



ANTIBACTERIAL ACTIVITY OF FOUR ESSENTIAL OILS IN THE BROTH DILUTION TEST

R. Mickiene^{1,2}, A. C. Springorum², B. Bakutis¹, J. Schulz², J. Hartung²

¹ Department of Food Safety and Animal Hygiene, Lithuanian Veterinary Academy

E-mail: mickiene@lva.lt

² Institute for Animal Hygiene, Animal Welfare and Behaviour of Farm Animals, University of Veterinary Medicine Hannover, Germany

E-mail: itt@tiho-hannover.de

ABSTRACT

Recently essential oils sprays were recommended as a new approach to reduce airborne micro-organisms in animal houses. The antibacterial activity of such oils from *Mentha arvensis* (Mea), *Malaleuca alternifolia* (Ma), *Cymbopogon citratus* (Cc), *Zingibe officinales* (Cc) was tested on 2 Gram + (*S. aureus*, *E. faecium*), 3 Gram - (*Ps. aeruginosa*, *E. coli*, *P. mirabilis*) species and the yeast *C. albicans* using the broth dilution method. Standard bacterial broth cultures were mixed with different concentrations of essential oils in steps from 0.5 to 50 % in order to determine the lowest effective antibacterial concentration. The oils showed a very wide spectrum of antibacterial activity. Concentrations of 0.5 % of Ma reduced total bacterial counts of *E. coli* and *P. mirabilis* to below 1 %, 5 % inactivated *Ps. aeruginosa* and *E. faecium* for *Staph. Aureus* 8 % were necessary. High antibacterial activity was also revealed for Cc with bactericidal concentrations of 0.8 % (*E. coli*); 0.5 % (*P. mirabilis*); 0.2 % (*E. faecium*); 5 % (*Ps. aeruginosa*) and 8 % (*S. aureus*). *C. albicans* is highly sensitive for Ma (0.5 %), Cc (0.5 %) and Mea (0.8 %). It seems that essential oil-water mixtures may be a promising alternative to air disinfection. More experiments are necessary to verify the effectiveness and to assess occupational and animal health aspects.

Keywords: essential oil, disinfection, micro-organisms.

INTRODUCTION

Control of micro-organisms in indoor environments has traditionally focused on source control, ventilation, and air cleaning [5, 6]. Various industries are now searching for sources of alternative, more natural and environmentally friendly antimicrobials and antibiotics as protection agents. The antimicrobial effects of essential oils have been widely described in several studies [3, 1].

The aim of this study was to evaluate the sensitivity profile of 2 Gram +, 3 Gram - species, and the yeast *C. albicans* using the broth dilution method to some essential oils with emphasis for a possible future use of the ones as alternative to air disinfection.

MATERIALS AND METHODS

The antibacterial activity of such oils from *Mentha arvensis* (Mea), *Malaleuca alternifolia* (Ma), *Cymbopogon citratus* (Cc), *Zingibe officinales* (Cc), was tested on 2 Gram + (*S. aureus*, *E. faecium*), 3 Gram - (*Ps. aeruginosa*, *E. coli*, *P. mirabilis*) species and the yeast *C. albicans* using the broth dilution method.

Essential oils of *Mentha arvensis*, *Malaleuca alternifolia*, *Cymbopogon citratus*, *Zingibe officinales* were obtained from a commercial source (Germany) and used different concentrations in steps from 0.5 to 50 %.

Test organisms: *Staphylococcus aureus* DSM-No. 799, *Enterococcus faecium* DSM-No. 2918, *Pseudomonas aeruginosa* DSM-No. 939, *Escherichia coli*, *Proteus mirabilis* DSM-No. 788, *Candida albicans* DSM-No. 1386 were obtained from the Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH.

C. albicans were cultivated on Maltextract-Agar at 25 °C for 48 h. Bacteria were cultivated on Blutagar-Basis Nr. 237 °C for 24 h.

RESULTS

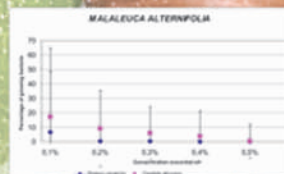


Figure 1. Effects of *Malaleuca alternifolia* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.

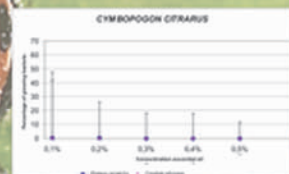


Figure 2. Effects of *Cymbopogon citratus* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.

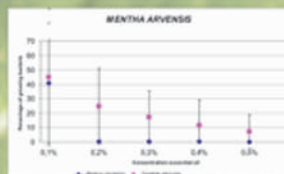


Figure 3. Effects of *Mentha arvensis* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.



Figure 4. Effects of *Malaleuca alternifolia* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.



Figure 5. Effects of *Cymbopogon citratus* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.

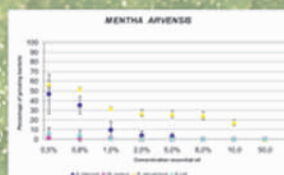


Figure 6. Effects of *Mentha arvensis* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.



Figure 7. Effects of *Zingibe officinales* on *E. faecium*, *Staph. aureus*, *Ps. aeruginosa*, *E. coli*.

DISCUSSION AND CONCLUSIONS

This study demonstrates that the four aroma chemicals - *Mentha arvensis*, *Malaleuca alternifolia*, *Cymbopogon citratus*, *Zingibe officinales* - definitely show antimicrobial effects on airborne bacteria, yeasts. Gram-positive bacteria are known to be more susceptible to essential oils than Gram-negative bacteria [2, 7]. The weak antibacterial activity against Gram-negative bacteria was ascribed to the presence of an outer membrane, which possessed hydrophilic polysaccharide chains as a barrier to hydrophobic essential oils [4, 8]. As a conclusion, it can be stated that the tested essential oils, can be used for air disinfection as an alternative to established air disinfectants.

ACKNOWLEDGEMENTS

Gratefully acknowledge to Prof. Dr. J. Hartung Institute of Animal Hygiene, Animal Welfare and Behaviour of Farm Animals, University of Veterinary Medicine Hannover, Germany.

REFERENCES

- Cox SD, Mann CM, Markham JL, et al. 2000. The mode of antimicrobial action of essential oil of *Malaleuca alternifolia* (tea tree oil). *Journal Applied Microbiology* 88:170 - 175.
- Farag R. S., Daw Z. Y., Hewedi F. M. & El-Baroty G. S. A., 1989. Antimicrobial activity of some Egyptian spice essential oils. *Journal of Food Protection* 52, 665 - 7.
- Kim J., Marshall MR., Cornell JA., et al. 1995. Antibacterial activity of carvacrol, citral, and geraniol against *Salmonella typhimurium* in culture medium and on fish tubes. *J. Food Scienc.* 60: 1364 - 1374.
- Mann C. M., Cox S. D. & Markham J. L., 2000. The outer membrane of *Pseudomonas aeruginosa* NCTC 6749 contributes to its tolerance to the essential oil of *Malaleuca alternifolia* (tea tree oil). *Letters in Applied Microbiology* 30, 294 - 7.
- Saleh M., Seedorf J., Hartung J., 2005. Influence of animal age and season on bio-aerosol concentrations in a broiler house. XIIth International Congress ISAH 2005 Warsaw, Poland. *Animals and Environment*. Vol.2: 37-40.
- Schulz J., Seedorf J., Hartung J., 2005. Estimation of a „Safe distance“ between a natural ventilated broiler house and a residential dwelling. XIIth International Congress ISAH 2005 Warsaw, Poland. *Animals and Environment*. Vol.2: 41-45.
- Smith-Palmer A., Stewart J. & Fyfe, L., 1998. Antimicrobial properties of plant essential oils and essences against five important food-borne pathogens. *Letters in Applied Microbiology* 26, 118-22.
- Tassou C.C. & Nychas G. J.E., 1995. Antimicrobial activity of the essential oil of Mastic gum (*Pistacia lentiscus* var. *chia*) on gram-positive and gram-negative bacteria in broth and model food systems. *International Biodegradation & Biodegradation* 36, 411-20.