DETECTION OF ECHINOCOCCUS COPROANTIGENS BY ENZYME-LINKED IMMUNOSORBENT ASSAY IN DOGS FROM THE NORTH-VEST OF ROMANIA

Seres Ştefania¹ , E. Avram², V. Cozma¹

¹University of Agricultural Sciences and Veterinary Medicine, Faculty of Veterinary Medicine, 3-5, Mănăștur Street, 400372, Cluj-Napoca, Romania, email: seresstefania@yahoo.com
²Parasitology and Parasitic Diseases Department of the Sanitary Veterinary and Food Safety Direction, Satu-Mare, Romania

Key words: ELISA, coproantigens, Echinococcus granulosus, dogs.

Abstract: A sandwich enzyme-linked immunosorbent assay (ELISA) was used for the detection of Echinococcus coproantigens in faecal samples from dogs in Satu Mare district. In view of the considerable public health significance of Echinococcus granulosus, the causative agent of the human cystic hydatidosis, there is an urgent need in Romania for reliable and simple techniques for the diagnosis of the infection in individual dogs. This technique should be considered as very important in implementation of the immunologic diagnosis tests in the control programmes of hydatidosis.

The aim of this study was to underline the easy application of the ELISA test Chekit Echinotest (Bommeli, Liebefeld-Bern, Switzerland) and to estimate the prevalence of Echinococcus spp. infection in dogs from four rural regions in Satu-Mare. The studies were performed in October 2005 and July 2006 at the Parasitology and Parasitic Diseases Department of the Sanitary Veterinary and Food Safety Direction, Satu-Mare. 40 dog faeces were collected from the field or from rectum, and examined using the sandwich ELISA enzyme-linked immunosorbent assay. The overall prevalence was 12.5%, respectively five dogs were found positive to Echinococcus spp.

INTRODUCTION

An accurate determination of the prevalence of Echinococcus granulosus in dogs is an essential requirement for establishing epidemiological base-line data, for surveillance and for estimating the potential infection risk of humans in endemic areas (Raoul et al., 2001). Studies performed in our country were lied mostly upon the intermediar hosts, thus is essential to improve the researches concerning the definitive hosts.

In the world the prevalence of cystic hydatid disease in animals is high, although the control programmes have been improved (Berke, 2001). In Romania, the prevalence of cystic hydatidosis is 30-60% in the rumminants (Coman, 2002). In Europe, the prevalence of echinococcosis in dogs and intermediate hosts is different according to the geographical area, but the existent data are of great concern. In certain areas of the western part of Romania there were registered prevalences up to 68% of Echinococcus granulosus in dogs (Morariu, 2004).

The diagnosis of Echinococcus granulosus infection in individual dogs depending on the classical parasitological techniques is unsatisfactory. The detection of adult worms in dogs is based on various methods. Although the most reliable technique for the diagnosis of Echinococcus granulosus infection in dogs as final hosts, due to it’s high sensibility (100%) and specificity (99%), is the parasitological examination of the small intestine at necropsy (Eckert et al., 2001), at present there are preferred the intra vitam methods.
In the past, the arecoline purgation was the method of choice for the diagnosis of echinococcosis in dogs, although it is a labourious, expensive procedure, with biological risque and low sensibility (65%) (Schantz et al., 1995). Recently, some methods have been developed and evaluated with the aim to simplify and improve epidemiological investigations in dog populations and to enable diagnosis of Echinococcus spp. infection in living animals. These methods include the detection of serum antibodies, of coproantigens and of DNA originating from eggs isolated from faecal samples.

As an alternative, in order to diagnose Echinococcus spp. infection in the small intestine of the dogs, various research groups have improved and evaluated the detection of specific coproantigens with the ELISA technique (Allan et al., 1992, Deplazes et al., 1992, Malgor et al., 1997). The ELISA kits are rapid and easy to use in epidemiological screenings. In the endemic areas Stieger et al. (2002) estimated a 88% sensibility of the ELISA.

Furthermore, due to the stability of the coproantigens, the ELISA test can be used in faeces recovered from field (Deplazes et al., 2004). The coproantigens released by adult taeniid parasites can be detected in both patent and pre-patent infections, suggesting that they are the product of active secretion, coat turnover and/or metabolism.

This is the first study upon the diagnosis of Echinococcus spp. in the definitive host (dog in our case) using the ELISA immunological technique in the North-Vest of Romania.

MATERIALS AND METHODS

Aim: The aim of the study was to underline the easy application of the ELISA test Chekit Echinotest (Bommeli, Liebefeld-Bern, Switzerland) for the detection of the specific coproantigens and to estimate the prevalence of Echinococcus spp. infection in dogs from four rural areas in Satu-Mare.

Period: The research was performed in October 2005 and July 2006 at the Parasitology and Parasitic Diseases Department of the Sanitary Veterinary and Food Safety Direction, Satu-Mare.

Experimental design: 40 faecal samples of dogs were collected from the field or from rectum. For safety reasons, faeces were stored at -80°C for at least 5 days before being further processed. The dogs were from rural areas belonging to Paulesti (10 dogs), Acas (11 dogs), Craidorolt (11 dogs) and Socond (8 dogs). The 8 dogs from Socond and 5 from Craidolrolt, were shepherd dogs, being in frequent contact with sheep. The rest of the dogs were from owners living in the rural areas mentioned above.

The faecal samples were examined with the ELISA technique for the detection of coproantigens using the commercially available Chekit Echinotest (Bommeli, Liebefeld-Bern, Switzerland). The Chekit Echinotest is designed for the detection of Echinococcus granulosus and Echinococcus multilocularis coproantigens in dogs, foxes and cats. The test was performed according to the manufacturers instructions, using 1g of faecal material diluted 1:4 in the kit’s sample dilution buffer. The samples were run in duplicates. After centrifugation of the sample suspension (3,000 g at room temperature for 10 min), the supernatants were used for ELISA.

RESULTS AND DISCUSSION

From the 40 dog faecal samples collected and examined using the ELISA technique for the detection of Echinococcus spp. coproantigens, 5 were positive representing an overall prevalence of 12.5% (Fig. 1 and Table 1).
The prevalence of 12.5% in dog echinococcosis indicates the great concern that we should have regarding this disease, especially because of its public health significance. Furthermore, these results underline the importance of an accurate epidemiological study upon echinococcosis in the final hosts in Satu-Mare.

For epidemiological investigations, especially in animal populations with low parasite prevalences, coproantigen detection by ELISA may be the method of choice (Allan et al., 1996a). We cannot omit the fact that this immunological technique has the great advantage of protecting the life of the animal.

Table 1. Prevalence of echinococcosis in rural dogs from Satu-Mare district

<table>
<thead>
<tr>
<th>Rural area</th>
<th>No. of dogs examined</th>
<th>ELISA positive dogs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Nr.</td>
<td>%</td>
</tr>
<tr>
<td>Paulesti</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>Acas</td>
<td>11</td>
<td>2</td>
</tr>
<tr>
<td>Socond</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td>Craidorolt</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>40</td>
<td>5</td>
</tr>
</tbody>
</table>

These are the results of our first studies in the North-Vest of Romania using the ELISA test for the detection of *Echinococcus* coproantigens in dogs. Our research will go on, in order to elucidate the epidemiological status of this parasitic zoonosis in the dog population from this part of the country.

CONCLUSIONS

The researches made in October 2005 and July 2006 on 40 dogs from four rural areas in the North-Vest of Romania (Paulesti, Craidorolt, Acas and Socond) in order to demonstrate the efficacy and easy application of the ELISA immunosorbent assay for the detection of *Echinococcus spp.* coproantigens, have showed the following:

- The commercially available *Chekit Echinotest* (Bommeli, Liebefeld-Bern, Switzerland) test is very comfortable to use in the examination of individual dogs.
- The overall prevalence of the dog population from the four rural areas (Paulesti, Craidorolt, Acas and Socond) was 12.5%. 

397
BIBLIOGRAPHY


