

6. CONCLUSIONS

The development of immunological diagnosis of dicrocoeliosis by ELISA could be of great consequence in countering the shortcomings of coproscopic examinations (i.e. long pre-patent period leading to false negatives) and in detecting early *D. dendriticum* infections in sheep.

Immunological methods can provide early diagnosis of dicrocoeliosis and consequently allow for early treatments, but offer no indications about parasitic burden or stage of infection, as was also the finding of other authors (GONZÁLEZ-LANZA *et al.*, 2000).

Moreover, further studies are required to investigate cross-reactivity of dicrocoeliosis with other concomitant parasitic diseases (i.e. fasciolosis, paramphistomatosis or gastrointestinal strongylosis).

In any case, ELISA methods always should be used together with classical diagnostic methods, anamnestic and coprological investigations. Only in this way can results be rightly interpreted, and false negatives or false positives recognized.

If this test was introduced in routinely use, large amount of antigen would be necessary. At the moment antigens are produced using whole worm cultures, with trematodes from experimentally infected animals. As described, this process is time-consuming and expensive, because the whole parasite life cycle has to be reproduced. Moreover, another disadvantage of this method is the low purity of antigen obtained.

A less expensive option could be the production of recombinant antigens by molecular biology techniques. This could also improve the test sensitivity and specificity, as purification of antigen proteins is guaranteed. Of course the proper antigenic fractions must be firstly isolated and sequenced. This result could be achieved by further immunoblot trials, leading to better defined proteic band patterns of *D. dendriticum*, and afterwards, to identification of antigenic fractions.

As described in the present study, serodiagnosis for helminthiasis is often difficult. Nevertheless, these methods are worth to be implemented. In the case of dicrocoeliosis, the persistence of elevated titres of anti-*D. dendriticum* antibody is a key

factor for seroepidemiological studies. In fact, this characteristic can help to track *Dicrocoelium* infections and to draw risk maps.

The rationale behind accurate diagnosis is that parasitic infections require specific targeted treatments (OTRANTO and TRAVERSA, 2002).

One of the best approaches to control the occurrence of diseases caused by flukes and by other parasitic disease is the implementation of proper husbandry practices and strategic treatments. This can be achieved only through a correct knowledge of parasitological status, which implies a correct diagnosis.

Focused treatments, in terms of timing and dosage, can play a fundamental role in solving the challenge of anthelmintic resistance (e.g. benzimidazole compounds) and in mitigating the economic impact of the disease (OTRANTO and TRAVERSA, 2002).