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ANTIBODIES TO BORRELIA SPIROCHETES IN SERA FROM SWEDISH CATTLE AND SHEEP

By

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HOVMARK, A., E. ÅSBRINK, O. SCHWAN, B. HEDERSTEDT and D. CHRISTENSSON: *Antibodies to Borrelia spirochetes in sera from Swedish cattle and sheep*. Acta vet. scand. 1986, 27, 479—485. — In the southern parts of Sweden a Borrelia infection transmitted by the tick *Ixodes ricinus* may affect man. In the present study antibodies to *Borrelia spirochetes* were studied in sera from 58 cows, 68 calves and 13 lambs from areas in southern Sweden where *Ixodes ricinus* occurs. For comparison, serologic studies were also performed on 88 cows and 10 lambs from the northern parts of Sweden.

Serum titers of > 80 were found in 14 of the calves and 23 of the cows from southern Sweden but in only 1 of the cows from northern Sweden. In 11 of the lambs from the south a serum titer of > 40 developed. None of the lambs from the north had a serum titer of > 40 . The results indicate that cattle and sheep in certain areas of Sweden are exposed to *Ixodes ricinus*-borne *Borrelia spirochetes*.

In 9 of the lambs from southern Sweden an endemically occurring arthritis had developed. The possibility that this arthritis may be caused by *Borrelia spirochetes* is discussed.

Ixodes ricinus; arthritis.

Recently discovered *Borrelia spirochetes*, transmitted by *Ixodes* (I.) ticks, may cause infection in man.

In the United States the name Lyme disease has been coined for *Ixodes*-borne borreliosis, which may affect different organ systems (Steere *et al.* 1977). The name *Borrelia burgdorferi* has been given to the spirochetes involved (Johnson *et al.* 1984). This infection can be transmitted by *I. dammini* and *I. pacificus* (Burgdorfer *et al.* 1982, Johnson *et al.* 1984).

In the southern parts of Sweden and in many other European countries ticks of the species *I. ricinus* occur. This tick can trans-

mit a spirochete, closely related to *Borrelia burgdorferi*, which in man may cause cutaneous manifestations (erythema chronicum migrans Afzelius, lymphadenosis benigna cutis, acrodermatitis chronica atrophicans), manifestations from the nervous system (meningitis, polyneuritis) and disorders of the joints (arthritis, subluxations) (Åsbrink 1985, Stiernstedt 1985).

In the United States mice and deer are important reservoirs of *Borrelia spirochetes* (Bosler et al 1984b). Antibodies to *Borrelia burgdorferi* have also been found in New England horses (Marcus et al. 1985). Apart from studies in humans, there have hitherto been no investigations on mammalian hosts in Scandinavia.

The island of Gotland in the Baltic, in the southern part of Sweden, is an area where borreliosis occurs endemically in humans. During the summer months Gotlandic lambs may develop an arthritis of unknown etiology. Empirically penicillin has been found to cure this arthritis if given during the early phase of the disease, and the farmers have suspected that ticks play an etiologic role. To our knowledge this type of arthritis is unknown among lambs in northern Sweden.

Bovine babesiosis in Sweden is caused by *Babesia divergens* (Christensson, to be published), a protozoan also transmitted by *I. ricinus*. Especially in the south east parts of Sweden clinical cases are common during the grazing season (Wahlgren et al. 1984). The geographical distribution of *Babesia divergens* in Sweden (Olson 1961, Wahlgren et al. 1984) is reasonably the same as that of *I. ricinus*.

The purpose of the present study was to determine whether cattle or sheep bred in areas of Sweden where *I. ricinus* occurs may develop a serologic response to *Borrelia spirochetes*.

MATERIALS AND METHODS

Cattle

Sera were collected from 3 different groups of cattle.

Group 1 consisted of sera from 88 cows from northern Sweden, which is beyond the limit of the natural distribution of *I. ricinus*. All animals had grazed during the summers, none had titers of antibodies to *Babesia*.

Group 2 consisted of 58 sera from cows bred in southern Sweden. As an indicator of contact with *Ixodes* ticks, only sera

which in a previous analysis had shown elevated titers of antibodies to *Babesia* were included.

Group 3 consisted of sera from 68 calves from stocks in southern Sweden, stocks in which babesiosis was known to occur. Blood samples were collected just before and after the grazing season, which lasted for about 4.5 months. Before grazing all calves were seronegative for *Babesia* and after grazing there were 15 seropositive animals.

Lambs

Sera were collected from 13 lambs living on the island of Gotland in southern Sweden. Nine of these lambs suffered from arthritis, which was being treated with penicillin at the time of the first serum sampling (test 1). From these lambs a second serum sample was drawn 3 to 8 weeks later (test 2). During the same period of the autumn, when the second samples were drawn, sera were also collected from 10 lambs from Jämtland in the northern part of Sweden.

Serologic tests

The sera were stored at -20 or -70°C until used in tests for assay of IgG antibodies to *Borrelia* spirochetes. A modification of the indirect immunofluorescence (IF) test described previously (Åsbrink *et al.* 1984) was employed and spirochetes isolated from a Swedish patient with tickborne borreliosis were used as antigen. The tests were performed with rabbit anti-bovine IgG, respectively rabbit anti-sheep IgG labeled with fluorescein isothiocyanate (Miles Laboratories). The serologic results are presented as reciprocal titers.

Sera from the cattle were also analyzed for antibodies to *Babesia divergens* with a modified IF-test (Christensen, to be published), where titers of 40 or more were considered as positive.

Statistics

The Wilcoxon rank-sum test was used in the statistical calculations.

RESULTS

Cattle

The results of the determinations of serum IgG antibodies to *Borrelia* spirochetes are presented in Tables 1 and 2. The sera

Table 1. Results of determinations of serum IgG antibodies to *Borrelia* in adult cows.

	No.	Titer						
		< 20	20	40	80	160	320	≥ 640
Cows from northern Sweden (area free from <i>I. ricinus</i>)	88	36	23	21	7	0	1	0
Cows from southern Sweden (area infested with <i>I. ricinus</i>)	58	1	2	9	23	19	1	3

from the group of *Babesia*-positive cows from southern Sweden showed a significantly higher median titer of antibodies to *Borrelia* spirochetes than sera from cows from northern Sweden ($P < 0.001$). A serum titer of > 80 was found in 23 of the 58 *Babesia*-positive cows from the south, but in only 1 of the 88 cows from the north (Table 1).

At the end of the summer 14 of the 68 calves had developed titers of antibodies against *Borrelia* spirochetes higher than 80 (Table 2). There was no significant difference in serum titers of antibodies to *Borrelia* spirochetes between the 15 calves, which after grazing were positive for *Babesia*, and those which remained *Babesia*-negative.

Table 2. Results of determinations of serum IgG antibodies to *Borrelia* in calves before and after grazing on pasture where *I. ricinus* occur.

	No.	Titer						
		< 20	20	40	80	160	320	≥ 640
Calves before grazing	68	16	18	23	10	1	0	0
Calves after grazing	68	7	11	17	19	11	3	0

Lambs

The results of the determinations of serum IgG antibodies to *Borrelia* spirochetes in lambs are shown in Table 3. In paired sera from the Gotlandic lambs the titers increased significantly by two titer steps or more in 7 of the 13 cases and increasing titers were found both among lambs with and without arthritis. In 11 of the 13 lambs from Gotland a serum titer of 80 or higher developed, but all lambs from Jämtland in northern Sweden displayed serum titers less than 80.

Table 3. Results of determinations of serum IgG antibodies to *Borrelia* in lambs.

	No.	Titer						
		< 10	10	20	40	80	160	320
Lambs from Gotland, test 1	13	0	0	4	4	5	0	0
, test 2	13	0	0	0	2	4	5	2
Lambs from Jämtland	10	2	6	1	1	0	0	0

DISCUSSION

The results of this study indicate that cattle and sheep in certain areas of Sweden are exposed to *I. ricinus*-borne *Borrelia* spirochetes. *Leptospira* organisms may cause cross-reacting antibodies. However, this spirochetal infection has only very rarely been demonstrated in Swedish ruminants (*Hurvell*, pers. comm.). All calves investigated came from stocks in which babesiosis was known to occur and it is likely that all calves in these areas run a risk of being parasitized by ticks. Among the calves increasing serum titers of antibodies to *Borrelia* spirochetes developed irrespective of whether or not serologic positivity to *Babesia* developed. Thus, the results do not suggest that ticks harbouring the *Babesia* protozoa bear *Borrelia* spirochetes more often than other *I. ricinus* ticks.

The question whether a *Borrelia* spirochete infection in ruminants may result in clinical symptoms, or just gives a sub-clinical infection, is still not answered. In man *Ixodes*-borne borreliosis may cause arthritis (*Burgdorfer et al.* 1982, *Åsbrink* 1985, *Stiernstedt* 1985). Clinical findings indicate that arthritis may also develop in dogs infected with tick-borne *Borrelia* spirochetes (*Bosler et al.* 1984a). In this investigation elevated titers of antibodies to *Borrelia* spirochetes were found both in Gotlandic lambs with and in those without arthritis. These results may be explained by the occurrence of both clinical and sub-clinical infections. However, the hypothesis that *Ixodes*-borne spirochetes may cause endemically occurring arthritis in lambs needs further investigation, including attempts to isolate spirochetes from affected animals. It is also important that the agent responsible for the seroresponse of the ruminants is identified, as ruminants may serve as reservoirs for the spirochetes causing borreliosis in humans.

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SAMMANFATTNING

Serum antikroppar mot Borreliaspiroketer hos nötkreatur och får i Sverige.

Det har nyligen visats att *Borrelia* spiroketer som överförs av *Ixodes ricinus* fästingar orsakar infektion hos människa. Denna borrelios förekommer endemiskt i södra och mellersta delarna av Sverige.

58 kor, 68 kalvar och 13 lamm från södra Sverige och 88 kor och 10 lamm från norra Sverige har undersökts med avseende på förekomsten av serum antikroppar mot *Borrelia* spiroketer. Serum titrar > 80 påvisades hos 14 av kalvarna och 23 av korna från södra Sverige men endast hos en av korna från norra Sverige. 11 av lammen från södra Sverige erhöll efter betessäsongen en serum titer > 40 . Inget av de nordsvenska lammen hade serum titer > 40 . Resultaten tyder på att nötkreatur och får i vissa områden av Sverige är exponerade för *Ixodes ricinus* överförda spiroketer.

Nio av de undersökta lammen från södra Sverige hade drabbats av en endemiskt förekommande artrit. Hypotesen att denna artrit orsakas av *Borrelia* spiroketer diskuteras.

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