

## THE ISOLATION OF *TOXOPLASMA GONDII* FROM THE SALIVARY GLAND OF *CLETHRIONOMYS GLAREOLUS*

Although numerous reports are available on isolations of the protozoan *Toxoplasma gondii*, on the finding of toxoplasma organisms in smears of various organs or on the identification of toxoplasmic antibodies in sera of different species of small rodents, the occurrence of toxoplasmosis in *Clethrionomys glareolus* Sehr. has received little attention.

Relatively many examinations of this rodent species have been carried out in the U.S.S.R. (LEVIT et al., Toxoplasmosis of animals, Nauka, Alma-Ata, pp. 238—273, 1965, in Russian). Of a total of 142 animals examined serologically with the complement fixation test, 5 animals reacted positively; also in smears of their organs, toxoplasma organisms were repeatedly found. DUNAYEVA et al. (Zool. zhurn. 52: 629—631, 1963) first isolated a strain of *T. gondii* from the bank vole caught in the forest zone near the town Tula. In Poland, UMIŃSKI et al. (Wiadom. parazytol. 10: 4—5, 1964) examined 47 bank voles with the complement fixation test without obtaining positive results. Negative results were obtained also by DOBY et al. (Čs. parazitol. 12: 133—144, 1965), who examined histologically the brains of 106 bank voles from various regions of France.

In Czechoslovakia, a total of 68 specimens of *Clethrionomys glareolus* were examined with the Sabin Feldman test (SFT); of these 6 reacted positively in titers of 1:4—1:16 (HAVLÍK O., HÜBNER J., Čs. Epidem. 9: 391—397, 1960). ŠEBEK (Folia zool. 11: 355—366, 1962) and ČATÁR (Bratisl. lek. Listy 47: 226—234, 1967) examining under the microscope impression smears of different organs of 485 bank voles obtained negative results only.

In November 1968, 8 specimens of *Clethrionomys glareolus* and 28 specimens of *Apodemus sylvaticus* were caught in the vicinity of the Hydrobiological station at Blatná in southern Bohemia. Blood was taken from the retroorbital venous plexus of all animals and examined serologically for toxoplasmosis with the technique of microprecipitation in agar gel (MPA) (HÜBNER J., UHLÍKOVÁ M., J. Hyg. Epidem. — in press 1969). The serum of one specimen of

*Clethrionomys glareolus* reacted positively showing lines of medium intensity. The remaining 40 animals were MPA negative, the positively reacting animal was unfortunately killed and its organs were frozen at  $-40^{\circ}\text{C}$  to be used for virological examination. Only a small portion of the salivary gland and of brown fat, kept after preparation in saline at  $-4^{\circ}\text{C}$  for more than 24 hrs after dissection, could be used for isolation experiments. Both organs were homogenized by hand in a glass mortar and then 2 ml of saline and PNC—STM were added to each crushed organ. Two groups of two mice per group received intraperitoneally 1 ml of mixture respectively. Fresh preparations were made from the homogenates of both organs and examined. In the tissue of the salivary gland we discovered 3 round cysts (size 15  $\mu$ ) resembling those of toxoplasma organisms.

The mice were examined serologically one month after the inoculation; both mice inoculated with the homogenate of brown fat were MPA and SFT negative and so was one of the mice of the group inoculated with the tissue of the salivary gland. The second mouse of this group, however, reacted positively to both MPA (one line of medium intensity) and to the SFT. The mouse was killed 50 days after inoculation; during this period there was no evidence of illness. The microscopical examination of the brain, however, revealed numerous cysts of *Toxoplasma gondii*.

The successful isolation of toxoplasma organisms from the salivary gland of the bank vole supports the assumption that toxoplasma organisms are expelled with the excrements, in this case with the saliva. This may occur when the cysts are liberated or are decomposing and the pathogenic agent is carried with the saliva to the oral cavity of the infected animal.

This is the first report on the isolation of *Toxoplasma gondii* from the bank vole in Czechoslovakia.

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