

Prevalence of *Taenia solium* porcine cysticercosis in the Eastern, Southern and Western provinces of Zambia

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Abstract

Tongue examination and detection of circulating antigen (Ag-ELISA) were used to establish the prevalence of *Taenia solium* porcine cysticercosis in free-range pigs in selected districts of Eastern, Southern and Western provinces of Zambia, and to determine if prevalence of porcine cysticercosis was associated with age, breed and sex. Households with pigs were identified using the snowballing technique. A total of 1691 pigs were examined out of which 183 (10.8%) were positive on tongue examination. Ag-ELISA gave a sero-prevalence of 23.3%. When considering the factors in a logistic regression analysis, only breed type was significantly associated with porcine cysticercosis (OR = 0.72; 95%CI = 0.63–0.81). The crossbred pigs were 72% more likely to have had cysticercosis than the Nsenga (dwarf local) breed as determined by Ag-ELISA. The result that crossbred pigs had a higher prevalence of *T. solium* cysticercosis suggests that pig breeds may display different susceptibility to cysticercosis. The limited use of latrines in these areas implies that people use the nearby bush for defecation, resulting in pigs having access to human faeces. Therefore, investigation of taeniosis and cysticercosis in humans is warranted to better comprehend the local epidemiology and transmission risks. This should then be followed by extension programs to communities so that the control plans that could be instituted are more sustainable.

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Introduction

Taenia solium cysticercosis is a serious public health problem in endemic regions (Garcia-Garcia et al., 1999; Geerts et al., 2002). Humans are the only definitive hosts while pigs act as intermediate hosts. Pigs are the source of human taeniasis, an intestinal tapeworm infection acquired by eating undercooked pork contaminated with *Cysticercus cellulosae* (cysticerci) the larval stage of the ces-

tode. The pig is infected by ingestion of parasite eggs or proglottids in human faeces and acquires cysticercosis. Thus, porcine infection with *T. solium* is limited to areas where animal husbandry practices are such that pigs come into contact with human faeces (Toledo et al., 2001). Cysticercosis may also occur in humans if eggs are conveyed to the mouth by unclean fingers after defaecation (Flisser, 1994), but the swallowing of *T. solium* egg-contaminated soil, water or vegetation are the most likely causes of the human condition (Schantz et al., 1992).

Garcia et al. (1999) suggested that determining the percentage of the porcine population that is infected is a better

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epidemiological indicator of *T. solium* infection pressure than measuring the infection in the human population. These authors listed the following reasons for their proposal: firstly, porcine infection is much more common than the human infection; secondly, the life span of pigs in field conditions is much shorter than that of humans; and thirdly, the bleeding of pigs for diagnostic purposes is more easily accepted by pig owners compared to obtaining blood and/or stool specimens from humans.

Surveys in pigs have contributed to an increased awareness of this zoonotic infection in many developing countries, including Eastern and Southern Africa (Phiri et al., 2003). A pilot field study conducted by Phiri et al. (2002) suggested that porcine cysticercosis was prevalent in the rural areas of the Southern and Eastern provinces of Zambia and they recommended that an extensive study covering more districts and larger areas be conducted in order to explore the extent of *T. solium* infection in these areas.

The present survey, therefore, was conducted with the aim of establishing the prevalence of *T. solium* porcine cysticercosis in rural free-range pigs in selected districts of Eastern, Southern and Western provinces of Zambia and to establish if the prevalence of porcine cysticercosis was associated with age, breed and sex.

Material and methods

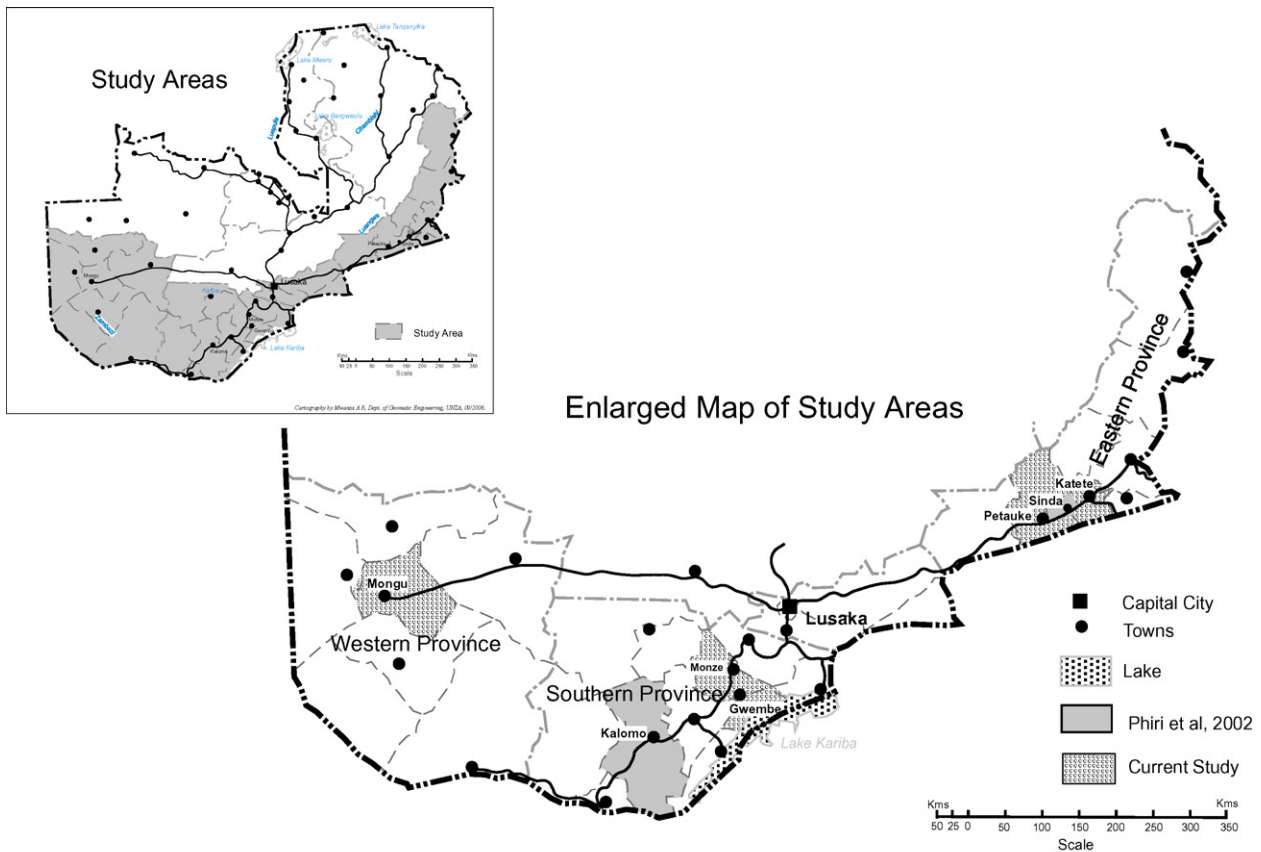
Study areas

The study was done in five districts; Katete and Petauke in the Eastern province, Gwembe and Monze in the Southern province, and Mongu in the Western province (Fig. 1). With the exception of Gwembe, Monze and Mongu, these districts were selected following a preliminary survey that was conducted by Phiri et al. (2002), which indicated the presence of free-ranging pigs.

Study design and animals

A cross sectional study was conducted between June 2002 and September 2003. Sample size estimation was calculated using the formula $n = Z^2PQ/L^2$, by Martin et al. (1987), where n is the required number of individuals to be examined, Z is the Z score for a given confidence level, P is a known or estimated prevalence, $Q = (1 - P)$, and L the allowable error of estimation. In the current study, we used 95% as the confidence level with an allowable error of estimation of 0.05. To get the maximum sample size, P was estimated at 50%. Thus $n = 1.96^2 \times 0.5 \times 0.5 / 0.05^2 = 384$. Therefore, at least 384 pigs were to be sampled from each district, with the exception of Mongu in the Western province, where the pig population was small. In the Eastern province, the local (dwarf) pig breed called Nsenga predominates. In the Southern and Western provinces, the common type of pig breed is a cross between Large White and Landrace.

In the villages, the principle investigator (CSS) explained the purpose of the study to the village headmen and requested permission to conduct the study. The animals that were examined belonged to farmers who were



Cartography by Mwanza A R, Dept. of Geomatic Engineering, UNZA, 09/2006.

Fig. 1. Map showing the study areas (districts) in Eastern, Southern and Western provinces of Zambia.

willing to participate in the study. Households with pigs were identified using the snowballing technique (Heckathorn, 2002; Salganik and Heckathorn, 2004).

A pig was classified as young if it was less than 1 year and as an adult if it was one year or older (Pouedet et al., 2002). All the pigs in a household were examined if the herd size was <5, and 50% were examined if it was >5. The breed, sex and age of the pigs were recorded at the time of sampling.

Tongue examination

The pig was firmly restrained in lateral recumbency and a hard wooden stick was used to open the mouth and keep it open. Using mutton cloth as an aid, the tongue was pulled out, examined and palpated along its entire ventral aspect to check for presence or absence of cysticerci. A pig found to have one or more cyst(s) on the tongue was considered positive for *T. solium* cysticercosis.

Blood collection for antigen-ELISA (Ag-ELISA)

Blood samples were obtained from the cranial vena cava into plain blood collecting tubes and the blood was allowed to clot. To obtain serum, the clotted blood was separated by centrifugation at 3200 g for 5 min. The supernatant (serum) was dispensed into 2 mL aliquots and stored in labelled cryogenic vials at -20°C until use.

Enzyme-linked-immunosorbent assay for the detection of circulating antigens of *T. solium* cysticerci (Ag-ELISA)

Antigen-ELISA was preferred as a second measure of infection in pigs in addition to tongue examination because many studies have reported shortfalls with antibody detection (Ab-ELISA) in animals (Pinto et al., 2000; Garcia et al., 2001; Dorny et al., 2003). Ag-ELISA has been shown to have a high sensitivity for detecting a pig with even a single cyst (Nguekam et al., 2003), and has the advantage of differentiating between recent infections with live metacestodes and older infections with degenerated metacestodes, which are no longer infective (Harrison et al., 1989). The Ag-ELISA was performed according to Pouedet et al. (2002) and slightly modified as described by Sikasunge et al. (2007).

Statistical analysis

SPSS Version 11.0 and Microsoft Excel software were used for analysis. Cross tabulations were done in SPSS, while 95% Confidence Intervals (CI) for prevalence rates were computed in Excel using the formula:

$P \pm 1.96\sqrt{(PQ/n)}$ (Daniel, 1991), where P is the prevalence, $Q = 100 - P$ and $n =$ sample size. Fisher's exact test or Yates corrected χ^2 were used to assess the association between porcine *T. solium* infection and the factors (sex, age and breed) at a 95% confidence level. The step forward regression analysis was used to adjust for confounding factors. The Ag-ELISA was entered as an outcome and the covariates that were included in the model were sex, age and breed.

Results

Study population

A total of 1691 pigs were examined comprising 769, 772 and 150 from the Eastern, Southern and Western provinces, respectively. Pigs were sampled from a total of 856 households.

Prevalence of *T. solium* porcine cysticercosis by district

Of the 1691 pigs examined, 182 (10.8%) were found to be positive on tongue examination. Ag-ELISA detected an overall sero-prevalence of 23.3%. Pigs in the Southern province had a higher prevalence of cysticercosis 15.2% ($P < 0.001$) on tongue examination than pigs in the Eastern and Western provinces, which had 7.0% and 7.3% positives, respectively (Table 1).

Prevalence of *T. solium* porcine cysticercosis according to sex, age and breed

On bivariate analysis, sex and breed were significantly associated with the prevalence of porcine cysticercosis on both tongue examination and Ag-ELISA (Table 2). Age was not significantly associated with prevalence of porcine cysticercosis in both tests. When considering the factors in a logistic regression, only breed type was significantly associated with porcine cysticercosis (OR = 0.72; 95%CI = 0.63–0.81). The crossbred pigs were 72% more likely to have had cysticercosis than the Nsenga breed as determined by Ag-ELISA.

Table 1
Prevalence of *T. solium* cysticercosis in pigs after tongue examination and Ag-ELISA by district in Eastern, Southern and Western provinces of Zambia

Province	District	<i>n</i>	Tongue examination +ve (%; 95%CI)	Ag-ELISA +ve (%; 95%CI)
Southern	Gwembe	385	83 (21.6, 17.5–25.7)	131 (34.0, 29.3–38.8)
	Monze	387	34 (8.8, 6.0–11.6)	88 (22.7, 18.6–26.9)
	Sub-total	772	117 (15.2, 12.6–17.7)	219 (28.3, 25.2–31.5) ^a
Eastern	Petauke	384	25 (6.5, 4.0–9.0)	56 (14.6, 11.1–18.1)
	Katete	385	29 (7.5, 4.9–10.2)	74 (19.2, 15.3–23.3)
	Sub-total	769	54 (7.0, 5.2–8.8)	130 (16.9, 14.3–19.6) ^b
Western	Mongu	150	11 (7.3, 3.2–11.5)	45 (30.0, 22.7–37.3)
	Sub-total	150	11 (7.3, 3.2–11.5)	45 (30.0, 22.7–37.3) ^a
Total		1691	182 (10.8, 9.3–12.2)	394 (23.3, 21.3–25.3)

^a Prevalence rates for porcine cysticercosis between Southern and Western provinces were not significantly different as determined by Ag-ELISA.

^b Prevalence rates for porcine cysticercosis in Eastern province was significantly lower ($P < 0.05$) than in Southern and Western provinces as determined by Ag-ELISA.

Table 2

Prevalence of *T. solium* porcine cysticercosis with reference to sex ($n = 1691$), age ($n = 1391$) and breed ($n = 1691$) on tongue examination and Ag-ELISA

		<i>n</i>	Tongue examination +ve (%; 95%CI)	Ag-ELISA +ve (%; 95%CI)
Sex	Male	472	66 (14.0, 10.9–17.1)	126 (26.7, 22.7–30.7)
	Female	1219	116 (9.5, 7.9–11.2)	268 (22.0, 19.7–24.3)
Age	Young	626	68 (10.9, 8.4–13.3)	163 (26.0, 22.6–29.5)
	Adult	765	62 (8.1, 6.2–10.0)	174 (22.7, 19.8–25.7)
Breed ^a	Nsenga	769	54 (7.0, 5.2–8.8)	130 (16.9, 14.3–19.6)
	Crossbreed	922	128 (13.9, 11.7–16.1)	264 (28.6, 25.7–31.6)

^a Statistically significant on logistic regression analysis (OR = 0.72; 95%CI = 0.63–0.81). Crossbred pigs were 72% more likely to have had cysticercosis than the Nsenga breed as determined by Ag-ELISA.

Discussion

We have conducted an extensive study in order to explore the endemicity of *T. solium* cysticercosis in pigs in Zambia, as recommended by Phiri et al. (2002) using two of the available diagnostic methods used in detection of *T. solium* cysticercosis infection in pigs, namely tongue examination and antigen-ELISA. The high prevalence of porcine cysticercosis observed in the Southern province on both tongue examination and Ag-ELISA, confirmed the findings of a preliminary field survey conducted by Phiri et al. (2002). These authors reported cysticercosis in 8.2% of pigs in Kalomo district (Southern province) and 5.2% pigs in Sinda area (Eastern province) by tongue examination. In the same survey, using the Ag-ELISA, 20.8% and 9.3% of the pigs were infected in the Southern and Eastern provinces, respectively.

The prevalence rates obtained in the present study were higher than those previously reported in Zambia. This difference was probably due to the wider survey coverage and the larger sample size. In the current study, we observed 7.0% positive pigs on tongue examination in the Eastern, 15.2% in the Southern and 7.3% in the Western provinces which are higher than those reported by Phiri et al. (2002). We have also shown that *T. solium* infection is also endemic in the Western province of the country, a region that had previously not been investigated.

In our study, the prevalence of porcine cysticercosis on tongue examination was lower than that obtained with Ag-ELISA in all provinces. This was a normal finding because tongue examination only offers an estimate of infection levels, and not all active infections can therefore be easily detected by this method (Sarti-G et al., 1992; Phiri et al., 2006). Sarti-G et al. (1992) reported that not all positive pigs necessarily have cysts on the tongue and that probably infection intensity could be the most important factor determining whether cysts are discernible by visual inspection of the tongue or not. Recently, a study conducted by Phiri et al. (2006), reported that tongue examination could only detect 61.3% of *T. solium* infected pigs

although it exhibited a high specificity of 100%. The finding that in the Western province, the Ag-ELISA result was very high but detected cyst prevalence (tongue examination) was low (high–low), contrasted with both Southern (high–high) and Eastern (low–low) provinces, and is difficult to explain.

Phiri et al. (2002), from the survey of pigs slaughtered at the Chibolya slab in Lusaka, found that 10% were positive by examination of the tongue. All of these animals were from the Southern province. This difference in prevalence clearly demonstrates that pig data obtained from abattoirs do not reflect the true disease picture in rural communities. Our results agree with those of Sarti-G et al. (1992) and Carrique-Mas et al. (2001) who reported that abattoir surveys appear to under estimate the real prevalence of the disease.

Bivariate analysis revealed that male pigs had a significantly higher prevalence than females on both tongue and Ag-ELISA. However, the association between sex and the prevalence of porcine cysticercosis that was observed on a bivariate analysis disappeared when other variables were taken into account in a step forward logistic regression analysis. Breed type was the only factor that was independently associated with porcine cysticercosis. This finding could suggest that the Nsenga breed that is predominant in the Eastern province may be more resistant to cysticercosis than the crossbreeds found in the Southern and Western provinces. The fact that crossbred pigs had a higher prevalence of *T. solium* cysticercosis further potentiates the hypothesis that in Zambia, pig breeds may display different susceptibility to cysticercosis (Phiri et al., 2006).

The limited use of latrines in these provinces implies that people use the nearby bush for defaecation, resulting in pigs having ready access to human faeces (Sikasunge et al., 2007). In the previous study conducted by Sikasunge et al. (2007), a free-range husbandry system was found to be a significant risk factor for infection in pigs. That study also revealed that people in the villages had limited use of latrines, slaughtered pigs were not inspected and many individuals consumed and sold infected pork.

Our study confirmed that there was a high prevalence of porcine cysticercosis in the areas that were surveyed, which, in turn, indicated that there were *T. solium* human carriers in the study areas – since pigs can only get the infection from humans. Moreover, infected pigs have been used as markers/sentinels of active transmission zones otherwise called “hot spots” (Dorny et al., 2003). This entails that a study on the taeniosis and cysticercosis complex in humans in Zambia is warranted to better understand the local epidemiology and transmission risks. The assessment of the burden of the disease in humans should then be followed by extension programs, community education (emphasis on maintaining hygienic and sanitary conditions) and other prevention and control initiatives so that the control measures that could be instituted are more sustainable.

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