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# Demonstration of antibodies to *Encephalitozoon cuniculi* in rabbits on some Slovak farms

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#### Summary

Rabbits from three farms were examined for seropositivity to *Encephalitozoon cuniculi*. Thirty seven rabbits were included in this experiment. The indirect immunofluorescent antibody test was performed. The serum of all rabbits from the laboratory colony reacted positively to antigen. Serum from animals of amateur breeders reacted negatively.

Key words: *Encephalitozoon cuniculi*; rabbit; immunofluorescence

#### Introduction

Encephalitozoonosis is a chronic, usually latent disease caused by the intracellular protozoon *Encephalitozoon cuniculi* (syn. *Nosema cuniculi*). The parasite can infect rabbits and other mammals (P a k e r s and G e rr i t y, 1994). Occasionally the clinical form of the disease and mortality can cause certain economic losses in some rabbit farms (S o m w a n s h i *et al.*, 1994).

Several serological tests have been developed for the diagnosis of encephalitozoonosis, including indirect fluorescence antibody techniques (IFAT) (C h a l u p s k ý *et al.*, 1971; 1973). The test has been used to screen large colonies of rabbits.

Rabbits of one colony had been showing sporadically clinical neurologic signs such as torticollis. Histologically, the granulomas in the cortex and medulla were seen, even in rabbits without neurological signs. These observations have prompted us to begin with a seroepidemiological study in Slovakia.

#### **Material and Methods**

The blood of rabbits was obtained by aspiration from the ear vessels. Sera obtained from the blood samples were frozen and maintained at -20°C until use.

The indirect immunofluorescence antibody test was performed according to the method described in detail by C h a l u p s k ý *et al.* (1971, 1973), using conjugate, specific to the rabbit. The serum and the conjugate were applied to the antigen for 30 min at 37 °C. Purified spores of *Encephalitozoon cuniculi* obtained from infected monkey kidney cell cultures were used as an antigen. The infected cell culture was kindly provided by C h a l u p s k ý (Charles University, Prague).

#### Results

Rabbits from three farms were examined. The animals whose sera reacted at a dilution of 1: 64 or higher, were considered to be positive (Fig. 1). The results obtained by examining the rabbits are summarized in Table 1. The samples of six rabbits reacted at a dilution of 1: 512, the sera of eight rabbits were antigen positive at a dilution of 1: 256, and the sera of three animals showed positive reaction at a dilution of 1: 128. Seropositive and seronegative animals were clinically healthy. The animals examined were of 1.5 to 3.5 years old.



Fig. 1. Positive reaction of examined rabbit at a dilution 1 : 265 (IFAT; x 1000)

# Tab. 1. Occurrence of the *Encephalitozoon cuniculi* seropositive rabbits

Colony coding	Colony type	No. of examined animal	No. of positively reacting animal	% of positivity
ZT	laboratory colony	17	17	100
TVB	amateur breeder	5	0	0
TVP	amateur breeder	5	0	0

#### Discussion

The results of the preliminary serological examination showed the occurrence of antibodies to *Encephalitozoon cuniculi* in rabbits in Slovakia. Sera showing positivity at a dilution of 1 : 64 and higher were chosen on the basis of C h a l u p s k ý *et al.* (1979) previous experience. Some neurological signs and granulomas sporadically observed earlier in the CNS confirmed the infection of this microsporidian in one colony examined. This study shows the presence of microorganisms in one colony, and an absence in the other. It is interesting that both negative colonies are kept by amateur breeders. These colonies are also numerous. Difference in the handling of the animals, and the spatial arrangement of the cages etc. might be important in the transmission of this disease (C h a l u p s k ý *et al.*, 1979).

Animals from the laboratory colony are used for scientific research. That is why experimental results may actually be altered because of modulation of the animals' immune system. The results can be miss-interpreted.

All animals examined were adults. In the case of the examination of young animals, the number of positively reacting animals can be different. The results of the experiments strongly suggest a horizontal transmission by various parenteral routes, oral, and rectal administration (N e I s o n, 1967; F u e n t e a I b a *et al.*, 1992). Vertical transmission of *Encephalitozoon* is strongly sus-

pected (N e l s o n, 1967), but few other reports refute this aspect (W i l s o n, 1986).

Our observation confirmed that encephalitozoonosis is a focal disease, endemic in some colonies. Further investigation will establish the spread of microsporidian parasites in rabbits in Slovakia.

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