Biochemical and molecular identification of species of Taenia

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The adults of 5 species of taenid cestodes are found in Australian canids. Three of these, Echinococcus granulosus, Taenia hydatigena and T. ovis, are of considerable public health and economic importance. E. granulosus may be readily identified by its small size. However, T. hydatigena, T. ovis and the other 2 species within the genus Taenia, T. pisiformis and T. serialis, cannot be so easily distinguished.

Beveridge and Gregory (1976) found that gross strobilar morphology and anatomy of the mature proglottid were reliable methods of differentiating the 4 species of Taenia in suitably relaxed, fixed and stained specimens. However, such procedures are not always possible and most surveys have used rostellar hook morphology to identify species (Jackson and Arundel 1971; Arundel 1972; Coman 1973). Despite its use by Jackson and Arundel (1971), hook shape does not appear to be reliable for specific identification (Beveridge and Gregory 1976; Edwards and Herbert 1981). Hook number and size are of more value, but there is considerable overlap in both characters between species and unequivocal identification is often impossible (Beveridge and Gregory 1976; Edwards and Herbert 1981).

Biochemical and molecular techniques may provide more reliable identification of taenid species. These techniques produce taxonomic characters which are less likely to be influenced by environmental factors than morphology (Thompson 1988). Although they are relatively expensive and time-consuming to set up, once established they provide reproducible data readily and cheaply. Two techniques established in our laboratory may provide markers for species of Taenia from canids and other definitive hosts.

In enzyme electrophoresis, proteins extracted from fresh or frozen parasite tissue are separated on horizontal starch gels and stained for specific enzymes as described by Lymbery and Thompson (1988). Enzymes of different mobility (isozymes) can often be interpreted as the products of different genetic loci or of alternative alleles at the same locus. Eight enzyme systems have provided isozyme markers which appear to distinguish between taenid species; adenosine deaminase, glucose phosphate isomerase, glutamate dehydrogenase, hexokinase, mannose-6-phosphate isomerase, nucleoside phosphorylase, peptidase and phosphoglucosaminate.

In restriction endonuclease analysis, DNA is isolated from fresh or frozen parasite tissue, cleaved with restriction enzymes, separated on agarose gels and hybridised to specific gene probes, as described by Yap and Thompson (1987) and Yap et al (1988). Differences in the mobility of restriction fragments may be interpreted as differences in restriction sites, resulting from insertions, deletions or base substitutions in homologous DNA sequences. Probes available in our laboratory include the ribosomal DNA gene unit of Echinococcus granulosus and the mitochondrial genome of Taenia hydatigena. By using different combinations of restriction enzymes and probes, we have found unique restriction fragment patterns for all the taenid species we have examined.

The potential of these techniques to identify taenids especially morphologically similar species from culains, is obvious. A number of overseas studies have already used total protein (Bursey et al 1980) and enzyme electrophoresis (Le Riche and Hewett 1977, 1978; Allopp et al 1987) for this purpose. Rishi and McManus (1987) constructed DNA probes which distinguished Taenia solium from T. saginita in humans.

We are not yet in a position to reliably distinguish between species of Taenia from Australian canids. Unequivocal identification requires diagnostic characters, that is, those that uniquely specify a given taxon. Before the isozymes and restriction fragment patterns we have found can be regarded as diagnostic characters, we need to determine the extent of variation within each species. This requires adequate sampling over the geographic range of each species; so far the maximum number of isolates we have examined for any species is five, and for some species only one isolate has been examined. Our most pressing requirement is, therefore, to analyse as many isolates as possible from the four species of Taenia found in Australian canids. To this end, we would like to hear from colleagues throughout Australia who may be able to provide us with samples of either adult or larval tapeworms. Such collaborative studies are essential if the full value of this approach is to be realised.

References

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Congenital lymphoedema in an Ayrshire-Friesian crossbred female calf

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Congenital oedema is due to genetic malformation of the lymphatic system involving an apparent failure of the central and/or peripheral lymphatic vessels to make adequate connections, resulting in hypoplasia of lymphatic endothelia and obstruction to lymphatic drainage (Donald et al 1952, Patterson et al 1967; Lugnibuld et al, 1967). It is also called hereditary lymphoedema and has been reported in dogs (Patterson et al 1967) and cattle (Donald et al 1952, Herrick and Eldridge 1955).

To date the syndrome in cattle has been reported only in Ayrshire calves (Donald et al 1952; Blood et al 1983), and for this reason has been called "inherited lymphatic obstruction of Ayrshire calves" by Jubb and Kennedy (1970). The condition occurred in 12% of calves from Ayrshire cows
TABLE 1
Analysis of the fluid aspirated from the distal limb of a calf with lymphoedema

<table>
<thead>
<tr>
<th>Colour</th>
<th>Clear</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appearance</td>
<td>Turbid</td>
</tr>
<tr>
<td>Specific Gravity</td>
<td>1.012</td>
</tr>
<tr>
<td>pH</td>
<td>8.0</td>
</tr>
<tr>
<td>Total protein</td>
<td>5 gml</td>
</tr>
<tr>
<td>RBC, WBC, bacteria</td>
<td>None evident</td>
</tr>
<tr>
<td>Cellular content</td>
<td>Nil</td>
</tr>
</tbody>
</table>

TABLE 2
The results of haematology and plasma protein estimation from a calf with lymphoedema

<table>
<thead>
<tr>
<th>PCV</th>
<th>0.26</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBC x 10^6/l</td>
<td>3.77</td>
</tr>
<tr>
<td>WBC x 10^6/l</td>
<td>4.3</td>
</tr>
<tr>
<td>Neutrophils %</td>
<td>26.0</td>
</tr>
<tr>
<td>Lymphocytes %</td>
<td>74.0</td>
</tr>
<tr>
<td>Total plasma protein g/ml</td>
<td>60.5</td>
</tr>
<tr>
<td>Albumin g/ml</td>
<td>25.5</td>
</tr>
<tr>
<td>Globulin g/ml</td>
<td>35.0</td>
</tr>
<tr>
<td>A/G Ratio</td>
<td>0.73</td>
</tr>
</tbody>
</table>

The following introduction of a carrier Ayrshire bull (Donald et al 1952). The abnormality appears to be determined by a single autosomal recessive gene influenced by modifiers in calves and by a single autosomal dominant gene in the dog (Jubb and Kennedy 1970).

The extent of the oedema varies from mild to severe. In mild cases the calf may appear normal at birth, whereas in more severe cases the calf may be stillborn and will usually cause dystocia (Blood et al 1983). The oedema of the limbs is generally symmetrical, uniform and diffuse, and distal to the elbows and stifles. The presence of secondary ear lobes has been reported to be associated with this lymphatic defect (Jubb and Kennedy 1970). The growth rate and general condition of the affected animal are usually not impaired.

In view of the fact that this condition is regarded as hereditary and has been reported only in Ayrshire calves, this case record that it can also occur in Ayrshire crossbreds.

A 5-month-old Ayrshire (sire) Friesian (dams) female calf was admitted to the University of Nairobi Veterinary Clinic with a history of swollen hind limbs since birth. On physical examination the only abnormalities detected were symmetrical swelling of both hind limbs below the stifle joint (Figure 1) and swelling of the entire tail. A small accessory ear lobe was present on the left pinna. The calf showed no signs of lameness, and its behaviour, appetite, body size and weight (78.5 kg) were all considered normal.

The swelling of the limbs was warm and painless with palpation and pitted readily on pressure. Fluid aspirated aseptically from the distal limb was a transudate (Table 1) and proved bacteriologically sterile after aerobic incubation at 37°C for 72 h. Laboratory examination revealed mild anaemia, no blood parasites and a slightly lowered plasma protein level (Table 2). It was thought that these mild alterations were unrelated to the condition. Faecal analysis for parasites was not performed.

This report of the condition in an Ayrshire x Friesian calf suggests that the use of a carrier Ayrshire bull could lead to the appearance of the defect in a non-Ayrshire herd.

References
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Recognition of the sucking louse Linognathus africanus on goats

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According to Ferris (1951) there are apparently four distinct species of Linognathus which occur on domestic sheep and goats. Linognathus ovillus, the face louse and L. pedalis, the foot louse, both occur on sheep in Australia (Ferris 1951: Roberts 1952) while L. stenopsis has been observed on goats (Seddon 1967). The fourth species of sucking louse L. africanaus, is mentioned by Roberts (1952) in his text on the insects that affect livestock in Australia in conjunction with L. stenopsis and it appears that he considered this species as a synonym of L. stenopsis. L. africanaus has been reported from Africa (Hopkins 1949), the Philippines (Tongson et al 1981), North America (Emerson et al 1984), Micronesia (Wilson 1972), South Africa (Thorold 1963), Spain (Portus et al 1977), Mexico (Lozoya 1986) and India (Mishra et al 1974) but apparently not from Australia.

On 8 April 1988 lice were collected during a post-mortem