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Brief Communication

LONGSTANDING COLONISATION OF THE RESPIRATORY TRACT OF CALVES BY MYCOPLASMA DISPAR

Mycoplasma dispar (M. dispar) was first described in pneumonic calf lungs in England by *Gourlay & Leach* (1970). It has since been found to be one of the most commonly isolated mycoplasmas from pneumonic lungs of calves in England, Australia, USA, Japan and Denmark. The present communication reports the finding of M. dispar in Finnish calves and presents some results from studies on the course of colonisation of the respiratory tract by M. dispar followed by means of nasal swab samples.

The animals studied comprised a group of 6 conventionally reared Ayrshire calves purchased by the research establishment. Five of the calves originated from one local dairy farm and the sixth from another farm. The calves were transported together and were housed for 9 months in the same room, in close contact with each other but completely isolated from other calves in the research establishment. On arrival, 5 of the calves were 3—5 weeks old and the sixth was 3 months old. Three of the calves were splenectomized. The first nasal swab samples were taken 3 weeks after arrival and then 1—3 times monthly, excluding months 4, 6 and 8, when no samples were taken. The animals were slaughtered 12—17 months after the start of the experiment.

Nasal samples were taken with swabs and placed immediately in mycoplasma broth (1.8 ml). The broths with swabs were incubated for 45 min at 37°C before dilution. At slaughter, the trachea and the main bronchus of the apical lobus, and in the splenectomized calves also the larynx, were sampled with swabs and apical lung tissue by preparing tissue-broth suspension with scissors before making the dilutions.

Glucose calf-serum (GS) broth described by *Gourlay & Leach* (1970), slightly modified, conventional Hayflick broth and corresponding solid media were used for culturing the mycoplasmas. Ampicillin (0.5 mg/ml) instead of penicillin was used in both

media. The inoculated broths were incubated at 37 °C for 3 weeks. Passages on the solid media were made conventionally. Broth-to-broth passages were sometimes used. The incubation of the intial broths was continued after subculturing on the solid media and subcultured again if the change of pH was more advanced. Solid media were incubated in air with increased CO_2.

At the first examination 3 weeks after arrival, all the calves were found to harbour in their respiratory tracts mycoplasmas which could be isolated by means of GS broth only. The growth characteristics and colony morphology of the isolated strains resembled those of M. dispar described by *Gourlay & Leach* (1970). The isolates were sensitive to digitonin. Three of the strains were cloned and after some broth-to-broth passages studied by serological methods. Positive results were recorded with antisera against M. dispar (462/2) in the epi-IF-test (*Rosendal & Black* 1972) and GI-test (*Glyde* 1964).

All six calves were regularly positive for nasal isolation of M. dispar for a period of 9 months after the first examination. During the 10th, 11th and 12th month, 4, 3 and 2 calves, respectively, were still detectably, although irregularly, shedding M. dispar in their nasal secretions. The last successful nasal isolation of M. dispar was in the 13th month from 1 of the calves. There appeared to be no difference in the duration of shedding of M. dispar between splenectomized and non-splenectomized calves.

The intensity of colonisation was highest in the first months of the study when the amounts of M. dispar per nasal sample were in most cases $\geq 10^6$ ccu, when diluted that far. The shedding of M. dispar then appeared to decrease gradually. After 9 months it was often difficult to isolate M. dispar directly from the initial broth sample without broth to broth passages. The titres of M. dispar were then always below 10^3 ccu. The presence of other mycoplasmas eg. M. bovirhinis or Acholeplasma laidlawii interfered with the isolation of M. dispar in the last months when the amounts of M. dispar in nasal samples were small.

Three non-splenectomized calves were slaughtered at months 12, 13 and 15, with the last nasal isolation of M. dispar 1, 0 and 5 months, respectively, before their death. M. dispar could not be isolated from the respiratory tracts of these calves at slaughter; however, the larynx and the proximal part of the trachea were not investigated.

The three splenectomized calves were slaughtered at months 13, 15 and 17, with last nasal isolations of M. dispar 1, 6 and 6 months earlier, respectively. M. dispar was isolated in small amounts (10^1—10^4 ccu/sample) from the larynx and the trachea of all of these calves, but not from distal parts of the respiratory tract.

The results establish the occurrence of M. dispar also in Finnish cattle. The main result of the studies, however, is the demonstration of the relatively long-standing overt colonisation by M. dispar of the respiratory tract in the calves. Continuous shedding over a period of several months is apparently an effective way of maintaining this mycoplasma in a continuously recruited and large enough population of calves. As calves grow the degree of colonisation by M. dispar tends to decrease and elimination of the mycoplasma may take place. This view is supported by the results of *Thomas & Smith* (1972). However, the possibility and the role of a more latent type of colonisation, with perhaps intermittent shedding, in older calves and adults remain to be studied.

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