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Equine Babesiosis

Pathology and Serology

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Infection: IFA Test.

1. Introduction

A case of equine babesiosis diagnosed in the Bowral area of New South Wales (Churchill and Best, 1976) was shown by Mahoney *et al.* (1977) to be due to *Babesia equi*. In the years since that diagnosis no further field cases have been observed, but the presence of *B. equi* infection in carriers has been confirmed at foci in New South Wales, Western Australia and Queensland. The infected horses either had a history of being imported from countries known to be enzootic for equine babesiosis or had been subjected to intravenous manipulations with hypodermic needles at the same times as in-contact, infected horses. There is no evidence that *B. caballi* is present in Australia or that vector spread of equine babesiosis has occurred.

2. Clinical and Pathological Changes

The experience of pathogenic equine babesiosis in Australia is limited to the observations by Churchill and Best (1975) of the field case, and those by Mahoney *et al.* (1977) of two laboratory horses, one splenectomised, the other intact. Both intact horses were mildly affected with fever to 40°C, listlessness, inappetence and jaundice. The packed cell volume of the laboratory horse fell to less than 50% of its initial value; parasites were readily found in blood films from both intact horses. The syndrome in the splenectomised animal was much more pronounced, and led to death. This horse also experienced haemoglobinuria, incoordination and, terminally, extreme weakness. At autopsy, jaundice was marked, there were subendocardial haemorrhages, fluid in body cavities, large yellow clots in blood vessels and a swollen liver.

Microscopic examination revealed generalised proliferation of reticuloendothelial cells and the presence of parasitised erythrocytes within vessels. Numerous macrophages were present in sinusoids and central veins of the liver as well as thrombi in large vessels and periportal accumulations of mononuclear cells. The liver lesions could be confused with those of equine infectious anaemia except for the presence of parasitised erythrocytes. Severe nephrosis was evident in the kidney with haemoglobin and protein casts and interstitial infiltration by mononuclear cells.

3. Diagnosis

3.1. Acute Cases

Symptoms and lesions of a febrile, haemolytic condition would suggest *B. equi* infection, and this diagnosis would be confirmed or discounted after examination of a blood film. *B. equi* is readily detected in peripheral blood, and should be present in thin as well as thick blood films

during primary attacks (see 8.3. of Callow *et al.*, 1993). Impression smears from organs as well as peripheral blood films might be taken at autopsy.

3.2. Carriers

3.2.1. Blood Films

B. equi infections persist in some horses for many years, and carriers experience recrudescences of parasitaemia. It is, therefore, worthwhile to collect thick and thin blood films from suspected groups, and perform this on several occasions when no serological testing can be done.

3.2.1. Serology

In Australia, a reliable indirect fluorescent antibody (IFA) test (see 5.1.) has been developed (Callow *et al.*, 1979). There is complete agreement in the results obtained from this test and from the standard complement fixation test used by the United States Department of Agriculture (Frerichs *et al.*, 1969). The IFA test has detected very longstanding infections—of at least four years duration in one group of horses and possibly much longer infections in another. There appear to be very satisfactory levels of specificity and sensitivity in the test.

4. References

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5. Appendixes

5.1. Appendix 1 — The Indirect Fluorescent Antibody Test

This test is essentially the same as that described for bovine babesiosis (see 8.5. of Callow *et al.*, 1993). Minor differences in the preparation of antigen and the performance of the test are given.

5.2. Appendix 2 — Antigen Slides

High parasitaemias can be attained with *B. equi*, and antigen should be made from blood as heavily infected as possible. Parasitaemia should not be less than 10% of erythrocytes infected; the best antigen has been prepared from a parasitaemia of over 30% (L.L. Callow, unpublished data, 1976).

On this occasion, the infection had not developed rapidly, but no difficulty with non-specificity was encountered. Some horse bloods contain a component causing autofluorescence, and preliminary screening might be useful to exclude these animals as donors of antigen.

5.3. Appendix 3 — Antihorse Ig G Conjugate

A commercial product useable in highly diluted form is available from ICN Biomedicals, PO Box 187, Seven Hills, NSW 2147. Tel. (02) 838 7422; Fax (02) 838 7390. Notwithstanding, commercial batches of conjugate may be variable and this factor should be borne in mind if problems are being experienced with the IFA test. All batches of antihorse Ig G conjugate should be carefully checked against a standard preparation before use in the test system.

5.4. Appendix 4 — Performance and Interpretation of the Test

Titres in IFA tests will depend on the optical system used to read, but, in general, much higher titres are observed in horses infected with *B. equi* than in cattle infected with *B. bovis*; on average, titres are 10 times higher in horses. Furthermore, the reactivity of sera from uninfected horses is minimal, so that there is little difficulty in interpreting reactions at the upper limit of the negative dilution range and the lower limit of the positive range. Fluorescence microscopy performed with a vertical illuminator and a HBA 200 W mercury lamp, used with x40 neofluor objective, x12.5 oculars, excitor filter No.1 (BG 12) and barrier filters 53 and 54 give positive titres from 1/270 to 1/7290, and negative titres (non-specific reactions) from 1/10 to 1/90 in the *B. equi* test.