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AGE-SPECIFIC SEROPREVALENCE OF *NEOSPORA* SPECIES INFECTIONS IN CATTLE ON SELECTED BRITISH DAIRY FARMS

Davison H.C.[1], Dannatt L.C.[2], Guy F.[1], Roberts J.[1], McGarry J.[1], Otter A.[3], Williams D.J.L.[1], Trees A.J.[1]

L'infection à Neospora est une cause récemment reconnue d'avortement chez les bovins dans le monde entier. Son épidémiologie est peu connue, excepté que l'agent causal, Neospora sp., peut être transmis par voie congénitale. La transmission horizontale de ce parasite n'a pas été prouvée, mais des éléments circonstanciels suggèrent qu'elle est possible. En Angleterre, 12 élevages de bovins Holstein/Friesian qui ont eu des taux d'avortements annuels supérieurs à 10% attribués à Neospora, et dans lesquels des espèces de Neospora ont été mises en évidence par la technique d'immunocytochimie à partir du cerveau du foetus ou par sérologie maternelle, ont été sélectionnés pour des études sérologiques. Tous les bovins ont fait l'objet d'un prélèvement de sang une seule fois et les anticorps spécifiques contre Neospora ont été détectés par IFAT (en utilisant la limite de séropositivité de 1:640) ou au moyen d'un ELISA commercial récemment développé, ou par la combinaison des deux tests. Des sérums ont aussi été obtenus à partir de vaches adultes qui ont été prélevées régulièrement pendant une année dans une autre exploitation. Les prévalences de troupeau obtenues, avec intervalle de confiance à 95%, allaient de 7% (5, 10) à 56% (50, 61). La prévalence animale la plus élevée était chez les veaux de moins de 7 mois d'âge et chez les vaches adultes. La valeur faible de la prévalence apparente chez certains groupes de génisses pourrait refléter l'inclusion d'un grand nombre de résultats faussement négatifs dans ces groupes à cause d'une diminution de la réponse immunitaire, comme il est observé avec les génisses supposées infectées par voie congénitale et surveillées sérologiquement par prise d'échantillons répétée. Les profils sérologiques obtenus seront discutés dans le contexte d'un rôle possible de la transmission verticale et horizontale.

INTRODUCTION

The protozoal parasite *Neospora* species has recently been recognised as an important cause of bovine abortion in many countries including the United Kingdom (Dubey and Lindsay, 1996). The parasite's full life-cycle is unknown. Congenital transmission is the only known natural mode of transmission in cattle, but the occurrence and source of horizontal transmission have yet to be determined. Currently, *Neospora* species infections in cattle on selected British dairy farms are being investigated by cross-sectional and longitudinal serological studies and the preliminary results are presented.

MATERIALS AND METHODS

Sample collection

Twelve Holstein/Friesian dairy farms situated in eight counties of England were selected using the following criteria: 1) high level of interest and co-operation of the farmer and veterinary surgeon; 2) history of *Neospora*-associated abortions (diagnosis by immunocytochemistry of foetal brain or maternal serology); 3) herd size; and 4) location. All cattle were blood sampled on the farms, which had herd sizes ranging from 143 to 793 and had had multiple *Neospora*-associated abortions over various periods, except two farms (Farm H and Farm N) that had had low reported abortion rates and only one or two *Neospora*-associated abortions. Sera were also obtained from adult cows that had been serially blood sampled every two months over a one year period on an additional *Neospora*-affected farm (Dannatt *et al.,* 1995). Sera were stored at -20°C until the tests were performed

Serological assays

The sera were tested for *Neospora*-specific antibodies either with an indirect fluorescent antibody test (IFAT) or with an ELISA[4] (Williams *et al.*, 1997) or with both tests. A cut-off value of 1:640 was used for the IFAT. The ELISA results were expressed as percent positivity values (PP), where the PP equals the test sample OD divided by the mean high-positive control OD, expressed as a percentage. The following interpretation was used to categorise the PP values: <20 PP: negative; 20-25 PP: inconclusive; >25 PP: positive.

[1] Liverpool School of Tropical Medicine/Faculty of Veterinary Science, Pembroke Place, Liverpool, L3 5QA, United Kingdom
[2] Veterinary Laboratories Agency, Veterinary Investigation Centre, Staplake Mount, Starcross, Exeter, Devon, EX6 8PE, United Kingdom
[3] Veterinary Laboratories Agency, Veterinary Investigation Centre, Job's Well Road, Carmarthen, SA31 3EZ, United Kingdom
[4] MAST DIAGNOSTICS, Mast Group Ltd, Mast House, Derby Road, Bootle, Merseyside, L20 1EA, United Kingdom

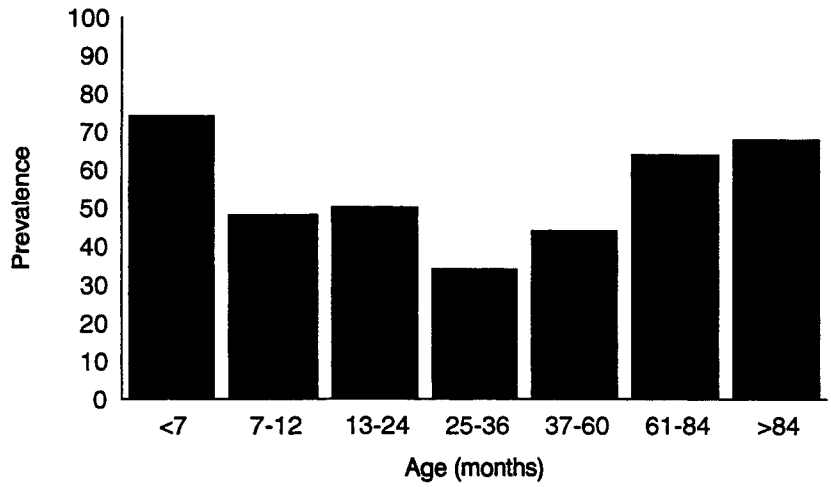
Data analysis

Prevalence values were stratified by herd and by age, and 95% confidence intervals were calculated using the software CIA[5]. Age-specific prevalence values were presented graphically for each herd and compared by Chi-squared analysis for 2 x k tables with an adjustment for continuity using the software EPIINFO version 6.02[6].

RESULTS

Herd-specific prevalence values ranged from 7% (5, 10) to 56% (50, 61). In general, the highest proportions of seropositive cattle were found with the calves less than seven months old and adult cows more than five years old, for example on Farm A, Farm C and Farm D as shown in Figure 1. On Farm H and Farm N (which had low abortion rates) 19% and 14%, respectively, of cattle more than five years old were found to be seropositive. The serially-sampled cows were found to have antibody responses that were either: 1) negative; 2) positive; or 3) fluctuated between positive/negative test status.

Figure 1
Age-specific prevalence of *Neospora* species by IFAT in cattle on three dairy farms (Farm A, Farm C and Farm D)



(n = 605)

DISCUSSION

Previously, only a proportion of cattle on selected British dairy farms have been tested for *Neospora*-specific antibodies. Age-specific prevalence profiles obtained by testing whole herds can be useful in the investigation of the role of congenital versus horizontal transmission. The results suggested that congenital transmission was the major mode of transmission in some of the herds tested, in agreement with previous work (Pare *et al.,* 1996), but did not exclude the possibility of horizontal transmission. Lower proportions of seropositive animals were found in some heifer groups, which may reflect the inclusion of higher numbers of false-negative results in these groups due to declining antibody responses, as observed in serially-monitored, putative congenitally-infected calves. Therefore, serial sampling of negative calves from birth would be more appropriate than monitoring heifers or cows to provide evidence of post-natal exposure. The observation that 14% to 19% of adult cows were seropositive on two farms that had low abortion rates indicates the importance of investigating other factors that may affect the outcome of *Neospora* infections.

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[5] Confidence Interval Analysis, British Medical Journal, BMA House, Tavistock Square, London, WC1H 9JR

[6] Central Disease Center, Atlanta, Georgia, 30333 USA and the World Health Organisation, Geneva, Switzerland