

## Bovine Abortions Associated with *Neospora* in Denmark

During recent years protozoa of the *Neospora* genus have been identified as a major cause of bovine abortions world wide (Barr *et al.* 1991, McIntosh & Haines 1994, Thornton *et al.* 1991, Yaeger *et al.* 1994). A presumptive diagnosis of *Neospora* abortion can be made on the foetal histopathology as infected foetuses have a distinctive pattern of multifocal necrotizing and non-suppurative encephalitis often with an accompanying nonsuppurative myocarditis and a varying degree of focal inflammation in other organs (Barr *et al.* 1990, Thornton *et al.* 1991). However, confirming the diagnosis of *Neospora* abortion requires detection of protozoa in foetal tissues that react positively by immunohistochemistry with *Neospora* specific antisera (Barr *et al.* 1990, 1991).

In routine diagnostic examinations of aborted bovine foetuses in Denmark, lesions presumptive of *Neospora* infections are commonly found, but a specific diagnosis has not been established. Therefore, a minor investigation which included immunohistochemical examination of paraffin embedded tissues was performed on 2 suspected cases of *Neospora* abortion.

This article presents the findings from the cases and reports for the first time bovine abortions caused by the bovine *Neospora* parasite in Denmark.

Bovine foetuses with the corresponding placenta and maternal blood were submitted for

routine diagnostic evaluation to the Danish Veterinary Laboratory from 2 dairy herds where multiple abortions had occurred. Microbiologic examinations of foetal tissues and fluids as well as serology on foetal pleural effusion and maternal blood were performed as previously described (Agerholm *et al.*, accepted) and were negative for viral or bacterial pathogens. Tissues collected for histology included foetal brain, lung, myocardium, liver, kidney, and placenta. The tissues were fixed in 10% buffered neutral formalin, paraffin embedded, sliced, and stained with haematoxylin and eosin. Based on the microscopic lesions found, a presumptive diagnosis of protozoal infection was made in each case. Tissues were sent to the California Veterinary Diagnostic Laboratory System, University of California for *Neospora* and *Toxoplasma gondii* immunohistochemistry. *Neospora* and *T. gondii* antisera were both developed in rabbits using respectively, a bovine *Neospora* isolate (BPA-1, optimal dilution 1:200) (Conrad *et al.* 1993) and combined ME-49 plus RH strains of *T. gondii* (optimal dilution 1:250).

*Case 1.* The aborted foetus originated from a Holstein-Friesian herd in which 6 abortions had occurred within a few weeks. The foetus was approximately 7 months old. Gross lesions were not present, but microscopically there was a mild multifocal encephalitis char-

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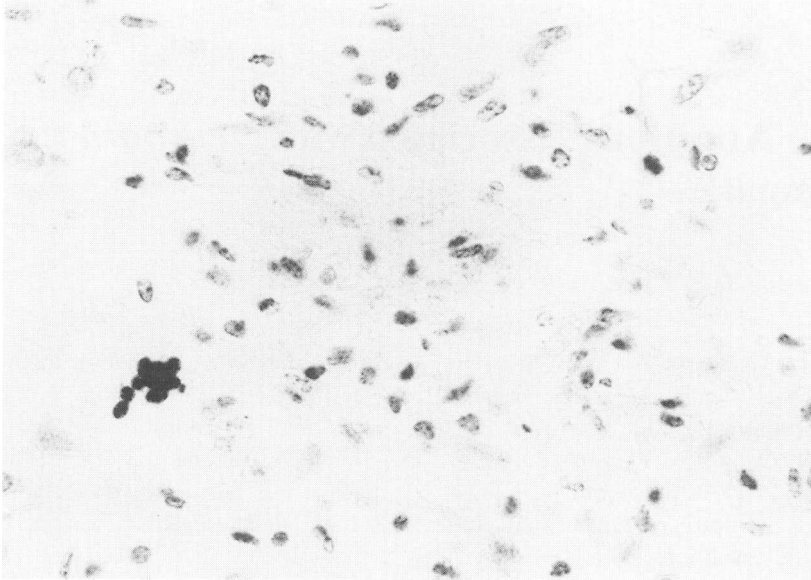


Figure 1. Photomicrograph of foetal brain from case No. 1. A cluster of tachyzoites staining positively with *Neospora* antiserum lies adjacent to a focus of gliosis. Immunoperoxidase, haematoxylin counterstain. 290 x.

terized by small random foci of gliosis. In addition, a focal necrotizing placentitis was found. By immunohistochemistry rare tachyzoites were found in association with foci of gliosis (Fig. 1) which reacted positively with *Neospora* antisera but failed to react with *T. gondii* antisera.

**Case 2.** The foetus was 5 months old and originated from a Holstein-Friesian herd in which 40 abortions had occurred during the last 2 years. Gross lesions were not present, but multifocal necrotizing encephalitis with gliosis, diffuse nonsuppurative myocarditis, and multifocal necrotizing hepatitis were found histologically. Immunohistochemical examination revealed the presence of protozoal tissue cysts and tachyzoites in the brain (Fig. 2) which reacted with the antisera as in case 1. The histopathological changes found in cases 1 and 2 are in accordance with other reports

(Barr *et al.* 1990, Thornton *et al.* 1991), and as *Neospora* tachyzoites or cysts were demonstrated by immunohistochemistry, there is strong evidence that these abortions were caused by a *Neospora* species. These abortions represent the first reported cases of *Neospora* infection in bovine foetuses in Denmark. It was not possible to determine whether all the abortions reported in the actual herds were due to *Neospora*. However, *Neospora* is well confirmed to be as a cause of multiple abortions (Anderson *et al.* 1991, Yaeger *et al.* 1994).

Even though the examined materials were very restricted, the findings suggest that *Neospora* infections may be a common cause of bovine abortions in Denmark. However, conclusions on the incidence and prevalence of the infection in Danish cattle must await further studies.

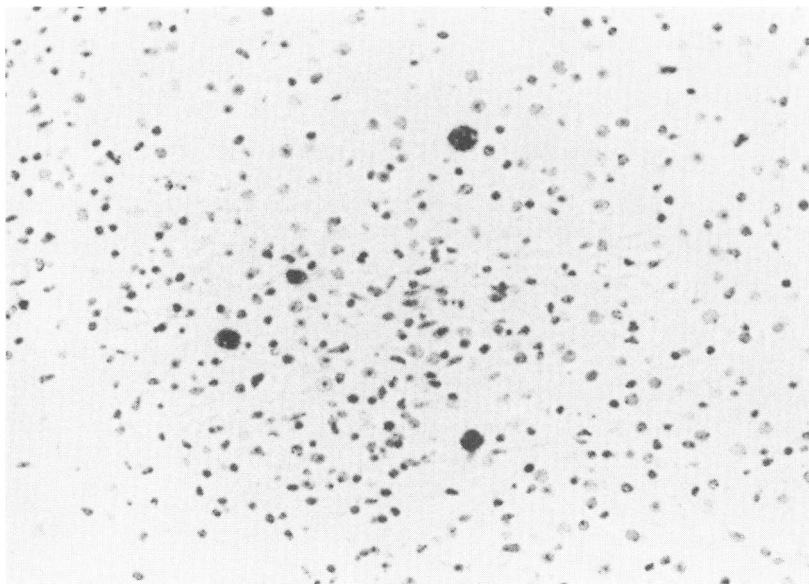


Figure 2. Photomicrograph of foetal brain from case No. 2. Several protozoal tissue cysts staining positively with *Neospora* antiserum lie around a focus of gliosis. Immunoperoxidase, haematoxylin counterstain. 140 x.

### Acknowledgements

We wish to thank John Barbano for technical expertise in performing the immunohistochemistry.

*J.S. Agerholm*

Danish Veterinary Laboratory  
Department of Pathology and Epidemiology  
Copenhagen, Denmark.

*B.C. Barr*

California Veterinary Diagnostic Laboratory System  
University of California  
School of Veterinary Medicine  
Davis, California, USA.

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*(Received November 18, 1994; accepted November 28, 1994).*

Reprints may be requested from: J. S. Agerholm, Danish Veterinary Laboratory, Department of Pathology and Epidemiology, Bülowsvej 27, DK-1790 Copenhagen V, Denmark.