MICROBIOLOGICAL CONTROL OF THE ENVIRONMENT IN AN INTENSIVE RABBIT REARING

MARTINO P.A.¹, LUZI F.², VERGA M.²

¹Sezione di Microbiologia e Immunologia, Dipartimento di Patologia Animale, Igiene e Sanità Pubblica Veterinaria, Università di Milano, Via Celoria, 10, 20133 Milano, Italy.
²Istituto di Zootecnica, Università di Milano, Via Celoria, 10, 20133 Milano, Italy.

piera.martino@unimi.it

ABSTRACT

The microbiological conditions of farm environment are very important to obtain good performance (e.g., daily live weight gains, feed conversion rate, meat guality, etc.), even if this parameter is often disregarded. Rabbits are particularly sensitive to environmental conditions and to stress due to changes in some parameters (e.g., temperature, relative humidity, etc.). Aim of this work is the evaluation of the number and kind of bacteria and fungi, using exposed plates with microbiological media, to which rabbits, bred in reproductive shed with forced ventilation, have been exposed. This work was performed in a meat rabbit farm located in the province of Bergamo (Lombardia region), in the North-West of Italy. Rabbits (commercial hybrids) are housed in two separate sheds with forced ventilation; one of them is dedicated to does and nests (reproduction sector) and the other one is for fattening rabbits. Metal cages are situated, for the 1st shed, on one level; on the contrary, for the 2nd on two levels (California systems). The environmental samples were collected using "opened plates", during springtime (March-June) in doe's shed and nests. *Micrococcus luteus* and *Staphylococcus* spp. were the most frequently isolated bacteria and among veasts Rhodotorula rubra, with a total charge always more than 50 UFC/plate. Among environmental fungi there is an important differentiation; beside Aspergillus spp., Penicillium spp. and Alternaria, there are Fusarium, Saksenaea (a Mucorales species) and Curvularia. For dermatophytes, high prevalence of Trichophyton mentagrophytes has been observed (from 50% to 70%): this microorganism is typical of rabbit farms and, like Microsporum canis, it's a zoonosic agent. The data reported in this paper demonstrate that a good environmental control, particularly a microbiological control, can be very useful for maintaining good health status in farm animals.

Key words: rabbit, rearing, environment, microbiological control

INTRODUCTION

The environmental conditions play a pivotal role in maintaining the health of the animals in intensive husbandry system; particularly rabbits are very sensitive to environmental changes (e.g., water, pH, temperature, relative humidity, concentration of NH₃ and CO₂,

etc.) (CRIMELLA et al., 1995; JACCHIA and MARTINO, 2000). These stressing environmental parameters can cause the spreading of pathogens (e.g., bacteria, viruses, fungi, parasites) and consequently high economic losses (HAFFAR and CHERMETTE, 1989; QUINN and CARTER, 1994; POLI and COCILOVO, 1996). Among bacteria, Staphylococcus aureus, Pasteurella multocida, Pseudomonas aeruginosa, other Gram-negative bacteria and, finally, enteropathogenic Escherichia coli (EPEC) (photo 1) are responsible for many diseases (pododermatitis, mastitis, respiratory and enteric infections) (Licois et al., 2003). Aspergillus, Penicillium, Alternaria, Mucorales (photo 2) are typical environmental fungi that are able to produce a high number of spores that are released in the air and that can cause respiratory diseases (above all Aspergillus species) (Pitt, 1994). Also, the presence in the rabbit farms of dermatophytes (such as Microsporum spp. or Trichophyton spp.), fungi with special tropism for cutaneous adnexa (e.g., skin, nail, coat/hair), are very important due to the fact that they are well-known zoonosic agents (photos 3-4-5) (ZAROR and CASAS, 1988; HAFFAR and CHERMETTE, 1989; POLI and COCILOVO, 1996; MARTINO et al., 1998; JACCHIA and MARTINO, 2000; VANGEEL et al., 2000). In detail, the factors or changes that could induce the development of dermatophytosis in rabbits reared in intensive husbandry system are numerous: fluctuations in relative humidity (>70%) and in temperature (>25°C); overcrowding, perturbation of homeostasis in normal cutaneous microbial flora (resident bacteria), the age of the animals and "poor" diet (e.g., lack of crude fibre with the appearance of alopecic areas) (JACCHIA and MARTINO, 2000). The aim of this work is the evaluation of the bacterial and fungal flora in an intensive rabbit farm. We evaluate the number and kind of bacteria and fungi in this environment with special attention to dermatophytes.



Photo 1. *Escherichia coli* on Mac Conkey Agar.



Photo 3. Microsporum canis.



Photo 2. Penicillium spp.



Photo 4. Trichophyton mentagrophytes.



Photo 5. Trichophyton mentagrophytes (electron microscope image). (Font: J.M. Rosell)

MATERIAL AND METHODS

Animal and environmental conditions

This work has been performed in a meat rabbit farm located in the province of Bergamo (Lombardia region), in the North-West of Italy. Rabbits (commercial hybrids) are located in two separate sheds with forced ventilation; one of it is dedicated to does and nests (reproduction sector) and the other one is for fattening rabbits. Metal cages are situated, for the 1st shed, on one level; on the contrary, for the 2^{nd} on two levels (California systems).

Microbiological test

The environmental samples were collected using "opened plates", during springtime (March-June) in doe's shed and nests. In detail, we have used "Surfair plate" (PBF, Italy) filled with *Tryptic Soy Agar* (Oxoid[®], Italy) for total bacterial count; *Sabouraud Dextrose Agar* (Oxoid[®], Italy) for environmental fungi and *Dermasel Agar* (Oxoid[®], Italy) for dermatophytes. All the plates exposed for 10 min, by opening the Petri dishes in doe's shed and by using a microbial air sampler (SAS[®], PBI, Italy) in nests, have been kept at 4 °C until they were taken to the laboratory.

Then, they have been incubated at 37° C (TSA) and at 25° C (SAB and Dermasel Agar); the reading of the plates has been made for bacteria after 18-24 h, for fungi after 72 h and for dermatophytes after 5-7 days. The bacterial identification has been performed using selective and differential cultural media and miniaturized biochemical system (API System, BioMerieux[®]) until genus and species definition. For fungi (environmental and *dermatophytes*) the identification has been performed with macroscopic evaluation of the "recto" and "verso" of fungal colonies on media and, microscopically, staining the sample with lactophenol-blue cotton and observing the macro- and microconidia and/or *hypha*e (40 x) (photo 6).



Photo 6. Macroconidia of *M. canis* (on the left); microconidia of *T. mentagrophytes* (on the right).

RESULTS AND DISCUSSION

Micrococcus luteus and *Staphylococcus* spp. were the most frequently isolated bacteria and among yeasts *Rhodotorula rubra*, with a total charge always more than 50 UFC/plate (on average). Among environmental fungi, there is an important differentiation; beside *Aspergillus* spp., *Penicillium* spp. and *Alternaria*, there are *Fusarium*, *Saksenaea* (a *Mucorales* species) and *Curvularia* (Figure 1). For dermatophytes, high prevalence of *Trichophyton mentagrophytes* is observed (from 50% to 70%); this microorganism is typical of rabbit farms and, like *Microsporum canis*, it's a zoonosic agent. We didn't observe the presence of *Microsporum* spp. (e.g., *M. canis, M. gypseum*, etc.) (Figure 1).



Figure 1. Frequence of environmental fungi and dermatophytes during the trial's period (month evolution expressed for each microorganisms).

Regarding the distribution of the environmental fungi during the four months (March, April, May and June) it's possible to observe that there is no particular progress, on the contrary, the various kind of fungi are present alternatively in the different periods. (Figure 2) As regards as the distribution of the dermatophytes, the presence of *Trichophyton mentagrophytes* is always very high and only in June we can observe a fall in the collected data (Figure 3).



Figure 2. Reproduction shed – Environmental fungi isolated during trials months.



Figure 3. Reproduction shed – Dermatophytes isolated during trials months.

A probably explanation could be connected to the environmental temperature (average values of the trial period: temperature 22°C, relative humidity 70%) because in March, April and May, there are good conditions for an optimal growth of these fungi (20-25°C). On the contrary, in June with an increase of the temperature and a decrease in the relative humidity in the shed, we can observe a decrease of their development.

The isolation of *T. mentagrophytes*, in the reproduction shed, could be explained with the presence of does "healthy carriers" and to the diffusion, in the air, of infected coat and of fungal spores, in spite of the periodical prophylactic treatments (disinfection).

CONCLUSIONS

The air sampling, using exposed plates with microbiological media, is a simple practice for the isolation and the identification of bacteria and fungi. Particularly the identification of dermatophytes has a great importance for animal welfare and its productivity, but, above all, for human health, because these microorganisms could have a zoonosic potential and the risk of transmission could be very high.

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