Full Length Research Paper

Prevalence and risk factors associated with faecal shedding of *Cryptosporidium* oocysts in piglets, Kaduna, Nigeria

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Cryptosporidiosis is predominant in piglets and other young farm animals and the infection can be severe resulting in financial loss to producers. Faecal samples from one hundred and thirty-two piglets were examined for *Cryptosporidium* oocysts using formol-ether concentration method and modified Ziehl-Neelsen staining technique. *Cryptosporidium* oocysts were found in 18 (13.6%) of the faecal samples and the prevalence were higher among males (20.0%) than females (5.3%), younger (19.0%) than older (8.7%) piglets, piglets raised under intensive (16.7%) than from under semi-intensive/extensive systems (0%), piglets with loose faeces (28.6%) than from well formed faeces (10.8%) and piglets that drank bore hole water (13.8%) than from those that drank well water (13.3%). There were associations between prevalence of *Cryptosporidium* oocysts and sex of piglets (OR = 0.22; 95% CI on OR: 0.05 < OR < 0.88), faecal consistencies (OR = 3.30; 95% CI on OR: 0.93 < OR < 11.47) and management systems (P < 0.05). On the other hand, there was no association between prevalence of *Cryptosporidium* oocysts and type of water and age of the piglets (P > 0.05). This study shows that sex, faecal consistencies and management systems influence the prevalence of *Cryptosporidium* oocysts in faeces of piglets in Kaduna metropolis.

Key words: Cryptosporidium, piglets, faeces.

INTRODUCTION

*Cryptosporidium* spp. are common parasites of humans, domestic animals and wild vertebrates and because of the wide host range of the organism, cryptosporidiosis has been considered to be a zoonotic disease for some time now (Xiao and Feng, 2008).

Cryptosporidiosis has long been a veterinary problem, being predominant in young farm animals such as calves (Avery et al., 2007). Calves, lambs, piglets and goat kids can become severely ill following infection resulting in financial loss to producers (Santin and Trout, 2007). Many species of livestock harbour *Cryptosporidium* species that are infectious for humans (Santin and Trout, 2007). *Cryptosporidium parvum* for example has received the most attention in zoonotic transmission of cryptosporidiosis because it is a major human pathogen and traditionally infects all mammals (Xiao and Feng, 2008). Recent genetic characterization studies have revealed that pigs are infected with a genetically distinct and apparently host-adapted form of *Cryptosporidium* (*Cryptosporidium* "pig" genotype) (Ryan et al., 2003). Pigs can also be infected with the zoonotic *C. parvum* "cattle" genotype, indicating that they can potentially play a role as reservoirs of infection for humans and other animals (Morgan et al., 1999). Sequencing and phylogenetic analysis of 18S rDNA by Ryan et al., 2003 identified two distinct genotypes of Cryptosporidium: the previously identified pig genotype (Morgan et al., 1999) and a novel pig genotype (pig genotype II).

Cryptosporidiosis was rarely reported in humans until 1982 when most of the earliest human cases were reported in animal handlers and the number of detected human cases began to rise rapidly alongside the AIDS pandemic (Avery et al., 2007). The close association of humans and livestock as well as ability of run-off from animal production operations to contaminate water supplies represents an ever present risk of human infection.
Table 1. Odds ratio (OR) and 95% confidence intervals on factors influencing the prevalence of faecal shedding of Cryptosporidial oocysts in Kaduna metropolis, Nigeria.

<table>
<thead>
<tr>
<th>Factors</th>
<th>No of faeces sampled</th>
<th>Positive samples</th>
<th>Specific rate (%)</th>
<th>Odds ratio (OR)</th>
<th>95% Confidence interval on OR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male ref</td>
<td>75</td>
<td>15</td>
<td>20.0</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>57</td>
<td>3</td>
<td>5.3</td>
<td>0.22*</td>
<td>0.05 – 0.88</td>
</tr>
<tr>
<td>Faecal Consistency</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Loose ref</td>
<td>21</td>
<td>6</td>
<td>28.6</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>111</td>
<td>12</td>
<td>10.8</td>
<td>3.30*</td>
<td>0.93 – 11.47</td>
</tr>
<tr>
<td>Age of piglets (weeks)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1-10 ref</td>
<td>63</td>
<td>12</td>
<td>19.0</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>11-20 ref</td>
<td>69</td>
<td>6</td>
<td>8.7</td>
<td>2.47</td>
<td>0.79 – 8.02</td>
</tr>
<tr>
<td>Water</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Well ref</td>
<td>45</td>
<td>6</td>
<td>13.3</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Bore hole</td>
<td>87</td>
<td>12</td>
<td>13.8</td>
<td>0.96</td>
<td>0.29 – 3.01</td>
</tr>
<tr>
<td>Management practices</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intensive ref</td>
<td>108</td>
<td>18</td>
<td>16.7</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Semi-int/ext.</td>
<td>24</td>
<td>0</td>
<td>0</td>
<td>0.00**</td>
<td>0.00 – 1.13</td>
</tr>
</tbody>
</table>

Reference standards (ref).
*p < 0.05 is significant; **P < 0.05 is significant for Fisher’s exact test.

(Santin and Trout, 2007). Though a greater under-standing of Cryptosporidium infection is critical from two perspectives, animal health and human health (Santin and Trout, 2007), the role of animals, especially farm animals and domestic pets in the transmission of human Cryptosporidium is nevertheless not clear (Xiao and Feng, 2008). Thus, this research was aimed at studying the prevalence and risk factors associated with faecal shedding of Cryptosporidium in piglets in Kaduna metro-polis, so that potential sources of transmission could be identified for possible control measures.

MATERIALS AND METHODS

The study was conducted between July and September, 2008. A total of one hundred and thirty two piglet faeces from various private farms in Kaduna metropolis, Nigeria were examined. The following information about the piglets examined in the study was obtained by the use of a questionnaire: age, sex, management systems, source of drinking water and presence of diarrhoea or loose faeces.

Faecal samples were collected rectally by use of polythene bags and brought to the laboratory immediately. If not tested upon collection, the samples were kept at 4°C in the refrigerator for a maximum of three days. The faecal samples were treated using formal-ether concentration method and stained using modified Ziehl-Neelsen technique (WHO, 1991). Briefly, about 1gm of faeces was mixed in 10 ml of 10% formalin in a universal bottle using an applicator stick. The homogenized faeces were sieved into a centrifuge tube using a funnel and gauze to which 3 ml of diethyl-ether was added to extract fat from the filtrate. The centrifuge tube was corked and shaken gently to mix properly. The tube was centrifuged at 5000 x g and supernatant decanted. The sediment was mixed, from which a thin smear was made on a clean glass slide. After air-drying, the smear was fixed in methanol for 2 - 3 min. The slide was flooded with cold carbol fuschin for 5 - 10 min and then with 1% hydrochloric-acid ethanol until colour ceases to flow out and rinsed in tap water. It was then counterstained with 0.25% methylene blue for 30 s, rinsed in tap water again and air-dried. The slide was examined microscopically using x 40 magnification. The cryptosporidial oocysts appeared as bright rose pink spherules on a pale green background.

Positives slides used were photomicrographs of oocysts of C. parvum from Department of Parasitic Diseases, Centers for Disease Control and Prevention (CDC), Atlanta, USA.

Association between various risk factors and the occurrence of Cryptosporidium infection was determined using chi-square test for association where appropriate with the use of Statistical Package for Social Sciences (SPSS). P < 0.05 was defined as significant. Odds ratio (OR) and 95% confidence intervals were calculated for dichotomous variables using EP1 INFO 2002 (Dean, 1994). OR values greater than unity denote association and less than unity denote that the factor may have a protective effect.

RESULTS

Out of the 132 piglet faeces examined, 18(13.6%) were positive for Cryptosporidium oocysts.

Table 1 shows the relationship between the prevalence of Cryptosporidial oocysts and sex, faecal consistency, source of drinking water, age and management practice. The prevalence was higher among males 15(20.0%) than females 3(5.3%), piglets with loose faeces 6(28.6%) than from well formed faeces12 (10.8%), younger (11 to 20 weeks old) 12(19.0%) than older (11 to 20 weeks old) piglets 6(8.7%). There was no much difference in the detection rate between piglets that drank bore hole 12(13.8%) from those that drank well water 6(13.3%) while all the 18 (16.7%) positive cases were raised under intensive system of management. There were associations between
prevalence of Cryptosporidium oocysts and sex of piglets (OR= 0.22; 95% CI on OR: 0.05 < OR < 0.88), faecal consistencies (OR= 3.30; 95% CI on OR: 0.93 < OR < 11.47) and management systems (P < 0.05). However, these associations were not statistically significant except for the sex that showed statistical significance. On the other hand, there was no association between prevalence of Cryptosporidium oocysts and type of water and age of the piglets (P > 0.05).

**DISCUSSION**

The 13.6% rate recorded in this study is lower than the previous reports of 15. 7% in piglets in some parts of Kaduna state (Kwaga et al., 1988) and 32.6% in pigs in Ille-Ife (Ayeni et al., 1985). Quilez et al. (1996) recorded a prevalence of 21.9% of C. parvum oocysts in pigs in Aragón, Northeastern Spain. The differences in the detection rates may be due to modifications in the techniques used and also differences in sampling periods as there has been noted seasonal variations in the prevalence of Cryptosporidium (Wuhib et al., 1994). Also, it has been found that faecal shedding of oocysts in infected animals coincides with clinical illness and mucosal damage as oocysts are often excreted in low numbers requiring more than one faecal sample to detect those shedding the oocysts (Tzipori et al., 1983; Quilez et al., 1996; Hannahs, 2007).

The higher detection rate in male piglets than in females is in agreement with the findings in other species of animals where Cryptosporidium oocysts were detected more in males than females (Rajkhowa, 2006; Ibrahim et al., 2007). This high rate may be because males are more likely than females to disperse to other colonies or be moved to other pens especially when the females are on heat thereby promoting the dissemination of the oocysts.

Even though none of the piglets examined had watery diarrhoea, those that had loose faeces still showed a higher prevalence than those with normal faeces because the organism affects the gastrointestinal system and the severity of the infection is dependent on the host immune system (Clark, 1999; Shukla et al., 2006). There can also be an in apparent carriage of the infection in some instances.

The high prevalence among the younger age group may be because; Cryptosporidium is considered a problem in newborn farm animals (Avery et al., 2007; Santin and Trout, 2007). This high prevalence in younger age group is contrary to the finding of Quilez et al. (1996) who failed to detect oocysts in suckling piglets. Some studies have also shown age related differences in the prevalence of different species of Cryptosporidium infecting pigs (Santin and Trout, 2007).

Though there has been reports of water outbreak of Cryptosporidium (Mackenzie et al., 1994; Rose et al., 2002), no record so far has implicated bore hole water as as source of cryptosporidial infections. The high prevalence of the oocysts among the piglets that drank from bore hole may have been associated with the system of management as the oocysts may have been easily spread among the piglets under intensive system due to faeco-oral route of transmission of the oocysts that may have been perpetuated by the close contact seen during such intensive system.

In view of the fact that Cryptosporidium is one of the causes of diarrhoeal illness in man and animals worldwide, close monitoring of the infection in farm animals such as piglets is required since there can be zoonotic transmission of the infection which is not only of public health significance but also of a great economic importance.

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