2005). We performed susceptibility tests on isolates of five common strains of *Mycoplasma hominis* and eleven strains of *Ureaplasma* from patients with urogenital infections. The antibiotics tested were, lincomycin, erythromycin, tetracycline, doxycycline, and rosaramicin as being conventionally used for the routine treatment practice.

MATERIALS AND METHODS

A total of 337 specimens including high vaginal swabs (HVS) and urine from infertile women, suffering from vaginitis and cervicitis of unexplained origin, were screened out for prevalence of *Mycoplasmas.* The study was conducted on the regularly attending women patients of gynaecology/obstetrics departments at the tertiary care hospitals of Rawalpindi-Isamabad (Pakistan). A written informed consent was also obtained in this regard for the investigations/diagnosis to meet the ethical standards of the study. These test specimens yielded five strains of *M. hominis*, FG 140,

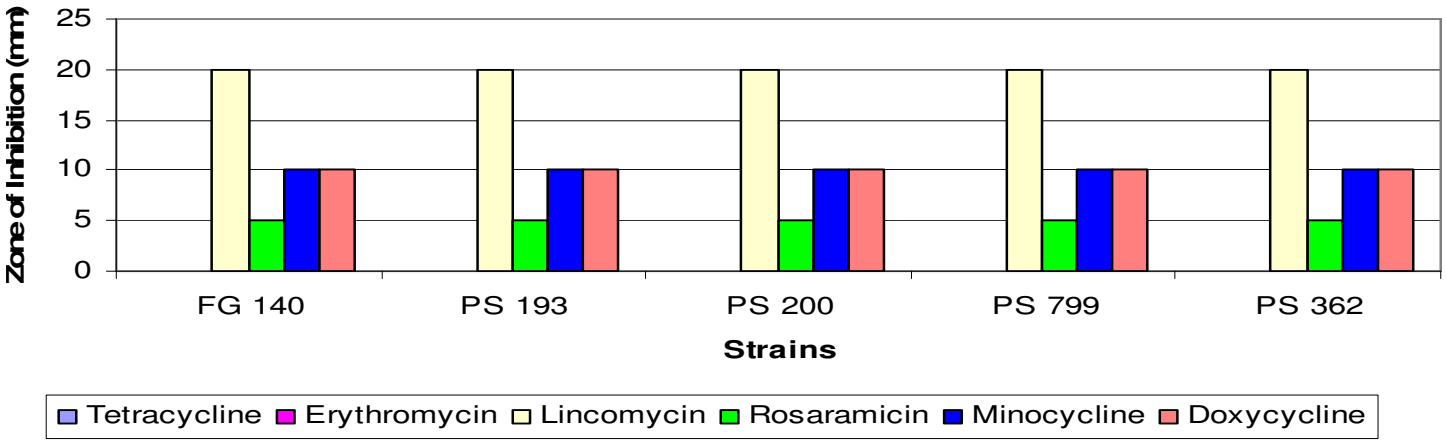

Figure 1. Antibiotic sensitivity of *M. hominis* strains and the pattern of zone of inhibition.

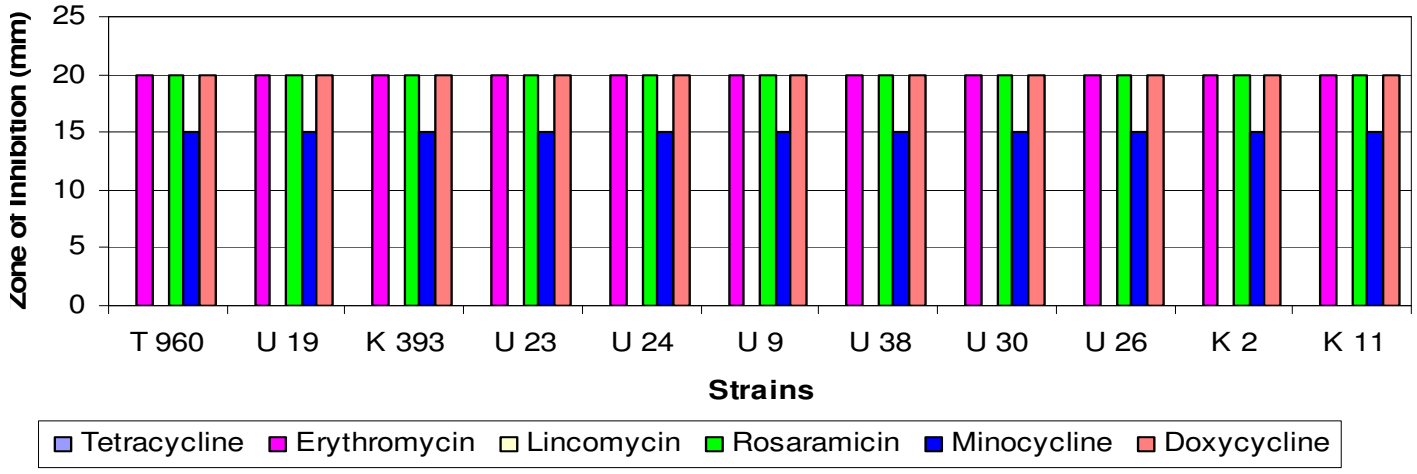

Figure 2. Antibiotic sensitivity of *Ureaplasma* strains and the pattern of zone of inhibition.

PS 193, PS 200, PS 799 and PS 362 and eleven *Ureaplasma strains*, T 960, U 19, K 393, U 23, U 24, U 9, U 30, U 26, K 2 and K 11. These strains were further tested for *in vitro* antibiotic sensitivity and inhibition zones were measured in mm according to National Committee for Clinical Laboratory Standards (NCCLS, 2002). The used sensitivity discs were minocycline, lincomycin, erythromycin, tetracycline, doxycycline and rosaramicin. All the antibiotics discs were stored at -20°C before use.

Briefly, over night fresh culture (log-phase broth culture) of *Mycoplasma* was flooded onto solid *Mycoplasma* medium plates with the identified strains and excess of culture was removed. The plates were left at 25°C for 24 h under sterile conditions. The antibiotic discs were applied and the culture plates were incubated at 37°C for 24 - 48 h. The plates were examined under a stereo microscope (Leitz) and zones of inhibition were measured in millimeters from the edge of the disc and presented as + (5 mm), + + (10 mm), + + + (15 mm) and + + + + (20 mm).

RESULTS AND DISCUSSION

The antibiotics rosaramicin and minocycline were found as active growth inhibitors of Ureaplasmas. Both *M. hominis* and *Ureaplasma* strains were highly resistant to tetracycline. Erythromycin and lincomycin were found differentially active against *M. hominis* and *Ureaplasma* strains. Lincomycins was found active against *M. hominis* but not against *Ureaplasmas,* whereas erythromycin on the other hand inhibited the growth of Ureaplasmas but showed very less activity against *M. hominis*. Minocycline and doxycycline were most active against Ureaplasmas and were also considerably effective against *M. hominis*. The rosaramicin and erythromycin, both were found similar in growth inhibition of *M. hominis* and *Ureaplasmas*. All these results are graphically represented in figures 1, 2 and 3.

The purpose of this study basically was to determine the antibiotic sensitivity of human genital *mycoplasmas*, which are isolated from high vaginal swabs and urine of infertile women. Prior studies documented the susceptibility of different Mycoplasmas species to several antimicrobial agents (Karamova et al., 2004; Ghaleh et al., 2008). It is evident in our observations that the strains

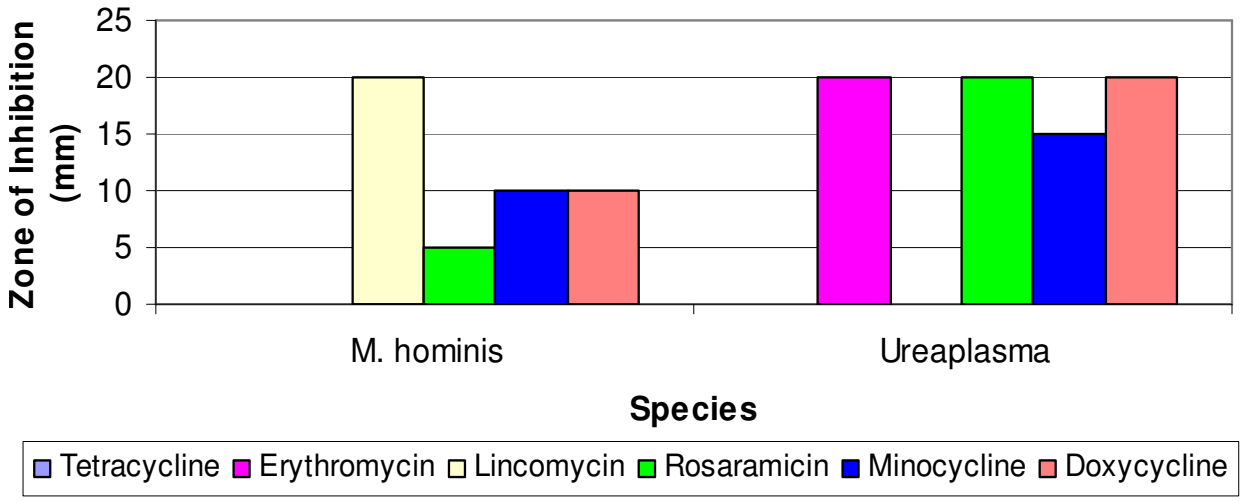

Figure 3. Comparison of antibiotic sensitivity of *Mycoplasmas* species.

of *M. hominis* have shown resistance against erythromycin which resembles similar findings in this regard as reported earlier (Huang et al., 2003; Karamova et al., 2004). However erythromycin is highly and exclusively active against all obtained stains of *Ureaplasma* (Huang et al., 2003; Nektaria et al., 2008).

The sequence of sensitivity of other 4 among 6 antimicrobial agents for *M. hominis* is lincomycin, minocycline, doxycycline and rosaramicin from highest to the lowest sensitivity, accordingly and these results are correlated with other studies as well (Wu et al., 2000; Huang et al., 2003). We concluded in our study that the obtained 11 strains of *Ureaplasma* were highly sensitive to erythromycin, rosaramicin, minocycline and doxycycline, while their resistance to lincomycin was also found as in other related studies to this effect (Huang et al., 2003; Ghaleh et al., 2008).

As all the strains of both species of *M. hominis* and *Ureaplasma* are resistant to tetracycline which could be due to the presence of tetracycline resistance gene tet (M) in their genome. Some researchers have found that high percentage of tet M-containing microorganisms in vagina, provide condition for tet M Gene transfer, a determinant gene of resistance in these *Mycoplasmas* cells (Taraskina et al., 2002). The changes in the spectrum of drug resistance could be due to inappropriate use of antibiotics. Thus, it is significantly important to offer the sensitivity test screening periodically and to use drugs rationally on the basis of regularly available sensitivity testing results. This is also very crucial for the early cure of patients and for the prevention of emergence of drug resistant strains.

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