A coproscopical study of helminthosis in domestic ruminants (cattle, buffalo, goat) in Khammouane Province (Lao PDR)

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SUMMARY

The purpose of this study was to investigate the prevalence of digestive and respiratory helminthiasis in cattle, buffalo and goat in Khammouane Province through a coproscopical survey and to assess the risk factors associated with gastrointestinal strongylosis (GIS). Four hundred and seven animals were randomly selected as a convenient sampling strategy representing the categories of interest: host species (cattle, buffalo, goat), physiological status (age classes, lactation status), husbandry practices (free grazing, food supplementation). Coproscopic examinations were made by individual quantitative McMaster technique (digestive nematodes) and group qualitative Stoll (trematodes) and Baermann (respiratory strongyles) techniques. For all host species, the most prevalent class of digestive nematodes was GIS (cattle: 69%, buffalo: 18%, goat: 72%) while prevalences for Trichuris sp., Capillaria sp. or Strongyloides sp. were much more lower (0 to 6%). The most prevalent trematode (at group level) was rumen fluke whereas the opposite was seen for goat. Physiological status and husbandry practices had no significant effect on GIS infection.

Keywords: Cattle; Buffalo; Goat; Helminths; Lao PDR

RÉSUMÉ

Etude coproscopique des helminthoses des ruminants domestiques (bovins, buffles, caprins) dans la province de Khammouane (Laos)

La prévalence des helminthoses digestives et respiratoires des bovins, buffles et caprins ainsi que les facteurs de risque associés aux strongyloses gastro-intestinales ont été déterminés par analyse coproscopique dans la province de Khammouane, Laos. Quatre cent sept animaux ont été prélevés lors d'un échantillonnage de convenance visant à représenter les catégories d'intérêt : espèces hôtes (bovins, buffles, caprins), statut physiologique (classes d'âge, lactation), pratiques d'élevage (pâturage libre, supplémentation). Les analyses coproscopiques ont été réalisées par la technique quantitative de McMaster (nematodes digestifs) sur prélèvements individuels, et par les techniques qualitatives de Stoll (trématoïdes) et de Baermann (protostrongyles) sur prélèvements de groupe. Pour les trois espèces hôtes, le groupe de parasites digestifs le plus prévalent a été les strongyles gastro-intestinaux (SGI) (bovins : 69%, buffles : 18%, caprins : 72%) tandis que les prévalences pour Trichuris sp., Capillaria sp. ou Strongyloides sp. ont été beaucoup plus basses (0 à 6%). Le type de trématode le plus prévalent (prévalence de groupe) pour tous les hôtes a été celui des paraplastihomes (bovins : 95%, buffles : 80%, caprins : 100%) tandis que Fasciola sp. n'a été détecté que dans quelques coproscopies de groupe (bovins : 14%, buffles : 7%, caprins : 6%). Des larves de protostrongyles ont été retrouvées dans 20 % des coproscopies de groupe. Les valeurs coproscopiques moyennes pour les SGI ont été de 123, 21 et 474 oeufs/g de fèces respectivement chez les bovins, les buffles et les caprins. Chez les buffles, les animaux adultes ont montré des prévalences et des valeurs coproscopiques plus basses tandis que l'inverse était constaté chez les caprins. Ni le status physiologique, ni les pratiques d'élevage n'ont montré de relations significatives avec l'infestation par les SGI.

Mots-clés : Bovin, Buffle, Caprin, Helminthe, Laos

Introduction

The Lao People's Democratic Republic (Lao PDR) is a Southeast Asian country with a tropical climate. It is a predominantly rural society with 85% of the population depending on agriculture for their livelihood with most of the rural households producing food for their own consumption [20]. About 90% of all households keep one or more species of livestock among which buffalo (1.1 million head) and cattle (1.8 million head) are the main ruminant species compared to goats and sheep (421,000 head) (Department of Livestock and Fisheries, Ministry of Agriculture and Forest, 2015). Lao indigenous livestock are mainly kept in low input systems and suffer from poor management, inadequate nutrition and minimal health care [23]. In lowland areas planted to paddy rice, available feed resources limit expansion of cattle and buffalo production [20]. Helminth parasites are a major animal health constraint to the poor keepers of livestock throughout the world particularly in the tropics/subtropics zone [8] where favorable environmental conditions for development and survival of infective stages and occurrence of highly pathogenic helminths (Haemonchus, Toxocara, Fasciola) are combined [22]. Previous published data on helminth infection of livestock in Lao PDR are few and indicated prevalences of 17 to 34% for Fasciola gigantica and of 23 % for Toxocara vitulorum in buffalo and cattle [12, 13]. Another study was conducted on gastrointestinal strongylosis of goat and cattle but was limited to a small number of animals [11].

The aim of this study was to investigate the prevalence of digestive and respiratory helminthiasis in cattle, buffalo and...
goat through coproscopic survey in Khammouane Province and to assess the risk factors associated with gastrointestinal strongylosis (GIS).

Materials and methods

STUDY AREA AND SAMPLING STRATEGY

The study was conducted in May and June 2015 in Khammouane Province (center of Lao PDR). A high level of poverty occurs in this Province, with 31.4% of inhabitants considered as poor [24]. The wet season lasts from May to October with cooler temperatures and the dry season from November to April. The annual rainfall is 2 381 mm/year with 80% occurring during rainy season. During the dry season, most ruminants graze freely during the day on crops residues and along roads, and are brought in enclosed paddocks for the night. During the rainy season, some cows and some buffalos are attached or put in closed yards during the day to avoid them eating crops. Most of ruminants do not receive any food supplement, but some of them, mostly during rainy season and the attached animals, are given rice straw or foliage.

Eight villages located in lowland rice area in the Xaibouathong district (Seevilaï, Nonghtad, Phahoy, Phakhong, Pakuaynongborn, Pakuaythong, Nakham and Dongnakham) were selected for the major part of sampling (371 animals) as they were involved in the rural development project of the French NGO AVSF (Agronomes et Vétérinaires Sans Frontières). Few additional sampling (36 animals) was performed in villages of Hinboun district (north-west of Khammouane province) thanks to the French NGO CEVEO (Coopération et Échanges Vétérinaires Est-Ouest). Animals were randomly selected in each village as a convenient sampling strategy representing the categories of interest: host species (cattle, buffalo, goat), physiological status (age classes, lactation status), husbandry practices (free grazing, food supplementation). Our sampling has been performed at the very beginning the wet season; most animals included in the study were also grazing freely. Three age class were defined for cattle and buffalo (≤ 3 months; >3 months ≤3 years;>3 years) and for goat (≤ 3 months;>3 months ≤3 years;>1 year). A questionnaire including management and animal information was fulfilled. None of the animals was defined for cattle and buffalo (≤ 3 months;>3 months ≤3 years;>3 years) and for goat (≤ 3 months;>3 months ≤3 years;>1 year). A questionnaire including management and animal information was fulfilled. None of the animals was given anthelmintic prior to fecal sampling and, generally, no anthelmintic was given routinely to the animals whatever the species.

FECAL SAMPLE COLLECTION AND ANALYSIS

Individual faecal samples were collected rectally, placed in a cooler box and carried to the parasitology laboratory of Thakhek hospital. Digestive nematode eggs were counted (Faecal Egg Count-FEC) in all individual faecal samples using a modified McMaster technique as described by RAYNAUD [16] with a saturated salt solution as flotation fluid (sensitivity: 50 eggs/g of faeces -epg-). Due to more labor-intensive techniques, faeces for trematode eggs and lungworm larvae recovery were processed at group level. Composite faecal samples were produced when mixing equal amounts of faeces from 7 to 10 animals to give one sample. Trematode eggs recovery was performed on adults (goats>1 year; cattle and buffalos>3 years). Lungworm larvae recovery was performed on goats >3 months and on cattle and buffalos >3 years. Sedimentation (Stoll) technique was used for trematode eggs, with a sodium hydroxide solution as sedimentation solution (sensitivity: 3 epg) while Baermann technique was used for lungworm larvae (sensitivity: 0.1 and 0.2 lpg for goat and cattle/buffalo groups respectively) as described by EUZEBY [3].

STATISTICAL ANALYSIS

The following quantitative descriptors were used for digestive nematode eggs: individual prevalence, mean intensity of excretion calculated from infected animals and mean FEC (or mean abundance of excretion) calculated from both infected and non-infected animals. For gastrointestinal strongyles (GIS), factors of variation of mean FEC included age classes, sex, lactation (Y/N), food supplementation (Y/N) and free grazing (Y/N). Chi-square/Fisher exact tests and Mann-Whitney/Kruskal-Wallis tests were used for prevalence and mean FEC comparisons respectively, in bivariate analysis. Differences were considered significant when P<0.05. For trematodes and lungworms, prevalences were calculated from composite (group) faecal results (group prevalence).

Results

Four hundred and seven fecal samples from 170 cattle, 61 buffalos and 176 goats were analysed. For all host species, the most prevalent group of parasites was GIS (cattle: 69%, buffalo: 18%, goat: 72%) while prevalences for Trichuris sp., Capillaria sp. or Strongyloides sp. were much more lower and ranged between 0 and 6% according to the host species. Regarding Trichuris sp. infection in goats, eggs were only observed from Stoll technique using composite samples. As far as group prevalences were concerned, the most prevalent trematode was rumen fluke in all host species (cattle: 95%; buffalo: 80%; goat: 100%) whereas Fasciola sp. was detected in only few groups of animals (cattle: 14%; buffalo: 7%; goat: 6%). Small lungworm larvae were found in 20% of goat groups (Table I).

Through bivariate analysis, sex and age were significantly associated to GIS prevalence or mean FEC (Table 2). Prevalence of GIS was significantly higher in goat and cattle compared to buffalo. Mean FEC were 474, 123 and 21 epg for goat, cattle and buffalo respectively. The effect of sex was not straightforward as it varied according to the host species, higher prevalence or FEC in male buffalo and cattle respectively and higher prevalence in goat female. Age showed a strong impact on GIS prevalence with differences between host species: older animals exhibited lower
prevalence in buffalo unlike goat where figures increased steadily with age. In contrast, no significant impact of age on GIS prevalence was seen for cattle. Regarding the effect of age on mean FEC, a similar tendency was observed: older cattle and buffalos had lower mean FEC, whereas mean FEC increased significantly in older goats (Table II). Lactation status, food supplementation and type of grazing had no significant effect on both GIS prevalence and mean FEC.

**Discussion**

GIS prevalences around 70% indicated that this infection is quite common in ruminants of Khammouane Province at least for cattle and goat. Mean FEC globally showed that goats shed 4 times more eggs than cattle and that buffalo both exhibited very low GIS prevalence and mean FEC. Previous data on ruminant coproscopical exams from Lao PDR are restricted to the study of OTAKE SATO et al. [11] showing also higher prevalence and FEC in goat (93 % and 1728 epg) and lower figures in cattle (36% and 86 epg) (14 and 11 animals respectively). Regarding cattle GIS infection, data from traditional grazing system in Vietnam and Cambodia have shown that FEC ranged from 15 to 177 epg according to the age, the maximum of shedding occurring before 7 months of age [2, 5]. These results are in agreement with our own study where the age class of 3 months-3 years showed the maximum prevalence (79%) and egg output (156 epg). However, all these figures for cattle suggest a rather low infection rate for GIS that could be related to the indigenous breed raised and even more to the traditional grazing system which could limit the intake of infective larvae as grazing time, collection of faeces for

<table>
<thead>
<tr>
<th>Type of eggs or larvae</th>
<th>Cattle</th>
<th>Buffalo</th>
<th>Goat</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of individual samples</td>
<td>170</td>
<td>61</td>
<td>176</td>
</tr>
<tr>
<td>No. of infected animals</td>
<td>117</td>
<td>11</td>
<td>126</td>
</tr>
<tr>
<td>Individual prevalence (%) [CI at 95%]</td>
<td>178.2 +/- 160.5</td>
<td>113.6 +/- 77.8</td>
<td>661.5 +/- 708.2</td>
</tr>
<tr>
<td>Mean intensity of excretion (epg)</td>
<td>Mean FEC (epg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nematodes</td>
<td>Trichuris sp.</td>
<td>6 [3-10]</td>
<td>3 [1-11]</td>
</tr>
<tr>
<td>Individual prevalence (%) [CI at 95%]</td>
<td>50.0 +/- -0.0</td>
<td>/</td>
<td>/</td>
</tr>
<tr>
<td>Mean intensity of excretion (epg)</td>
<td>Mean FEC (epg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Capillaria sp.</td>
<td>6 [3-10]</td>
<td>3 [1-11]</td>
<td>10 [6-15]</td>
</tr>
<tr>
<td>Individual prevalence (%) [CI at 95%]</td>
<td>4960.0 +/- -14 076.7</td>
<td>50.0 +/- -0.0</td>
<td>120.6 +/- -83.0</td>
</tr>
<tr>
<td>Mean intensity of excretion (epg)</td>
<td>Mean FEC (epg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Strongyloides sp.</td>
<td>291.8 +/- -3452.9</td>
<td>1.6 +/- -9.0</td>
<td>11.6 +/- -43.7</td>
</tr>
</tbody>
</table>

*eggs of *Trichuris sp.* in goats were only seen in composite samples for Trematodes: the prevalence given in the table is thus a group prevalence

(1) McMaster technique (2) Stoll technique (3) Baermann technique

**Table I**: Prevalence, mean intensity of excretion and mean FEC of gastrointestinal and pulmonary helminths from 407 coproscopical examinations according to host species (cattle, buffalo, goat) in Khammouane Province of Lao PDR.
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<table>
<thead>
<tr>
<th>Risk factors</th>
<th>Species</th>
<th>Factors studied</th>
<th>Total No.</th>
<th>Prevalence (%) (1)</th>
<th>Mean FEC (epg) (2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>Cattle</td>
<td>♀ / ♂</td>
<td>128 / 42</td>
<td>66 / 79 (ns)</td>
<td>110 / 163 (&lt;0,05)</td>
</tr>
<tr>
<td></td>
<td>Buffalo</td>
<td>♀ / ♂</td>
<td>43 / 17</td>
<td>9 / 41 (&lt;0,01)</td>
<td>13 / 41 (ns)</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>♀ / ♂</td>
<td>127 / 47</td>
<td>77 / 57 (&lt;0,05)</td>
<td>510 / 383 (ns)</td>
</tr>
<tr>
<td>Lactation</td>
<td>Cattle</td>
<td>(♀&gt;3 years) Yes / No</td>
<td>39 / 17</td>
<td>67 / 47 (ns)</td>
<td>115 / 91 (ns)</td>
</tr>
<tr>
<td></td>
<td>Buffalo</td>
<td>(♀&gt;3 years) Yes / No</td>
<td>11 / 11</td>
<td>0 / 0 (ns)</td>
<td>0 / 0 (ns)</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>(♀&gt;1 year) Yes / No</td>
<td>36 / 14</td>
<td>100 / 100 (ns)</td>
<td>769 / 493 (ns)</td>
</tr>
<tr>
<td>Food</td>
<td>Cattle</td>
<td>No / Yes</td>
<td>124 / 18</td>
<td>69 / 61 (ns)</td>
<td>118 / 194 (ns)</td>
</tr>
<tr>
<td>supplementation</td>
<td>Buffalo</td>
<td>No / Yes</td>
<td>48 / 7</td>
<td>19 / 14 (ns)</td>
<td>20 / 7 (ns)</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>No / Yes</td>
<td>165 / 9</td>
<td>73 / 56 (ns)</td>
<td>471 / 561 (ns)</td>
</tr>
<tr>
<td>Free grazing*</td>
<td>Buffalo</td>
<td>Yes / No</td>
<td>48 / 7</td>
<td>19 / 14 (ns)</td>
<td>20 / 7 (ns)</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>Yes / No</td>
<td>161 / 11</td>
<td>72 / 64 (ns)</td>
<td>470 / 577 (ns)</td>
</tr>
<tr>
<td>Age</td>
<td>Cattle</td>
<td>≤ 3m/ 3m-3y/ &gt; 3y</td>
<td>17 / 79</td>
<td>59 / 79 / 61 (ns)</td>
<td>71 / 156 / 99 (&lt;0,01)</td>
</tr>
<tr>
<td></td>
<td>Buffalo</td>
<td>≤ 3m/ 3m-3y/ &gt; 3y</td>
<td>31 / 30</td>
<td>35 / 0 (&lt;0,01)</td>
<td>40 / 0 (&lt;0,01)</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>≤ 3m/ 3m-1y/ &gt; 1y</td>
<td>50 / 69</td>
<td>30 / 81 / 97 (&lt;0,01)</td>
<td>119 / 548 / 695 (&lt;0,01)</td>
</tr>
</tbody>
</table>

*All the cattle graze freely (1) Chi-square test/ Fisher exact test (2) Mann&Whitney test/ Kruskal Wallis test ; ns: non significant at 0.05

Table II: Bivariable analysis between some individual risk factors and GIS prevalence (%) or mean FEC (epg)

fertilizing, rice fields lowly contaminated, among others [2, 5]. On the other hand, our study was performed at the very beginning of the wet season and could explain the low level of infection as transmission may increase thereafter, especially in young cattle [2]. For technical reasons, no coproculture was performed in our study. Data regarding genera of GIS in Lao PDR are those of OTAKE SATO et al. [11] describing Haemonchus, Trichostrongylus and Oesophagostomum in goat and Haemonchus, Cooperia and Oesophagostomum in cattle. In Cambodia, DORNY et al. [2] showed also the occurrence of Mecistocirrus and Bunostomum in cattle, two other haematophagous nematodes like Haemonchus.

Our study confirmed the presence of Trichuris sp. and Strongyloides sp. in cattle and goats in Lao PDR [11]. In contrast, no egg of Moniezia sp. was seen whereas this parasite had already been described in Thailand in cattle and goat [6, 15, 18].

No Toxocara vitulorum egg was found in this study. Toxocara eggs excretion occurs during a very short period of time in cattle and buffalo i.e. when calves are between 3 and 10 weeks of age [1]. Our sampling was limited to 17 cattle and 2 buffalo calves less than 3 months because calving mainly occurs from November to February. Furthermore, farmers were often uncertain about the correct age of their calves. According to RAST et al. [12], 23% of cattle and buffalo calves younger than 3 months were found infected by T. vitulorum in northern Lao PDR. Higher egg excretions were recorded in buffalo compared to cattle calves in Lao PDR [12] and in Cambodia [1]. This level of infection in buffalo calves might result from their predilection for water and the survival of T. vitulorum eggs in water leading to a greater infection of buffalo dams and then of their offspring [17]. On the other hand, more than 80% of farmers in Lao PDR had none knowledge about T. vitulorum and only 2.5% of them effectively treated their calves with the recommended anthelmintic regime [14].

Lungworm larvae were found only in groups with a group prevalence of 20% and were of Protostrongylidae type. Although not yet described to our knowledge in Lao PDR, the occurrence of protostrongylid larvae in goat is not surprising considering their large distribution throughout over the world [9]. The distribution of the cattle lungworm (D. viviparus) includes temperate and subtropical areas in Europe and America as well as some highlands in Africa [21]. Regarding South East Asia, D. viviparus has been recorded in cattle in Malaysia [10].

Group prevalences for Fasciola sp. ranged from 6 to 14%. These results are not strictly comparable to individual prevalence data collected in cattle or buffalo through coproscopy in previous studies, i.e. 13-25% in Lao PDR [13], 22% in Vietnam [5] or 5-25% in Cambodia [2, 19]. When examining gross liver lesions at slaughterhouse on >5 years old animals, the prevalence of F. gigantica, reaching 71 %, was much higher than this estimated by coproscopy [13]. The true prevalence of F. gigantica, its impact on body condition in adult cattle [2] and on general illness status in
adult cattle/buffalo [13] highlight the importance of this cumulative infection with age in south-Asian environment. Group prevalences for rumen fluke were very high in all host species studied (80-100%). Few published data are available but indicated rather high levels of prevalences in cattle: from 28% in Thailand [7] to 95-100% in Cambodia even in cattle less than 1 year of age [2, 25]. Pathogenicity of rumen flukes is related to the migration of immature stages in the small intestine but some rumen fluke species, belonging to the Gastrothylacididae family, are also pathogenic at the adult stage because of their haematophagous and histopagous habits [4].

In conclusion, a large range of helminths was found in cattle, buffalo and goat in Khammouane Province of Lao PDR. A more accurate estimation of the genera/species involved and of the fluctuations of prevalence/intensity of infection is needed to evaluate their impact on the livestock productivity before proposing systematic drenching. It is actually crucial for rural poor smallholder farmers facing so many constraints (knowledge, investment, extension service among others) to prioritize the most relevant health or nutritional levers.

Acknowledgments

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References


