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ORIGINAL ARTICLE

Seasonal pattern of bovine amphistomosis in traditionally reared cattle in the Kafue and Zambezi catchment areas of Zambia

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Abstract Seasonality of bovine amphistomosis in the Southern province of Zambia was established after examining 268 faecal samples from cattle presented for slaughter at Turnpike slaughter slab, Mazabuka. Amphistomosis was found present throughout the year but the highest abundance rate was found during the post-rainy season (47.8%) and the lowest during the cold dry season (24.8%). In the rainy and post-rainy seasons, higher mean egg counts and cattle found positive were recorded than in any other season. The distribution of amphistome eggs was significantly different ($p < 0.001$) among the four seasons, with the rainy season having higher median egg counts than others. There were no significant differences in abundance rates between sexes or between ages of cattle. A similar seasonality to that of fasciolosis exists and may help in strategic management of *Fasciola* and amphistomes.

Keywords Amphistomosis · Cattle · Seasonality · Trematodes · Zambia

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Introduction

Amphistomosis is widely distributed throughout the world, but its highest frequency has been reported in tropical and subtropical regions (Brotowidjoyo and Coperman, 1979; Asanji, 1989; Rangel-Ruiz *et al*., 2003; Keyyu *et al*., 2005; Pfukenyi *et al*., 2005). Economic assessments have repeatedly demonstrated that losses from helminths can be enormous (McLeod, 1995; Perry and Randolph, 1999; Rangel-Ruiz *et al*., 2003). Amphistomosis poses a major obstacle to livestock production in ruminants worldwide, especially in young animals where it causes high morbidity and mortality. Outbreaks of clinical amphistomosis caused by immature flukes in calves often pass undiagnosed. Therefore, the importance of this disease may be considerably underestimated.

Fasciola and amphistome infections are highly prevalent in Western and Southern provinces of Zambia, especially in the Kafue wetlands and Zambezi flood plains where they occur mostly as double infections (Phiri *et al*., 2005a). Current infection figures reported in the country are among the highest in Africa and the infection plays an important role in the economy of the livestock owners.

Four seasons occur in Zambia: the rainy (December to February), the post-rainy (March to May), the cold dry (June to August) and the hot dry (September to November) seasons (Phiri *et al*., 2005b). Seasonal evolution of infection might be related to climatic variations, with rainfall being the most important (Rolfe *et al*., 1991). Microclimate can vary considerably from one region to another, from one farm to another or between neighbouring open grasslands (Rangel-Ruiz *et al*., 1999).

The aim of this investigation was to determine the seasonality of amphistomosis in cattle in the Southern province of Zambia using faecal samples stored in 10% formalin. The catchments of the Kafue and Zambezi rivers and their associated wetlands, which are major traditional cattle-rearing areas of Zambia, are found in this province. A comparison of amphistome seasonality was made with previously reported *Fasciola* seasonality.

Materials and methods

Study sites

The seasonality study was carried out from March 2002 to February 2003 at Turnpike slaughter slab in Mazabuka district of the Southern province. During this period, 268 faecal samples of cattle from Gwembe and Siavonga districts near the Zambezi river and Mazabuka, Monze and Namwala districts along the Kafue wetlands presented for slaughter were collected and stored in 10% formalin before coprological examination. At this slaughter slab, only cattle from traditional farmers in the Southern province of Zambia were slaughtered.

Although lying within the tropical zone, the average temperature in June and July (the coldest months) was 16°C and in January (the hottest month) was 21°C. Annual rainfall was about 750 mm and nearly all the rain fell between December and February. The onset of the rainy season coincided with the warmest period of the year when maximum monthly temperatures equalled or exceeded 30°C. Rainfall variability was greatest in the southernmost portions where mean rainfall was least (less than 72 days of rain) but above normal. Southern province is usually one of the driest provinces in the country.

Study design and animals

On average, 24 cattle were sampled monthly with a range of 9 to 45 (at the beginning and end of the month) on the first 20 cattle tendered for slaughter. The study animals were distributed according to sex (male 151; female 117) and age (young 116; adult 152). Age estimation was done by inspection of the incisor teeth according to a method of Yeates and Shmidt (1974) which is based on incisor and temporary teeth replacement and the degree of wear of permanent teeth. Cattle estimated to be less than 4 years old were classified as young cattle, and those 4 years and above as adults.

Coprological examination

A sieving and sedimentation technique with a glass bead layer for the detection and quantitation of amphistome and *Fasciola* eggs as described by Bonita and Taira (1996) was used to determine the egg count from faecal samples. The abundance rate was recorded using the ratio between the number of infected cattle and that of examined cattle and expressed as a percentage. The abundance grading system was done according to a proposal of Brotowidjoyo and Coperman (1979) in which less than 10% was considered low, 10–20% was considered moderate, and $>20\%$ was considered high.

Statistical analysis

The Pearson's chi-square, Fisher's exact test and Yates's corrected chi-square test using the SPSS version 11.0 for Windows (SPSS Institute, Chicago, IL, USA) were used to determine the associations of origin, age and sex of cattle with the seasonality of amphistomosis. Mean abundance of eggs was defined as the total number of amphistome egg counts divided by the total of animals examined including non-infected ones. Faecal egg count medians of males and females were compared by Mann–Whitney test. Kruskal–Wallis (H) test was used to compare distributions of faecal egg counts among the different seasons. A p-value of less than 0.05 was considered significant.

Results

The highest abundance rate of amphistomosis was found in January (66.7%) and the lowest rate was observed in September (15.0%) (Fig. 1). Mean monthly and therefore seasonal amphistome egg counts varied throughout the study period (Table 1). The months of January, March and April had recorded mean egg counts of above 10 eggs per gram (epg). The highest abundance rate was observed during the rainy season (47.8%) and the lowest during the cold dry season

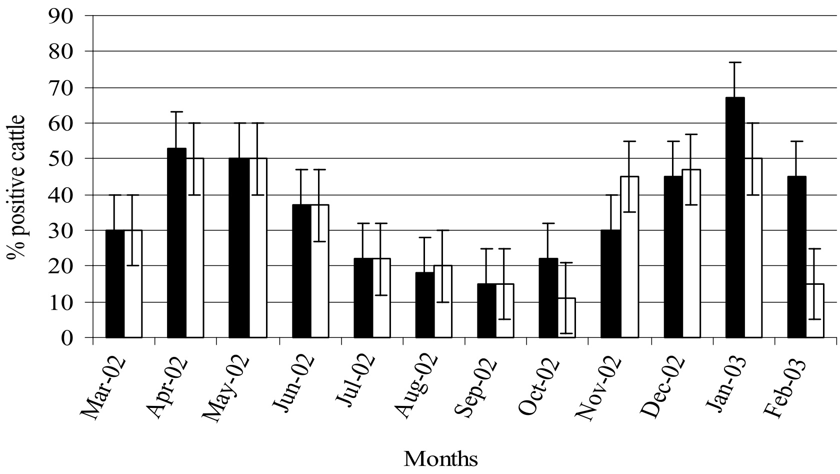

Fig. 1 Monthly pattern of amphistomosis (solid bars) and fasciolosis (open bars) (±SEM) in examined cattle presented for slaughter using coprological examination in 2002/2003

Table 1 Seasonal distribution of amphistome abundance, egg range and mean egg counts using coprological examination of examined cattle

Season	Number of cattle examined	Percentage of egg-positive cattle	Egg range	Mean egg count	SEM
Rainy	46	47.8	0–114	11.6	3.6
Post-rainy	80	45.0	0–110	6.6	1.9
Cold dry	113	24.8	0–67	3.3	0.9
Hot dry	29	34.5	0–79	5.6	3.1

(24.8%) (Table 1). Upon grading the level of abundance, all the seasons had high abundances. The seasonal pattern observed was that more eggs were found from November (end of dry season) to February/March (end of rains). In the rainy and post-rainy seasons, higher mean egg counts and cattle found positive were recorded as compared to any other season with the highest egg counts of 114 and 110 epg, respectively (Table 1). As the rains ended and drier conditions set in, egg counts became lower in some cattle in the cold dry season and the hot dry season, in which mean egg counts of less than 6 epg (ranges 0–67 and 0–79 epg, respectively) were found.

The distribution of eggs was significantly different ($p < 0.001$) among seasons. The rainy season (median 146) had higher egg counts compared with both the cold dry season (median 118) and the hot dry season (median 118). The abundance rates in the rainy (47.8%) and post-rainy (45.0%) seasons were not significantly higher ($p = 0.050$) than in the cold dry (24.8%) or hot dry (34.5%) seasons.

Female cattle had higher abundance rates in all seasons than male cattle (Table 2). In the rainy and post-rainy seasons higher abundance rates were recorded in both sexes. Male cattle had the lowest prevalence in the hot dry season, while in female cattle it was found in the cold dry season. However, the differences in the prevalences were not significant.

Table 2 Number of cattle examined and the seasonal pattern of amphistome infection according to sex of the examined cattle using liver inspection and coprological examination

	Male		Female	
Season	*n*	Number (%) of positive cattle	*n*	Number (%) of positive cattle
Rainy	27	11 (40.7)	19	11 (57.9)
Post-rainy	55	23 (41.8)	25	13 (52.0)
Cold dry	59	14 (23.7)	54	14 (25.9)
Hot dry	10	2 (20.0)	19	8 (42.0)

Adult cattle had higher prevalences in all seasons than the young cattle, but the differences were not statistically significant (Table 3). Higher abundance rates were found in the rainy and post-rainy seasons in both young and adult cattle. Lowest prevalences were noted in the cold season for adult (32.8%) and young (15.4%) cattle.

Table 3 Number of cattle examined and seasonal pattern of amphistome infection according to age of the examined cattle using liver inspection and coprological examination

	1–3 years (young)		4 years (adult)	
Season	*n*	Number (%) of positive cattle	*n*	Number (%) of positive cattle
Rainy	26	12 (46.2)	20	10 (50.0)
Post-rainy	23	11 (47.8)	57	31 (54.4)
Cold dry	52	8 (15.4)	61	20 (32.8)
Hot dry	15	5 (33.3)	14	5 (35.7)

Discussion

This study determined the seasonal pattern in the four seasons and also examined the associations of age, sex and origin of cattle using coprological examination. More cattle were found positive during the rainy and post-rainy seasons than in any other season. During these periods, two peaks were identified as being in April/May and November/December. However, amphistomosis was determined to occur throughout the year, agreeing with Pfukenyi and colleagues (2005) in Zimbabwe and Rangel-Ruiz and colleagues (2003) in Mexico. The high abundances during the rainy and post-rainy seasons may be attributed to the dispersion and multiplication of snails, dispersion of defecated material by rains, the likelihood of self-development of the miracidia, infection of snails and contamination with metacercariae in flooded areas (Rangel-Ruiz *et al.*, 2003). However, the cold dry season had the lowest abundance. Nonetheless, Lima and colleagues (2001) reported that the highest level of parasite contamination of pasture occurs during the dry season. A low abundance in the hot dry season could have arisen because of ongoing infection or harbouring of immature and/or light infections. The seasonal trend found is similar to that recorded for *Fasciola gigantica* fasciolosis (Phiri *et al.*, 2005b), implying that examined cattle become infected by both parasites as a result of grazing in the same pasture zones where their appropriate snail intermediate hosts are present (Phiri *et al.*, 2005a).

Since most of the time cattle of all ages accessed the same pasture zones, infection was picked up without regard to age and sex. Although adult cattle were found to have higher prevalences than young cattle, these differences were not statistically significant. However, Negesse (1994) and Pfukenyi and colleagues (2005) reported that adult animals had significantly higher prevalences than young cattle. In Zambia, adult cattle acquire such infections because they move long distances in search of scarce pastures and water, thereby increasing their chances of infecting as well becoming infected at overcrowded watering holes. The finding of positive cattle among the young agrees with Lima and colleagues (2001), who demonstrated that infection in calves can be picked up from as early as 2 months of age. Female cattle had higher prevalence rates than the male cattle despite both groups grazing together in the communal pastures. There was, however, no significant difference between male and female cattle in the fluke egg distribution in our study.

There is limited literature on the pathological effects of amphistomes in cattle. In our study we did not relate the numbers of worms or their eggs to pathological effects. Although some adverse effects on ruminant health have been attributed to infection by amphistome species, amphistomosis is still underestimated and generally there is limited information about the infection. The size of the amphistome burden is the most important factor determining the degree of small-intestinal pathology (Hansen and Perry, 1984). The immature stages of amphistomes can cause severe pathology and have been documented to cause death of the host in heavy infections (Silvestre *et al.*, 2000). Digestion and absorption are affected, and appetite is also depressed, resulting in diarrhoea, anorexia, anaemia and weakness (Spence *et al.*, 1996).

In Zimbabwe, Mavenyengwa and colleagues (2005) performed an experimental infection in cattle involving different doses of an amphistome, *Calicophoron microbothrium*. Groups of animals received a low dose of 5000 metacercariae, a medium dose of 15 000 metacercariae and a high dose of 25 000 metacercariae, and one additional animal was kept as an uninfected control. After infection, one animal from each group was slaughtered on days 28, 42, 56 and 84 post infection (pi). They observed significant pathological lesions in the duodenum and the abomasum. These changes, however, tended to decrease in severity as the infestation aged and had disappeared completely by day 56 pi. On the other hand, there is little evidence regarding the pathogenicity of adult amphistomes to their hosts, but severe damage to the mucosa of the rumen is provoked in heavy infections. Singh and colleagues (1984) demonstrated that tissue changes in the rumen appear only after 80 days post infection. Currently, we are undertaking a study to investigate concurrent effects of *Fasciola*, schistosome and amphistome worm and egg burdens in naturally infected cattle.

Since amphistome and *F. gigantica* infections have been found to occur throughout the year in Zambia, any strategic anthelmintic treatment scheme (e.g. at the beginning of the cold dry season and end/early rainy season) must be targeted at both parasites. There is also the need for other epidemiological studies involving other cattle production systems that will include levels of intensity for different geographical regions of the country to come up with a representative pattern. As estimated monthly variations of abundance were done on a limited number of cattle from communal grazing lands, a second study involving more cattle over a longer period and using worm burdens in the rumen is suggested to determine whether the variations were due to sampling of small numbers or to a real seasonal variation.

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Tendances saisonniéres de l'amphistomose bovine chez le bétail issu d'un élevage traditionnel dans les zones de captage de kafue et de zambezi de la zambie

Résumé – La saisonnalité de l'amphistomiase bovine dans la province du Sud de la Zambie a été établie après l'examen de 268 échantillons fécaux prélevés de bétail présenté à l'abattage à l'abattoir de Turnpike, à Mazabuka. L'amphistomiase s'est avérée être présente tout le long de l'année mais le taux de fréquence le plus élevé a été relevé durant la saison faisant suite aux pluies (47.8%) et le plus bas durant la saison froide et sèche (24.8%). Durant la saison des pluies et la saison faisant suite aux pluies, il a été enregistré des comptages d'œufs moyens et de bétail détecté positif plus élevés par comparaison à toute autre saison. La distribution des oeufs d'amphistomes a été significativement différente ($p < 0.001$) durant les quatre saisons avec la saison des pluies enregistrant des comptages médians d'œufs plus élevés que durant les autres saisons. Aucune différence significative n'a été enregistrée dans les taux d'abondance entre les sexes et également l'âge du bétail. Une saisonnalité similaire à celle de la fasciolase existe et pourrait faciliter la prise en charge stratégique de Fasciola et des amphistomes.

Patrón estacional de anfistomiasis bovina en el ganado criado tradicionalmente de los distritos de kafue y zambezi en zambia

Resumen – Se estableció la estacionalidad de la anfistomiasis bovina en la provincia sur de Zambia después de examinar 268 muestras fecales de ganado presentado para la matanza en la mesa mortuoria del matadero de Turnpike, en Mazabuka. Se encontró anfistomiasis durante todo el año, pero la mayor tasa de abundancia se encontró durante la estación post lluviosa (47.8%) y la tasa menor durante la estación fría y seca (24.8%). En las estaciones lluviosas y post-lluviosas, se registraron los más altos recuentos promedios de huevos y el mayor número de reses de ganado positivas, en comparación con cualquier otra estación. La distribución de huevos de *amphistomas* fue significativamente diferente ($p < 0.001$) entre las cuatro estaciones, teniendo la estación lluviosa los recuentos promedios de huevos más altos que las otras. No hubo diferencias significativas en las tasas de abundancia entre los sexos o con respecto a la edad del ganado. Existe una estacionalidad similar a la característica de la fasciolosis y esto podría ayudar en el manejo estratégico de la Fasciola y las *Amphistomas*.