ANTIFUNGAL SENSITIVITY AGAINST FUNGAL ISOLATES FROM BOVINE MASTITIS

K. Sukumar

Department of Veterinary Microbiology,
Veterinary College and Research Institute,
Namakkal- 637 002, Tamil Nadu, India

Received 20-3-2012 Accepted 5-8-2012

ABSTRACT

In vitro antifungal susceptibility of mould isolates showed that Clotrimazole was the drug of choice followed by Itraconazole, Ketoconazole and Pimaricin. Less sensitivity was shown against Amphotericin B and Nystatin. All the mould isolates showed resistance to Fluconazole and Griseofulvin.

KEYWORDS: Bovine Mastitis, Antifungal drugs

INTRODUCTION

Bacteria are the predominant causative agents of bovine mastitis but fungi especially yeast and yeast like fungal organisms are also being recognized as primary agent for this condition. Antibiotics used for intramamary therapy may become contaminated with fungi and can be considered as a source of udder infection. The incidence of mycotic mastitis appears to be increasing because of extensive, rather indiscriminate use of antibiotics for the treatment of mastitis (Gupta et al., 1981). The present study was undertaken to compare *in vitro* efficacy of different commercially available antifungal drugs against fungi isolated from bovine mastitis cases.

MATERIALS AND METHODS

Eight drugs namely Amphotericin- B, Clotrimazole, Fluconazole, Griseofulvin, Itraconazole, Ketoconazole, Nystatin and Pimaricin were used in this study. The drug sensitivity was studied by disc diffusion method as adopted by Bauer et al. (1966). The strengths used in each disc were Amphotericin - B (100 units), Clotrimazole (10µg), Fluconazole (10µg), Nystatin (100 units), Griseofulvin (250µg,125µg & 50µg), Itraconazole (0.3125µg, 0.0156µg & 0.0078µg), Ketoconazole (100µg, 80µg & 60µg) and Pimaricin (5µg, 2.5µg & 1.25µg).

The different antifungal discs were then placed on the medium suitably spaced with the help of sterile forceps. Yeasts and yeast like fungi viz. Candida tropicalis, *Candida parapsilosis, Candida guillermondii, Geotrichum candidum, Trichosporon cutaneum, Saccharomyces cerevisiae, Rhodotorula rubra* and *Torulopsis Sp.* inoculated plates were incubated at 37°C for 24 to 48 hours and the filamentous fungi *Penicillium Sp., Sepedonium Sp., Aspergillus ochraceous group, Cladosporium carrionii* and *Trichophyton verrucosum* inoculated plates were incubated at room temperature for 5 days. Inhibitory zones were read visually and measured with the help of scale. The inhibitory zone of 15 mm and above was marked as highly sensitive, 10-15mm as moderately sensitive, 5-10 mm as less sensitive and below 5 mm was declared as resistant as adopted by Shah et al. (1986).

RESULTS AND DISCUSSION

Twenty species of yeasts and yeast like fungi viz. *Candida tropicalis* (7 isolates), *Candida parapsilosis* (1 isolate), *Candida guillermondii* (1 isolate), *Geotrichum candidum* (4 isolates), *Trichosporon cutaneum* (3 isolates), *Saccharomyces cerevisiae* (1 isolate), *Rhodotorula rubra* (1 isolate) and *Torulopsis Sp.* (1 isolate) and mould isolates namely *Penicillium Sp.* (2 isolates), *Sepedonium Sp.* (1 isolate), *Aspergillus ochraceous* group (1 isolate), *Cladosporium carrionii* (1

isolate) and *Trichophyton verrucosum* (1 isolate) isolated from mastitic milk were used for sensitivity test.

In vitro antifungal susceptibility of yeast and yeast like fungal isolates showed 100% sensitivity to Amphotericin B (100 U), Clotrimazole (10µg), Nystatin (100 U) and Ketoconazole (100µg), 95% sensitive to Itraconazole (0.03125µg); 80% sensitive to Pimaricin (5µg) and 30% sensitive to Fluconazole (10µg). These isolates showed 100% resistance to Griseofulvin. High degree of sensitivity to Itraconazole, Ketoconazole and Pimaricin might be due to lowest MIC values of these drugs for fungi. The degree of sensitivity was found to decrease with the higher dilutions of these antifungal agents. Fluconazole, Griseofulvin had no effect on yeast and yeast like fungal organism even in higher concentrations. Similar findings were reported by Jand et al. (1978) and Bansal et al. (1989). Resistance to Griseofulvin might be due to distribution of this drug to dermal tissues (keratophilic), which is beneficial in the treatment of dermal fungal infections rather than the systemic infection.

In vitro antifungal susceptibility of yeast and yeast like fungal isolates showed that Itraconazole was the drug of choice followed by Ketoconazole, Pimaricin, Clotrimazole, Nystatin and Amphotericin-B. Mould isolates were 83.33% sensitive to Amphotericin B (100 U), Clotrimazole (10µg), Itraconazole (0.031µg) and Pimaricin (5µg); 50% sensitive to Ketoconazole (100µg); 33.33% to Nystatin (100 U); 100% resistant to Griseofulvin (250µg) and Fluconazole (10µg). Clotrimazole (10µg) and Itraconazole (0.031µg) were found to give high degree of inhibition followed by Pimaricin (5µg). Griseofulvin (250µg) failed to show antifungal activity against any of the organisms.

REFERENCES

Bansal, B.K, Singh K.B, Jand S.K, Nauriyal D.C and Randhawa S. S. (1989). In vitro drug sensitivity trails against Candida spp isolated from clinical cases of mastitis in dairy animals. Indian J. Comparative Microbiol. Immunol. Infectious Dis., **10** (4): 208-211.

Bauer, A.W, Kirby W.M.M , Sherris J. C and Turek M (1996). Antibiotic sensitivity testing by a standardized single disk method American. J. Clinical Pathol., **45**: 496.

Gupta, P. R. K, Kalra, D. S, Verma, P. C and Lodha, B C. (1981). Final Technical Report on studies on mycotic mastitis in domestic animals with particular reference to its incidence, Pathology and diagnosis. Final Technical Report, HAU, Hissar.

Jand, S.K, Singh, K.B and Narula, A S.(1978). In vitro trails of drug sensitivity against fungi. Indian Vet. J., **55** (10): 807-809.

Shah, N.M, Dholakia, P.M and Simaria, M. B. (1986). In vitro drug sensitivity trails against yeasts and moulds isolated from bovine udder. Indian Vet. J., **63** (1): 5-8.

❑