

## TICK-BORNE FEVER (EHRlichIA PHAGOCYTOPHILA) AS A SIGNIFICANT DISEASE OF CATTLE IN SWITZERLAND

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Tick-borne fever (TBF) is an infectious rickettsial disease caused by Ehrlichia phagocytophila (Rickettsiaceae), which parasitizes leucocytes, particularly neutrophils and eosinophils, and more rarely monocytes. The causative agent is transmitted by the sheep tick, Ixodes ricinus. The disease naturally affects cattle, sheep and wild ruminants.

TBF was first recognised in 1932 in the United Kingdom and was later reported from central and northern countries of Europe including Norway, The Netherlands, Finland, Ireland, Austria (1). In these countries, so far most research efforts have mainly been conducted on sheep rather than on cattle.

In Switzerland, TBF is a disease of cattle commonly named "pasture fever" (in german called "Weidefieber" ) and has been known to farmers for many years in some large areas, but the causative agent however has only recently been described (2, 3).

Owing to increasing rates of TBF and the discovery of other infected areas, detailed investigations based on clinical, epidemiological and serological aspects were initiated.

### MATERIALS AND METHODS

At present, TBF in cattle is diagnosed by two methods: a direct diagnosis by the detection of the typical forms of

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Ehrlichia in blood cells and an indirect diagnosis by the detection of specific antibodies.

In sick animals, E. phagocytophila is made visible in leucocytes by using a Giemsa staining technique. Samples of blood, from cattle presenting characteristic clinical symptoms of TBF, are collected and centrifuged. Then, the plasma is removed and Giemsa-stained buffy-coat smears are made.

During the acute phase of the disease, the causative agents are found in considerable numbers as isolated particles of varying size or as tightly or loosely packed particles in the cytoplasm of leucocytes. These typical forms, which appearing as dark blue clusters, are called morulae (Fig. 1).

Older and subclinical cases are better revealed serologically by an Indirect Immunofluorescent Antibody Test (IFAT). Presently, the following method, an adaptation of that of Jongejan et al (4), has been used: Calves are experimentally infected with E. phagocytophila by administrating intravenously 20 ml of thawed stabilate of a Swiss bovine strain. The animals are monitored by daily rectal temperature records. When the temperature is higher than 40 °C (3 to 5 days after inoculation), Ehrlichiae usually become visible in leucocytes. Then 300 ml of infected blood are collected, neutrophils are isolated and thereafter, E. phagocytophila are cultivated in these cells for 24 hours at 37 °C (5 % CO<sub>2</sub>). E. phagocytophila from infected neutrophil cultures are used as antigen on glass slides (Fig. 2).

## RESULTS AND DISCUSSION

In order to get a more detailed idea of the contaminated sites and the frequency of bovine TBF in Switzerland, we have examined more than 400 blood samples collected throughout the country from cattle presenting clinical signs of this infection.

Consequently, a map of the geographical distribution of TBF-endemic areas could be drawn (Fig. 3). Except for three

small and isolated areas in the north-west (Northern Berne (B), Neuchâtel (D) and Soleure (E)), we noticed that nearly all the infected sites are located in two large areas: one in the south-west (Vaud-Valais (C)) and another in the center-west of Switzerland (Southern Berne (A)).

The latter two areas clearly represent the majority of the positive cases diagnosed in Switzerland (90 %).

Therefore, we have directed our efforts towards the area with the highest prevalence of TBF in cattle (Southern Berne (A) 73 % of the positive cases) and we have examined the seasonal distribution of bovine ehrlichiosis' cases.

The examination of the seasonal distribution of the bovine ehrlichiosis cases revealed a close link to the seasonal appearance of the vector, Ixodes ricinus. The first cases of "pasture fever" appear in spring (april-may), when the conditions of temperature and humidity are favourable to the emergence of ticks. Thereafter, the number of cases reaches its maximum in June and falls abruptly in July and August (Fig. 4). A second wave of TBF occurs in autumn, in September and October, but to a lesser extent.

The difference between the number of declared cases in 1988 (85 cases) and 1989 (60 cases) can be explained by the fact that the veterinarians in this area have become more and more familiar with the typical clinical symptoms of TBF and immediately treat the animals, without sending blood samples for diagnosis in the laboratory.

The number of infected cattle is related to the vectors' activity. The appearance of the disease in two successive waves, one in spring and the other in autumn, corresponds to the two peaks of the vector's activity (Fig. 5).

The symptoms in cattle occur according to their age one to two weeks after arrival on tick-infested pastures.

The main clinical signs include: A high fever (41 °C or more) and a dramatic decrease in the milk production of dairy cows of more than 50 % between two milkings. During the febrile period of 4 to 7 days, E. phagocytophila is detected in large numbers in host-cells. These two symptoms are often

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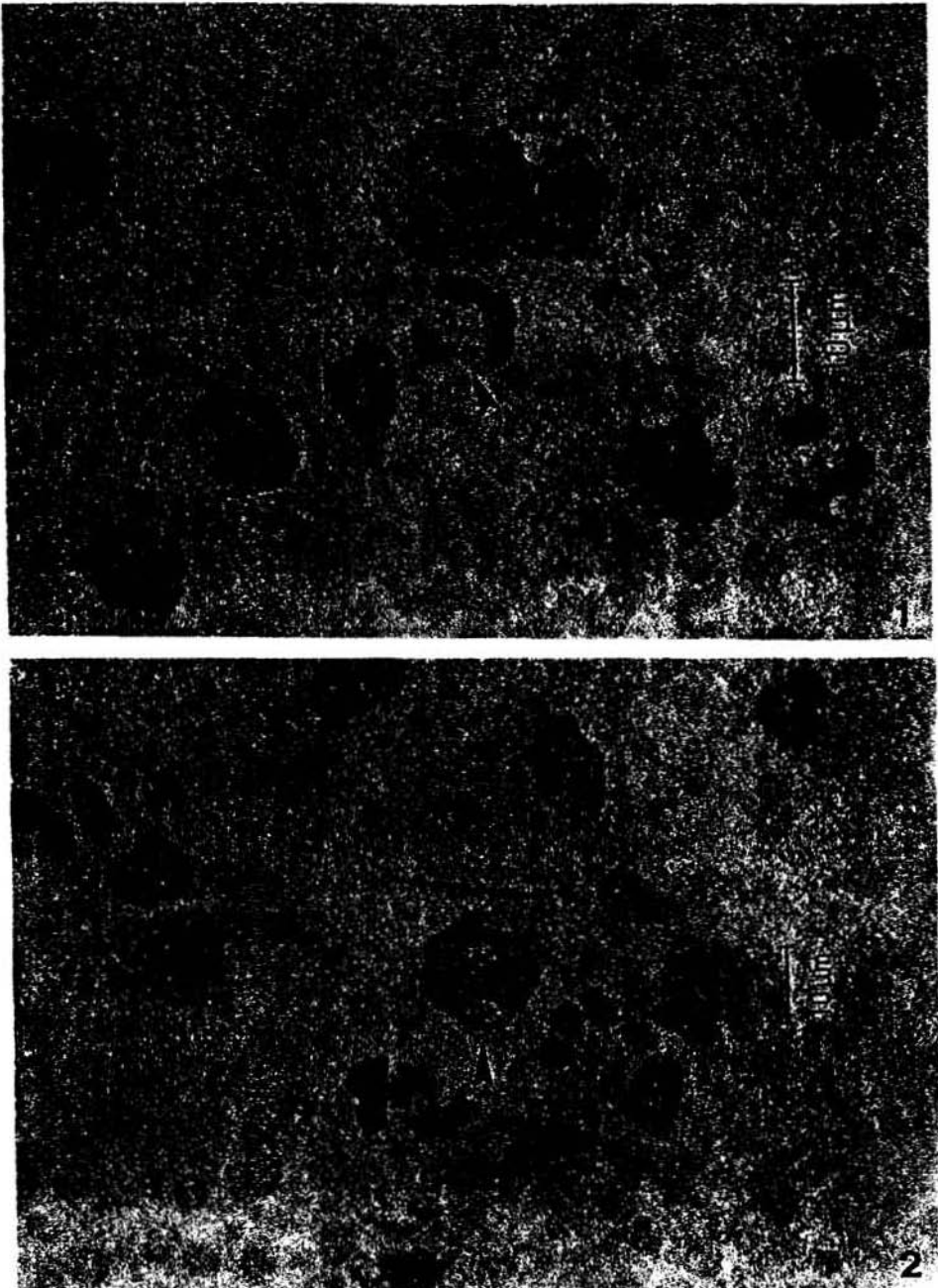
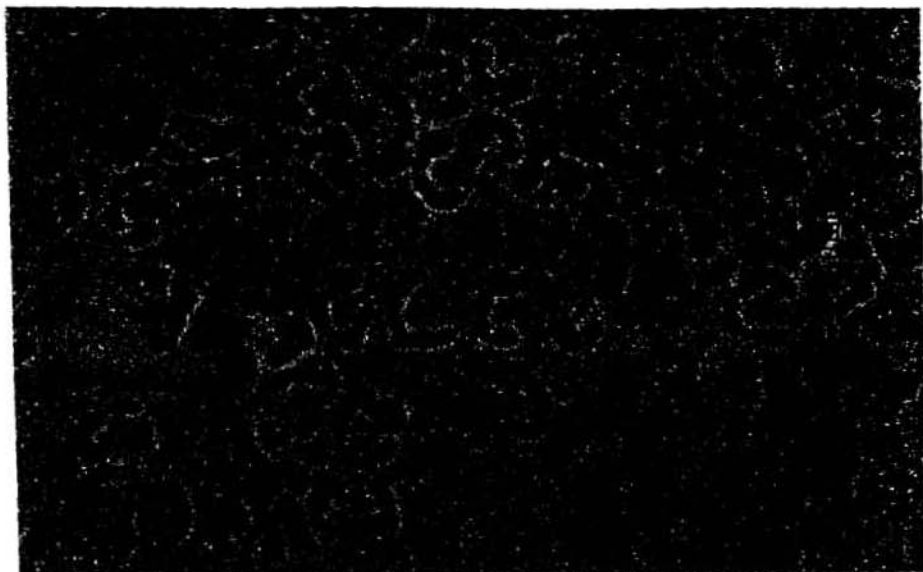


Fig. 1. Characteristic forms (morulae) of E. phagocytophila in a neutrophil (1) and in an eosinophil (2).



2. Neutrophil cultures infected with E. phagocytophila.  
Before incubation (1) and 24 hours after (2).

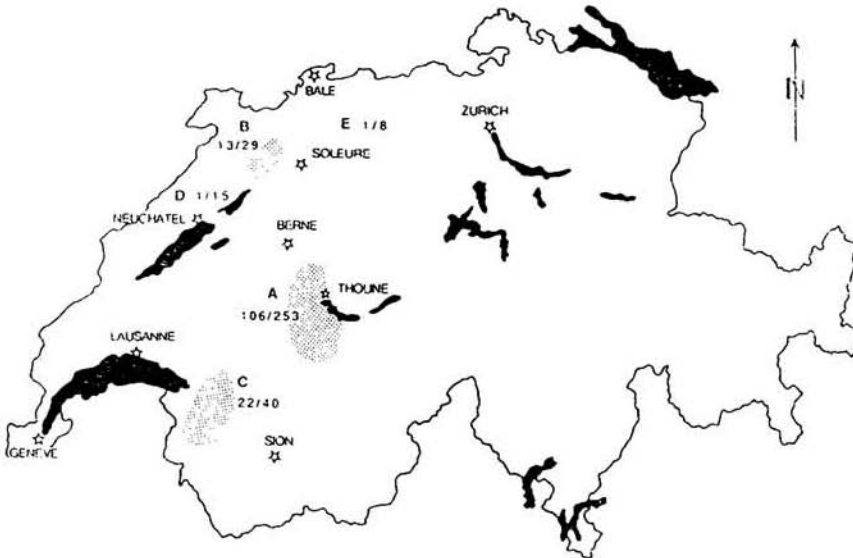


Fig. 3. TBF-infected areas in Switzerland:

A. Southern Bern B. Northern Bern C. Vaud-Valais  
 D. Neuchâtel E. Soleure.

For each area, the first number represents the diagnosed cases by direct detection and the second, the declared cases from cattle presenting clinical signs of TBF

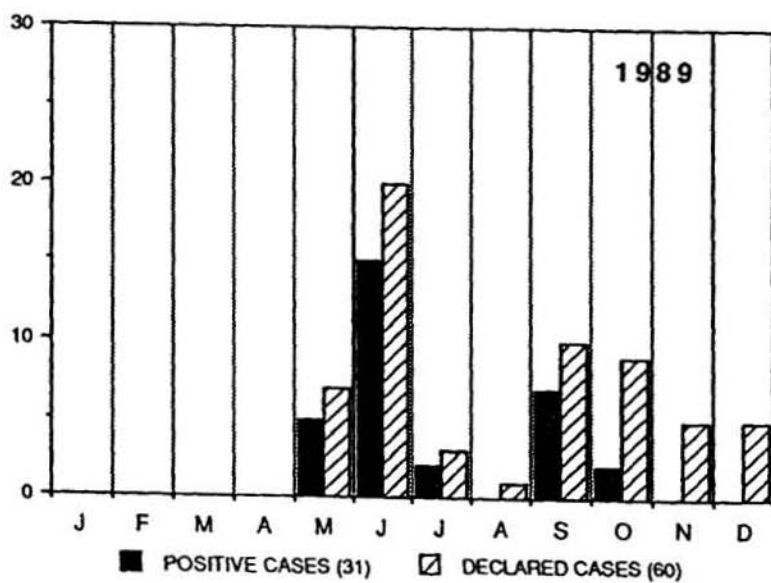
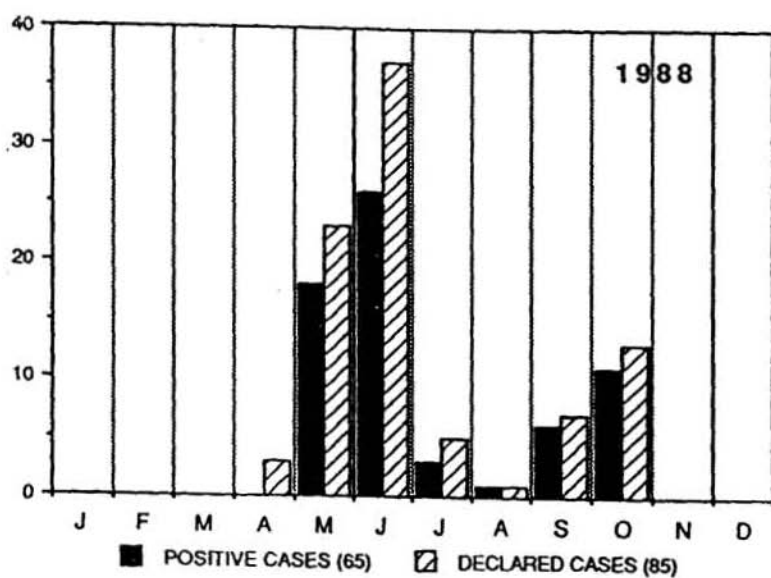


Fig. 4. TBf-cases in the Southern Bernese area.

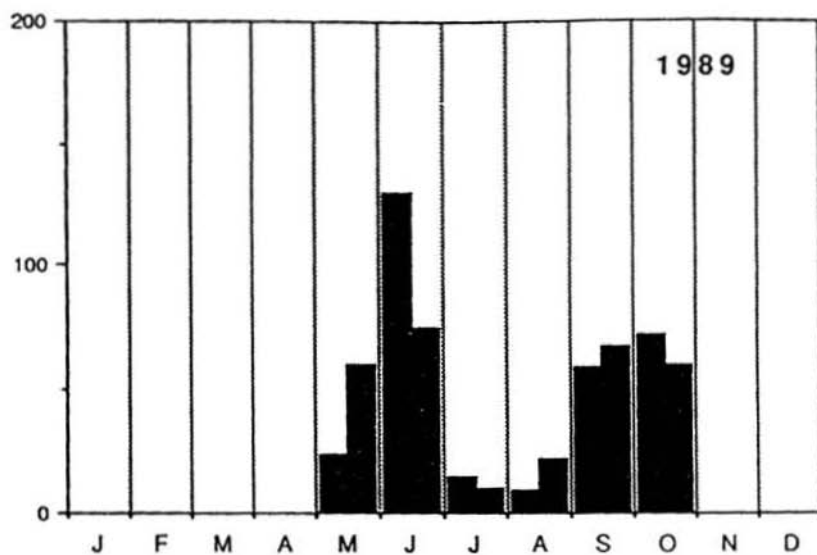


Fig. 5. *Ixodes ricinus* adults removed from cattle (from 14 cows and 14 calves) in the Southern Bernese area. (Streit, personal communication)

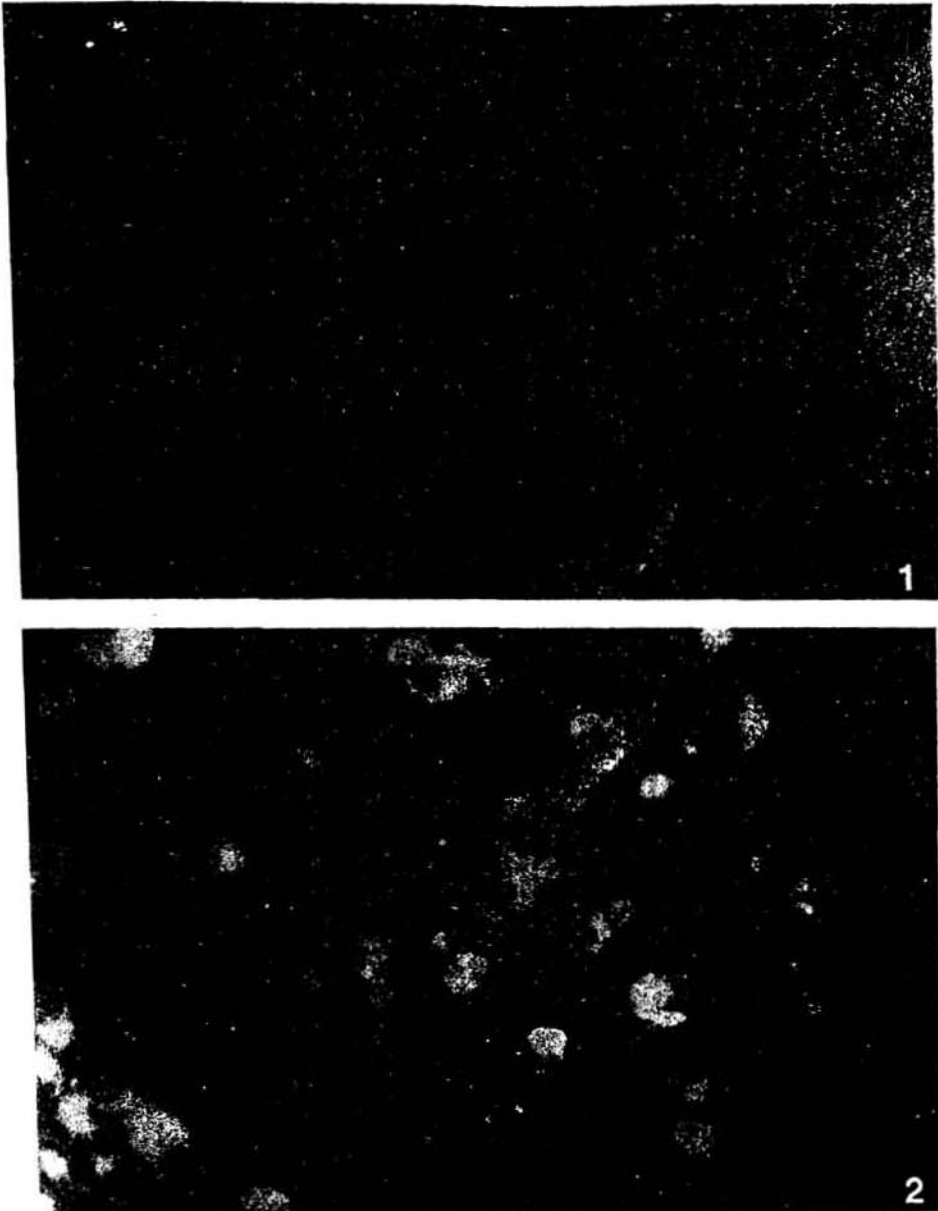


Fig. 6: Serological reaction against E. phagocytophila with IFA test (Anti-bovine IgG FITC) (500 x).

1. With negative serum (1/160 titer).
2. With positive serum (1/160 titer).