DIAGNOSIS OF GASTROINTESTINAL PARASITES IN CATS: A COMPARISON OF DIFFERENT METHODOLOGIES

[Diagnóstico de parasitoses gastrintestinais de felinos e comparação entre técnicas coproparasitológicas]

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ABSTRACT – We evaluated the effectiveness of the Willis-Mollay, Faust, sedimentation and direct test methods for diagnosing endoparasitic diseases in 198 domestic cats. Comparisons between these techniques were made using the chi-square test (χ²) and Cochran’s Q-test. No infection was detected in 47 (23.7%) out of the 198 cats examined. Ancylostoma spp. proved to be the most important parasite in cats, occurring in 129 animals (65.2%), followed by Cystoisospora spp. in 61 (30.8%), Dipylidium caninum in 18 (9.1%), Toxocara cati in 9 (4.5%), Taenia spp. in 9 (4.5%), Sarcocystis spp. in 2 (1.0%) and Giardia spp. in 2 (1.0%). No association was noticed between the parasites diagnosed and the variables of breed, gender and age of the animals examined (P > 0.05). The Willis technique showed the highest effectiveness in diagnosing Ancylostoma spp. (76 cases; 45.5%), while the direct test had the least effectiveness (49 cases; 29.3%). The Willis and Faust techniques were superior for detecting Cystoisospora spp. and showed the same positivity (30 cases; 18.0%). Most of the Dipylidium caninum cases were diagnosed by means of the sedimentation technique (14 cases; 8.4%). We recommend that the Willis and sedimentation techniques should be used in association for diagnosing gastrointestinal parasites in cats.

Keywords: Cats; Helminths; Protozoa; Parasitological Techniques.

RESUMO – Nós comparamos a eficiência dos métodos de Willis-Mollay, Faust, Sedimentação e Exame Direto no diagnóstico de infecções por helmintos e protozoários em felinos. Um total de 167 gatos domiciliados do Município de Araçatuba, São Paulo, foram examinados por meio das técnicas supramencionadas, sendo que as comparações entre as mesmas foram efetuadas pelo teste Qui-Quadrado (χ²) e pela prova Q de Cochran. Não foi detectada infecção, em 47 (23,7%) dos 198 gatos examinados. Ancylostoma spp. foi o principal parasito de gatos, tendo ocorrido em 129 animais (65,2%), seguido por Cystoisospora spp. em 61 (30,8%), Dipylidium caninum em 18 (9,1%), Toxocara cati em 9 (4,5%), Taenia spp. em 9 (4,5%), Sarcocystis spp. em 2 (1,0%) e Giardia spp. em 2 (1,0%). Não foi observada associação entre os parasitos diagnosticados e a raça, sexo e idade dos animais examinados (P > 0,05). A técnica de Willis teve maior eficiência para o diagnóstico de Ancylostoma spp. (76 = 45,5%), enquanto o Exame Direto mostrou-se o menos eficiente (49 = 29,3%). As técnicas de Willis e Faust foram superiores na deteção de Cystoisospora spp. e apresentaram a mesma positividade (30 = 18,0%). A maioria dos casos de Dipylidium caninum foi diagnosticada pela técnica de Sedimentação (14 = 8,4%). Nós recomendamos a associação das técnicas de Willis e Sedimentação para o diagnóstico de parasitos gastrintestinais de felinos.

Palavras-Chave: Gatos; Helmintos; Protozoários; Técnicas Parasitológicas.

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INTRODUCTION

Because of the close bonds between humans and domestic animals, there is a need to know about the frequency of occurrences of endoparasites in cats that live in human homes, especially because of the zoonotic potential of these enteroparasites (Riggio et al., 2013).

Various methods are used for fecal examinations in cats, including the direct fecal examination, Willis-Mollay and sedimentation methods, Faust method and Sheather method (Ketzis et al., 2015).

Given that these fecal examination techniques are essential for diagnosing intestinal parasites and therefore for treating infected animals, assessment of the efficiency of these techniques is fundamental. However, only a few scientific papers have made comparisons between the results obtained from different laboratory tests methods relating to cats.

We evaluated occurrences of endoparasitic diseases and their associations with the variables of the animals’ breed, gender and age, and also analyzed the efficiency of four techniques that are routinely used for making laboratory parasitological diagnoses on stool samples from cats.

MATERIAL AND METHODS

The experimental group was composed of 198 cats that lived in human homes, of both genders and different breeds and ages. Dental arch analysis was used to classify cats aged up to six months as young and those aged seven months or over as adults. However, the age of one animal in this study was not registered.

All of these animals were sent by their owners to the city of Araçatuba/SP, during the year 2007, to be put down. In this procedure, thiopental 2% in association with potassium chloride was administered intravenously, in accordance with the principles established by the Animal Experimentation Ethics Committee, under protocol number 2007-006842.

Stool samples were processed by means of the following methods: flotation or Willis-Mollay, using a saturated sodium chloride solution with density of 1.182 (Willis, 1921); centrifugal flotation or Faust, using zinc sulphate solution with a density of 1.200 (Faust et al., 1938); spontaneous sedimentation in water (Hoffman et al., 1934) and direct fecal examination.

The samples from 31 cats could not be processed using these four techniques because of the small amount of fecal matter available. In these cases, the samples were processed by means of the Willis and direct examination methods and also, whenever possible, using the Faust method. For this reason, comparison of the efficiency between the direct examination, Willis, Faust and sedimentation techniques were performed with only 167 animal stool samples.

The results obtained through these techniques were compared using Cochran’s Q test (Curi, 1998). The existence of associations between the variables studied and the presence of endoparasites was evaluated using the chi-square test (χ²) (Zar, 1999). The significance level was 5% and the statistical analyses were performed using the SAS software (1999).

RESULTS

In 23.7% (47/198) of the animals examined, no intestinal parasites were recorded. In 42.9% (85/198) of the cats, only one parasite genus was identified. Mixed infections with two, three and four genera occurred in 57 (28.8%), five (2.5%) and four (2.0%) animals, respectively. *Ancylostoma* spp. was detected in 129 cats (65.2%), followed by *Cystoisospora* spp. in 61 (30.8%), *Dipylidium caninum* in 18 (9.1%), *Toxocara cati* in nine (4.5%), *Taenia* spp. in nine (4.5%) and *Sarcocystis* sp. and *Giardia* spp., in two animals each (1%).

No associations between the parasites and the animals’ ages were observed (P > 0.05) (Table 1). There were also no associations between occurrences of the parasites and the variables of breed and gender (P > 0.05) (Tables 2 and 3).

| Table 1. Occurrence of *Ancylostoma* spp., *Dipylidium caninum* and *Cystoisospora* spp. in stool samples from 109 and 88 cats aged up and above six months, respectively. |
|-----------------|-----------------|-----------------|-----------------|
| Parasite       | Up to six months | Above six months | P-value*       |
| *Ancylostoma* spp. | 65 (59.6)        | 63 (71.6)        | Ns             |
| *Cystoisospora* spp. | 37 (33.9)        | 24 (27.3)        | Ns             |
| *Dipylidium caninum* | 11 (10.1)        | 7 (8.0)          | Ns             |

χ²-test*: Ns = not significant (P > 0.05). The percentages are given in parenthesis.
Table 2. Occurrence of *Ancylostoma* spp. and *Dipylidium caninum* eggs and *Cystoisospora* spp. oocysts in fecal samples from 115 female and 83 male cats.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Females</th>
<th>Males</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ancylostoma</em> spp.</td>
<td>78 (67.8)</td>
<td>51 (61.5)</td>
<td>Ns</td>
</tr>
<tr>
<td><em>Cystoisospora</em> spp.</td>
<td>39 (33.9)</td>
<td>22 (26.5)</td>
<td>Ns</td>
</tr>
<tr>
<td><em>Dipylidium caninum</em></td>
<td>9 (7.8)</td>
<td>9 (10.8)</td>
<td>Ns</td>
</tr>
</tbody>
</table>

χ²-test*: Ns = not significant (P > 0.05).
The percentages are given in parenthesis.

Table 3. Occurrence of *Ancylostoma* spp. and *Dipylidium caninum* eggs and *Cystoisospora* spp. oocysts in fecal samples from 180 mongrel cats and 18 pedigree cats.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Mongrel cats</th>
<th>Pedigree cats</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ancylostoma</em> spp.</td>
<td>114 (63.3)</td>
<td>15 (83.3)</td>
<td>Ns</td>
</tr>
<tr>
<td><em>Cystoisospora</em> spp.</td>
<td>58 (32.2)</td>
<td>3 (16.7)</td>
<td>Ns</td>
</tr>
<tr>
<td><em>Dipylidium caninum</em></td>
<td>16 (8.9)</td>
<td>2 (11.1)</td>
<td>Ns</td>
</tr>
</tbody>
</table>

χ²-test*: Ns = not significant (P > 0.05).
The percentages are given in parenthesis.

The comparisons of efficiency between the direct fecal examination, Willis, Faust and sedimentation methods for diagnosing intestinal parasites in cats' stool samples are shown in Table 4.

Table 4. Comparison of the efficiency of Direct Examination, Willis, Faust and Sedimentation methods for diagnosis of intestinal parasites in stool samples from 167 cats.

<table>
<thead>
<tr>
<th>Diagnosed Parasites</th>
<th>Direct</th>
<th>Willis</th>
<th>Faust</th>
<th>Sedimentation</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ancylostoma</em> spp.</td>
<td>49 (29.3)</td>
<td>76 (45.5)</td>
<td>62 (37.1)</td>
<td>72 (43.1)</td>
<td>P &lt; 0.001</td>
</tr>
<tr>
<td><em>Cystoisospora</em> spp.</td>
<td>16 (9.6)</td>
<td>30 (18.0)</td>
<td>30 (18.0)</td>
<td>19 (11.4)</td>
<td>P &lt; 0.01</td>
</tr>
<tr>
<td><em>Dipylidium caninum</em></td>
<td>13 (7.8)</td>
<td>12 (7.2)</td>
<td>2 (1.2)</td>
<td>14 (8.4)</td>
<td>P &lt; 0.001</td>
</tr>
<tr>
<td><em>Taenia</em> spp.</td>
<td>5 (3.0)</td>
<td>1 (0.6)</td>
<td>0 (0)</td>
<td>6 (3.6)</td>
<td>*</td>
</tr>
<tr>
<td><em>T. cati</em></td>
<td>1 (0.6)</td>
<td>2 (1.2)</td>
<td>2 (1.2)</td>
<td>3 (1.8)</td>
<td>*</td>
</tr>
<tr>
<td><em>Giardia</em></td>
<td>1 (0.6)</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>2 (1.2)</td>
<td>*</td>
</tr>
<tr>
<td><em>Sarcocystis</em></td>
<td>0 (0)</td>
<td>1 (0.6)</td>
<td>0 (0)</td>
<td>1 (0.6)</td>
<td>*</td>
</tr>
</tbody>
</table>

χ²-test: *Cochran’s Q-test was not applied to these parasites due to their low occurrence.
The percentages are given in parenthesis.

**DISCUSSION**

Direct examination was generally less efficient for diagnosing helminths and protozoa. Thus, we recommend that the Willis-Mollay and sedimentation methods should be combined to detect these parasites. We also emphasize the importance of educational measures for controlling parasitic zoonosis, since the rate of occurrence of parasites in the animals analyzed was quite high.

Infections caused by parasites are observed especially in cats aged less than one year, since adult animals usually have acquired a certain degree of immunity, especially against roundworms (ascarids) (Martínez-Barbosa, 2003).

*Ancylostoma* spp. was the most common parasite in cats in our study (with occurrence in 62.5%). Lower proportions (Serra et al., 2003) and higher proportions (Krecek et al., 2010) have previously been reported.

High prevalence of *Ancylostoma* was found in the necropsies on cats in the city of Araçatuba (Ishizaki et al., 2006). Those results differed from those in the studies by Gennari (2001), Ragozo et al. (2002), Robben et al. (2004), Sohn (2005), Gates (2009), Mircean (2010), Capári et al. (2013) and Riggio et al. (2013), who all reported that *Toxocara* spp was predominant. These differences in results were possibly due to the fact that most of the animals in the present study were adults, which are more resistant to *Toxocara* spp. infection.

The occurrence rate found for *D. caninum* (9.1%) was lower, since this parasite is commonly diagnosed through findings of proglottids in fecal matter or through adult-stage helminths in necropsies (Gennari et al., 1999). Most findings of *D. caninum* were made by means of the sedimentation technique (14 cases; 8.4%), although the Willis and direct examination techniques showed similar values (12 cases; 7.2%; and 13 cases; 7.8%, respectively) (Table 4). Sloss et al. (1999) considered that the sedimentation technique
was the one most indicated for recovery of some cestodes; and Táparo et al. (2006) confirmed its applicability for detection of this endoparasite in dogs.

For diagnosing *Ancylostoma* spp., *Cystoisospora* spp. and *D. caninum*, significant differences were observed between the four techniques used (P < 0.001). The Willis technique showed the highest efficiency for diagnosing *Ancylostoma* spp. eggs (76 cases; 45.5%), while direct examination proved to be the least efficient method (49 cases = 29.3%). This was also observed in dogs by Táparo et al. (2006).

Samples positive for *Cystoisospora* spp. were found more frequently through using the Willis technique (30 cases; 18.0%) and Faust technique (30 cases; 18.0%), thus showing higher efficiency for diagnosing this protozoon. It is noteworthy that the centrifugation-flotation method was more efficient than Willis and sedimentation with regard to examination of canine stool samples (Oliveira-Sequeira et al., 2002).

In the present study, only one stool sample from each animal was examined and no *Giardia* spp. cysts were detected using the Faust method. The results regarding occurrences of *Giardia* spp. in this study, through fecal tests, may have been underestimated, given the likelihood of intermittent cyst elimination, particularly in felines (Melo et al., 2003). For diagnosing *Giardia* diagnosis, the recommendation is that three samples should be taken on alternate days (Heymans et al., 1987; Adam, 1991; Mundin & Cury, 2002), in order to maximize the chance of detecting the parasite.

In conclusion, we recommended that a combination of the Willis-Mollay and sedimentation methods should be used for detecting helminth eggs and protozoan oocysts in stool samples from cats.

**CONFLICT OF INTEREST**

The authors declare no conflict of interest.

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