

Rickettsioses studies.

3. Natural foci of rickettsioses in south Bohemia

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Antibodies against Coxiella burnetii and against rickettsiae of the spotted fever group were found in human sera and in sera from domestic and wild animals collected in south Bohemia. Spotted fever group rickettsiae were also discovered in the tick Ixodes ricinus. These results indicate the presence of both types of rickettsiae in this part of Czechoslovakia. As no epizootics or epidemics of Q fever have as yet been reported in the area, it can be assumed that C. burnetii occurs in the latent state. The occurrence of spotted fever group rickettsiae is probably endemic among I. ricinus ticks and among small and larger wild mammals.

Several sporadic cases and epidemics of Q fever occurred in central and west Bohemia in 1952–54 (5, 7, 9, 10) and later in north and north-east Bohemia (4, 8). The presence of *Coxiella burnetii* was further confirmed in east Bohemia by detecting this rickettsia in *Sorex araneus* and the tick *Ixodes ricinus* (15), and by the existence of rickettsial antibodies in the pheasant and the roebuck (16). Since that time no data have been published on the occurrence of Q fever in Bohemia or Moravia.

When the role of small mammals as reservoirs of anthroponoses was studied in south Bohemia, antibodies against *C. burnetii* were found in one *Clethrionomys glareolus* in 1968, in one *C. glareolus*, and in four small rodents (*Apodemus* spp.) in 1969. To obtain more information, animals collected in three districts of Bohemia and two districts of Moravia were screened for the presence of antibodies against *C. burnetii* and against spotted fever (SF) group rickettsiae. For comparison, the same tests were conducted in one known focus of rickettsioses in Slovakia. This investigation revealed the presence of both rickettsiae in all three territories of

Czechoslovakia, the proportions of positive sera being very similar, as shown in Table 1. These findings, together with reports on rickettsioses in some countries adjacent to Bohemia, e.g., in Austria (17) and the Federal Republic of Germany (6), led us to investigate the ecology of rickettsioses in Bohemia more closely. Since 1972, we have made a thorough search for rickettsioses in south Bohemia by serological examination of wild mammals, domestic animals, and human subjects for the presence of rickettsial antibodies, and by testing for the existence of rickettsiae in ticks.

COLLECTION OF MATERIALS

Although serological examination of man, cattle, and hares was in some cases extended to adjacent areas, most of the localities studied were situated in the Českobudějovická depression. These areas are low-lying, with many ponds, and are characterized by lower average temperatures and substantially higher rainfall than other parts of Bohemia and Slovakia. They also share peculiarities of flora and fauna, including those species that are either possible reservoirs or vectors of rickettsiae. For example, there is an absence of the species of *Haemaphysalis* and *Dermacentor* that occur in south Moravia (*H. concinna* and *D. reticulatus*) and in Slovakia (*H. punctata*, *H. inermis*, and *D. marginatus*). Among small mammals, *Apodemus agrarius* does not occur in south Bohemia but it is abundant in east Slovakia, and members of the genus *Crociodura* are also very rare.

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Table 1. Evidence of antibodies to *C. burnetii* and SF group rickettsiae in small mammals collected in Czechoslovakia in 1971

| Territory | District | Locality | Date of collection | Species | No. of animals tested | No. of sera with antibodies to | |
|-------------------|---------------|------------------------|--------------------|-----------------------|-----------------------|--------------------------------|----------|
| | | | | | | <i>C. burnetii</i> | SF group |
| Bohemia | Turnov | Sedmihorky | 7 July | <i>A. flavicollis</i> | 2 | 1 | — |
| | | | | <i>C. glareolus</i> | 9 | — | 2 |
| | | | | <i>S. araneus</i> | 1 | 1 | — |
| | Beroun | Srbsko | 15 July | <i>M. arvalis</i> | 5 | — | 2 |
| | | | | <i>A. flavicollis</i> | 6 | — | — |
| | | | | <i>C. glareolus</i> | 2 | — | — |
| | Č. Budějovice | Borek | 16 July | <i>M. arvalis</i> | 2 | — | 1 |
| | | | | <i>A. flavicollis</i> | 4 | — | — |
| | | | | <i>C. glareolus</i> | 19 | — | 11 |
| <i>S. araneus</i> | | | | 6 | — | — | |
| Total | | | | 56 | 2 (3.6%) | 16 (28.6%) | |
| Moravia | Blansko | D. Lhota | 13 July | <i>A. flavicollis</i> | 9 | — | — |
| | | | | <i>C. glareolus</i> | 27 | 1 | 5 |
| | | | | <i>S. araneus</i> | 1 | 1 | — |
| | Znojmo | Znojmo | 17 July | <i>A. flavicollis</i> | 3 | — | — |
| | | | | <i>C. glareolus</i> | 5 | — | 3 |
| Total | | | | 45 | 2 (4.4%) | 8 (17.8%) | |
| Slovakia | V. Krtíš | Senné, Šula, Milotínce | 8–10 July | <i>M. arvalis</i> | 4 | — | 1 |
| | | | | <i>A. flavicollis</i> | 16 | — | 4 |
| | | | | <i>C. glareolus</i> | 6 | — | 2 |
| | | | | <i>Crocidura</i> spp. | 2 | — | — |
| | | | | <i>S. araneus</i> | 3 | 1 | 2 |
| | | | | <i>S. minutus</i> | 3 | — | — |
| Total | | | | 34 | 1 (2.9%) | 9 (26.5%) | |

Ticks were collected by trailing a white woollen blanket over the low vegetation. Small mammals were captured in special traps (2) and blood samples for serological examination were taken from the orbital sinus. Some animals were dissected and their organs (brain, liver, spleen, and kidneys) were aseptically removed and stored at -35°C for isolation experiments. The sera of larger wild mammals (hares and foxes), which were hunted and shot, were obtained from blood samples taken by cardiac puncture. The sera of domestic animals were supplied by the Central State Veterinary Institute in České Budějovice, and human sera by the regional hygiene and epidemiology centre in České Budějovice.

LABORATORY STUDIES

Detection of rickettsiae in ticks, and isolation experiments

Unfed ticks were examined for infection with rickettsiae, using the haemocyte test (11) with Giménez staining (3), and the indirect immunofluores-

cence technique with rabbit immune sera. The isolation experiments were carried out only on ticks that were positive for rickettsiae in the haemocyte test. Suspensions of ticks containing rickettsiae were inoculated into the yolk sacs of 7-day-old chick embryos. The presence of rickettsiae in ticks was further confirmed by antibody responses in white mice inoculated with tick suspensions.

The experiments on isolation from organs of small mammals were carried out with groups of 5 animals of each species, each group collected in the same locality. The method was that used in the study of natural foci of rickettsioses in east Slovakia (14).

Serological examination

All human and animal sera were examined using the complement fixation test with soluble antigen of *Rickettsia slovaca*, strain B (1), and the microagglutination test with stained phase-II corpuscular *C. burnetii* antigen. Both antigens were prepared in the Department of Rickettsiae, Institute of Virology, Slovak Academy of Sciences, Bratislava. Sera with antibody titres $\geq 1:8$ were considered positive.

Table 2. Infection of *I. ricinus* ticks collected in April 1975 in south Bohemia with SF group rickettsiae

| District | Locality | No. of ticks investigated | | No. of ticks containing rickettsiae as detected by | | | |
|---------------|------------------|---------------------------|-------|--|-----------|------------------------------|-----------|
| | | | | Giménez staining | | immunofluorescence technique | |
| | | females | males | females | males | females | males |
| Č. Budějovice | Chlumeč | 86 | 87 | 3 | 1 | 2 | not done |
| | Hluboká | 118 | 145 | 2 | 3 | 1 | not done |
| | Češňovice | 120 | 139 | 1 | 2 | 1 | 2 |
| | Čejkovice | 185 | 131 | 1 | 4 | 1 | 4 |
| | Němčice | 97 | 90 | 1 | 4 | 1 | 3 |
| Prachatice | Žitná | 127 | 112 | 1 | 3 | 1 | 1 |
| Total | sexes separately | 733 | 704 | 9 (1.29%) | 17 (2.4%) | 7 (1.0%) | 10 (1.4%) |
| | both sexes | 1437 | | 26 (1.8%) | | 17 (1.2%) | |

RESULTS

Detection of rickettsiae in ticks

Examination of *I. ricinus* ticks caught in south Bohemia in 1975 for infestation with rickettsiae gave positive results as evidenced by both tests. Table 2 shows that out of 1437 ticks examined, 26 (1.8%) contained rickettsiae according to Giménez staining, among them 9 females (1.29%) and 17 males (2.4%); by the immunofluorescence technique, 17 (1.2%) were found to be positive, of which 7 were females (1.0%) and 10 males (1.4%): all these rickettsiae belonged to the SF group. However, rickettsiae could not be isolated in the yolk sacs of chick

embryos, nor could specific rickettsial antibodies be found in white mice inoculated with tick suspensions.

Detection of rickettsiae in small wild mammals

As shown in Table 3, serological examination of 342 small wild mammals collected in 1974-76 revealed the presence of antibodies against *C. burnetii* in 1 *Mus musculus* (0.03%) and against SF group rickettsiae in 28 animals (8.2%) belonging to all the species tested. Most frequently found in the area were rodents of the species *Apodemus*; of 202 sera collected from these animals, 14 (6.9%) had antibodies against SF group rickettsiae. However, the

Table 3. Antibodies to *C. burnetii* and SF group rickettsiae in sera of small mammals collected in south Bohemia in 1974-76

| District | Locality | Date of collection | Species | No. of animals tested | No. of sera with antibodies to | |
|---------------|---------------------------------------|----------------------|---------------------|-----------------------|--------------------------------|-----------|
| | | | | | <i>C. burnetii</i> | SF group |
| Č. Budějovice | Němčice, Čejkovice, M. Chrást'any | 28-31 Aug. 1974 | <i>M. arvalis</i> | 2 | — | — |
| | | | <i>M. agrestis</i> | 5 | — | — |
| | | | <i>Apodemus</i> sp. | 97 | — | 7 |
| | | | <i>C. glareolus</i> | 18 | — | 5 |
| | | | <i>Mus musculus</i> | 72 | 1 | 1 |
| | Němčice, Čejkovice, Břehov, Češňovice | 29 Sept.—2 Oct. 1975 | <i>M. agrestis</i> | 3 | — | — |
| | | <i>Apodemus</i> sp. | 54 | — | 4 | |
| | | <i>Mus musculus</i> | 6 | — | — | |
| Prachatice | Žitné | 30 Aug. 1974 | <i>M. arvalis</i> | 9 | — | 3 |
| | | | <i>Apodemus</i> sp. | 34 | — | 1 |
| | | | <i>C. glareolus</i> | 5 | — | 2 |
| Č. Krumlov | Křemže (15 km radius) | 30 June—9 July 1976 | <i>M. arvalis</i> | 9 | — | 2 |
| | | | <i>M. agrestis</i> | 3 | — | — |
| | | | <i>Apodemus</i> sp. | 17 | — | 2 |
| | | | <i>C. glareolus</i> | 4 | — | 1 |
| | | | <i>S. araneus</i> | 4 | — | — |
| Total | | | | 342 | 1 (0.3%) | 28 (8.2%) |

Table 4. Antibodies to *C. burnetii* and SF group rickettsiae in sera of hares collected in south Bohemia in 1975

| District | Locality | No. of animals tested | Sera with antibodies to: | | | | | | | | | |
|---------------|------------|-----------------------|------------------------------|--------|-----|------|----------|------------------------------|--------|---|---|---|
| | | | <i>C. burnetii</i> | | | | SF group | | | | | |
| | | | No. and (%) of sera positive | Titres | | | | No. and (%) of sera positive | Titres | | | |
| 1:8 | 1:16 | 1:32 | | 1:64 | 1:8 | 1:16 | 1:32 | | 1:64 | | | |
| Č. Budějovice | Břehov | 37 | 3 (8.1) | 1 | 2 | — | — | 1 (2.7) | — | 1 | — | — |
| | Holašovice | 31 | 2 (6.5) | 1 | — | — | 1 | 4 (12.9) | 3 | — | 1 | — |
| | Žabovřesky | 47 | 3 (6.4) | 2 | — | 1 | — | — | — | — | — | — |
| | Zliv | 109 | 2 (1.8) | — | 1 | 1 | — | 3 (2.8) | 2 | — | — | 1 |
| Tábor | Měšice | 39 | 4 (10.3) | — | — | 3 | 1 | — | — | — | — | — |
| Total | | 263 | 14 (5.3) | 4 | 3 | 5 | 2 | 10 (3.8) | 5 | 1 | 1 | 1 |

highest prevalence of antibodies against rickettsiae of the SF group was noted in *Clethrionomys glareolus* and *Microtus arvalis*, namely in 8 (29.6%) out of 27 and in 5 (25%) out of 20 animals, respectively. Though antibody titres of 1:32 and 1:64 were found in two individual *Apodemus* sp., suggesting their recent infection with SF group rickettsiae, no rickettsiae could be isolated from the organs of small mammals. Apart from these two animals, all the antibody titres were 1:8 or 1:16.

Serological examination of larger wild mammals

Interesting results were obtained by examination of sera from 263 hares collected in 6 different localities in the winter of 1974–75: antibodies against *C. burnetii* were found in 14 (5.3%) and against SF group rickettsiae in 10 (3.8%) (Table 4).

Antibodies against *C. burnetii* with a titre of 1:64 were also found in 1 fox trapped in December 1974.

Serological examination of domestic animals

Sera from cattle, sheep, and horses were examined. Of 1329 cattle sera, 17 (1.3%) contained antibodies against *C. burnetii* and 87 (6.5%) against SF group rickettsiae. In the sera from 139 horses, antibodies against *C. burnetii* were found in 2 (1.4%) and against SF group rickettsiae in 4 (2.8%). Sheep sera also contained antibodies against both types of rickettsiae; of 52 sera tested only 1 (1.9%) had antibodies against *C. burnetii*, but 19 (36.5%) had antibodies against SF group rickettsiae (Table 5).

Serological examination of the human population

Of a total of 1086 human sera collected in 1975 and 1976 (see Table 6), antibodies against *C. burnetii*

were found in 19 (1.7%) and against SF group rickettsiae in 15 (1.4%).

DISCUSSION

Our investigation shows that the prevalence of antibodies against *C. burnetii* was similar in human and animal sera collected in south Bohemia. Even though a limited sample was examined, it was sufficient to conclude that *C. burnetii* was most probably transmitted by direct contact among animals and by human contact with infected animals or their excrement.

The 1086 human cases tested comprised 312 men and 774 women, the proportions of positive sera being 3.5% and 1.0%, respectively. Of those whose sera were found positive, none remembered having received a tick bite. Anamnestic data could be taken into account in only 4 patients who were suffering from an influenza-like disease, with chronic bronchitis. The antibodies against *C. burnetii* found in the remaining persons could hardly be connected with Q fever, because they were either hospitalized (in urological, dermatological, or dental wards) in the regional hospital, or had undergone general medical examinations that had revealed nothing unusual. No connexion could be discovered between the presence of antibodies against *C. burnetii* and possible exposure to Q fever in nature or at the workplace, because persons with Q fever antibodies had such different occupations as driver, engine-driver, crane-driver, general labourer, and government official. Four of them were pensioners, but none of them was a farmer, as might have been expected.

Of considerable interest is the comparatively high

Table 5. Antibodies to *C. burnetii* and SF group rickettsiae in sera of domestic animals collected in south Bohemia in 1973-76

| District | No. of localities tested | Date of collection | Animal species | No. of sera tested | Sera with antibodies to: | | | | | | | | | |
|---------------|--------------------------|--------------------|----------------|--------------------|------------------------------|--------|------|------|------------------------------|----------|----|---|---|---|
| | | | | | <i>C. burnetii</i> | | | | | SF group | | | | |
| | | | | | No. and (%) of sera positive | Titres | | | No. and (%) of sera positive | Titres | | | | |
| | 1:8 | 1:16 | 1:32 | | 1:8 | 1:16 | 1:32 | 1:64 | 1:128 | | | | | |
| Č. Budějovice | 7 | 1973-76 | cattle | 573 | 11 (1.9) | 9 | 2 | — | 69 (12.0) | 40 | 19 | 8 | 1 | 1 |
| | 1 | 1973-76 | horses | 139 | 2 (1.4) | 1 | 1 | — | 4 (2.8) | 4 | — | — | — | — |
| | 1 | 1976 | sheep | 52 | 1 (1.9) | 1 | — | — | 19 (36.5) | 11 | 6 | 2 | — | — |
| Prachatice | 1 | 1976 | cattle | 133 | 3 (2.3) | 1 | — | — | 10 (7.5) | 5 | 1 | 2 | 2 | — |
| Soběslav | 2 | 1976 | cattle | 67 | — | — | — | — | — | — | — | — | — | — |
| Milevsko | 4 | 1976 | cattle | 469 | — | — | — | — | 4 (0.9) | 1 | 2 | 1 | — | — |
| Pacov | 2 | 1975-76 | cattle | 65 | 3 (4.6) | 1 | 1 | 1 | 4 (6.2) | 4 | — | — | — | — |
| Tachov | 1 | 1976 | cattle | 22 | — | — | — | — | — | — | — | — | — | — |
| Total | | | | 1520 | 20 (1.3) | | | | 110 (7.2) | | | | | |

Table 6. Antibodies to *C. burnetii* and SF group rickettsiae in human sera in south Bohemia in 1975-76

| Sex | No. of sera tested | Sera with antibodies to: | | | | | | | | | |
|--------|--------------------|------------------------------|--------|------|------|------------------------------|----------|----------|------|------|---|
| | | <i>C. burnetii</i> | | | | | SF group | | | | |
| | | No. and (%) of sera positive | Titres | | | No. and (%) of sera positive | Titres | | | | |
| | | 1:8 | 1:16 | 1:32 | 1:64 | 1:128 | | 1:8 | 1:16 | 1:32 | |
| Male | 312 | 11 (3.5) | 6 | 1 | 2 | — | 2 | 2 (0.6) | — | 2 | — |
| Female | 774 | 8 (1.0) | 6 | 1 | 1 | — | — | 13 (1.7) | 10 | 2 | 1 |
| Total | 1086 | 19 (1.7) | 12 | 2 | 3 | — | 2 | 15 (1.4) | 10 | 4 | 1 |

prevalence of antibodies against *C. burnetii* in hares, in contrast with a low prevalence of antibodies against SF group rickettsiae. Previous research revealed no antibodies against *C. burnetii* in hares shot throughout Slovakia, and often directly in natural foci of rickettsioses, whereas sera of those shot in central Slovakia contained a high proportion of antibodies against SF group rickettsiae (12).

The transmission of *C. burnetii* in south Bohemia remains problematical. The possibility of mutual transmission between cattle and hares on the pastures cannot be taken into account at present, because cattle are not pastured in this area. The examination of ticks for infestation with *C. burnetii* also showed negative results. In view of this, it will be necessary to make further studies of the role of other ectoparasites in transmission of *C. burnetii*.

The finding of antibodies against *C. burnetii* in a fox is not surprising, because foxes are often infected with *C. burnetii* (12). They may serve as a good indicator of the circulation of *C. burnetii* in nature. The prevalence of rickettsiae in wild carnivora has always been high, owing to the many possibilities for infection. This could occur from a bite by an ectoparasite, from vegetation, or from eating infected prey, as well as through direct contact with *C. burnetii* in pastures, nests of reservoir animals, and in farm buildings.

At first sight the comparatively low prevalence of antibodies against *C. burnetii* and rickettsiae of the SF group in human beings and domestic animals is surprising. This can be related to the low infestation of the ticks with rickettsiae. Indeed, SF group rickettsiae were detected in *I. ricinus* ticks, collected

in south Bohemia, in a far lower proportion than in *D. marginatus* ticks collected in Slovakia (12) or in *I. ricinus* ticks, collected in the surroundings of the town of Bratislava (Řeháček, unpublished data). On the other hand, wild rodents in south Bohemia contained antibodies against SF group rickettsiae in proportions similar to those of wild rodents trapped in Slovakia. It is quite possible that ectoparasites

other than *I. ricinus* ticks (e.g., fleas and mites) may serve as vectors of SF group rickettsiae in south Bohemia, as postulated for central Slovakia (13).

Although the results indicate only a sporadic occurrence of Q fever and SF group rickettsiae in south Bohemia, without any concomitant epizootics and epidemics, they deserve the attention of public health and veterinary institutions.

ACKNOWLEDGEMENTS

The authors are grateful to Mrs E. Kachaňáková, Mrs A. Španělová, Mrs V. Ďudárková, and Mr S. Hanák for their technical assistance.

RÉSUMÉ

ÉTUDES SUR LES RICKETTSIOSES. 3. FOYERS NATURELS DE RICKETTSIOSES DANS LE SUD DE LA BOHÈME

Des foyers naturels de fièvre Q et de rickettsioses du groupe de la fièvre pourprée sont décrits et étudiés. En 1975, dans le sud de la Bohême, on a recherché la présence de rickettsies chez 1437 tiques *Ixodes ricinus*. Des rickettsies du groupe de la fièvre pourprée ont été trouvées chez 1,2% des arthropodes examinés, à l'exclusion de toute autre souche de rickettsies. Parmi 342 petits mammifères capturés en 1974-1976, on a trouvé des anticorps contre *Coxiella burnetii* chez un seul *Mus musculus*, et des anticorps contre les rickettsies du groupe de la fièvre pourprée chez 28 animaux appartenant à toutes les espèces représentées dans ce groupe de petits mammifères. Parmi les 263 lièvres capturés en 1974-1975, 14 possédaient des anticorps contre *C. burnetii* et 10 contre des rickettsies du groupe de la fièvre pourprée. On a examiné 1329 sérums de bovins, dont 17 contenaient des anticorps contre *C. burnetii* et 87 contre les rickettsies du groupe de la fièvre pourprée. Parmi 139 chevaux dont les sérums ont été éprouvés, il y avait des anticorps contre *C. burnetii* chez 2 d'entre eux, et des anticorps contre les rickettsies du groupe de la fièvre pourprée chez 4. Parmi les sérums de 52 moutons, 1 seulement contenait des anticorps contre *C. burnetii*, mais 19 en contenaient contre les rickettsies du groupe de la fièvre pourprée. En 1975 et 1976, 1086 sérums

humains ont été recueillis et on a trouvé des anticorps contre *C. burnetii* dans 19 d'entre eux, et contre les rickettsies du groupe de la fièvre pourprée dans 15.

La fréquence relativement faible des anticorps contre *C. burnetii* et contre les rickettsies du groupe de la fièvre pourprée chez les hommes et les animaux domestiques peut être rapportée à la faible infestation des tiques par les rickettsies. Un fait de grand intérêt est la fréquence relativement élevée des anticorps anti-*C. burnetii* chez les lièvres. Il apparaît que le problème de la circulation de *C. burnetii* dans le sud de la Bohême est assez compliqué. Il ne peut s'agir d'une transmission réciproque de *C. burnetii* entre les bovins et les lièvres sur les pâturages, car il n'y a pour ainsi dire pas de bovins paissant dans cette région. Les résultats négatifs donnés par la recherche de *C. burnetii* chez les tiques montrent que d'autres ectoparasites interviennent probablement dans la transmission de cet agent.

D'après les résultats ci-dessus, la présence de *C. burnetii* et des rickettsies du groupe de la fièvre pourprée dans le sud de la Bohême est démontrée; les rickettsies de la fièvre Q et du groupe de la fièvre pourprée n'apparaissent que sporadiquement, sans qu'il y ait aucune épizootie ou épidémie concomitante.

REFERENCES

1. BREZINA, R. ET AL. *Acta virologica*, 13: 142 (1969).
2. CHMELA, J. *Vertebratologické zprávy*, 2: 25 (1969).
3. GIMÉNEZ, D. *Stain technology*, 39: 135 (1964).
4. KAIZL, L. *Pracovní lékařství*, 8: 300 (1956).
5. KUBÁSEK, M. *Časopis lékařů českých*, 93: 474 (1954).
6. LIEBISCH, A. *Deutsche Tierärztliche Wochenschrift*, 83: 274 (1976).
7. PATOČKA, F. & KUBELKA, B. *Československá hygiena, epidemiologie a mikrobiologie*, 2: 340 (1953).
8. PRIX, R. ET AL. *Praktický lékař*, 40: 1085 (1960).
9. RAŠKA, K. ET AL. *Československá hygiena, epidemiologie a mikrobiologie*, 3: 1153 (1954).
10. RAŠKA, K. ET AL. *Československá hygiena, epidemiologie a mikrobiologie*, 4: 26 (1955).

11. ŘEHÁČEK, J. ET AL. *Acta virologica*, **15**: 237 (1971).
 12. ŘEHÁČEK, J. ET AL. *Folia parasitologica*, **19**: 41 (1972).
 13. ŘEHÁČEK, J. ET AL. *Journal of hygiene, epidemiology, microbiology and immunology (Praha)*, **19**: 329 (1975).
 14. ŘEHÁČEK, J. ET AL. *Bulletin of the World Health Organization*, **53**: 31 (1976).
 15. REHN, F. *Československá hygiena, epidemiologie a mikrobiologie*, **10**: 335 (1961).
 16. REHN, F. & RADVAN, R. *Journal of hygiene, epidemiology, microbiology and immunology (Praha)*, **11**: 192 (1967).
 17. SIXL, W. ET AL. *Wissenschaftliche Arbeiten aus dem Burgenland*, **1**: 80 (1973).
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