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Risk factors associated with porcine cysticercosis in selected districts of Eastern and Southern provinces of Zambia

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Abstract

To determine the risk factors associated with *Taenia solium* transmission in humans and pigs in the rural areas of Eastern and Southern provinces of Zambia, a questionnaire was administered in 788 households from 155 villages. Pigs were examined from 800 households. Tongue examination and enzyme-linked immunosorbent assay (Ag-ELISA) for the detection of circulating antigens of *T. solium* cysticerci were used to measure infection in pigs. A snowballing technique was utilised to select households with pigs. Prevalence of households with pigs infected with *T. solium* on tongue examination by district ranged from 12.7% to 32.1% with Ag-ELISA having a range of 30.0–51.7%. Of the total number of households visited, 18.8% and 37.6% had at least one pig positive for porcine cysticercosis on tongue examination and Ag-ELISA, respectively. Risk factors associated with *T. solium* infection were lack of pork inspection at slaughter (96.7%), consumption of pork with cysts (20.1%), selling of pork infected with *T. solium* cysticerci (18.3%), free-range husbandry system (83.2%) and absence of latrines (58.0). Free-range husbandry system (OR = 1.68; 95% CI = 1.36–2.07) was a significant risk factor for porcine cysticercosis in the surveyed areas. The result that pigs were mostly kept on free-range and semi-intensive husbandry systems may have permitted them to have access to eating human faeces that could be contaminated with tapeworm eggs. This study has shown that *T. solium* infection poses a high public health risk in the study areas and urban areas as well. We recommend that a human survey be conducted to verify the human exposure to taeniasis and/or cysticercosis in Zambia.

Keywords: *Taenia solium*; Risk factors; Snowballing technique; Questionnaire; Pig; Zambia

1. Introduction

Occurrence and prevalence of *Taenia solium*, the pig tapeworm, is associated with certain cultural practices such as eating raw or undercooked pork and poor socio-economic conditions. Studies have demonstrated that in endemic areas, *T. solium* porcine infections have been associated with poverty, absence of latrines and free access by scavenging pigs to human faeces (Diaz et al., 1992; Schantz et al., 1992; Sarti et al., 1997). In the usual cycle of transmission, humans acquire intestinal infection (taeniasis, or tapeworm infection) by ingestion of undercooked pork infected with *T. solium* cysticerci. Pigs become infected when they ingest human faeces containing either the ova or proglottids of

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the tapeworm and develop cysticercosis, the larval form of *T. solium* otherwise called *Cysticercus cellulosae* (Schantz et al., 1998).

Human cysticercosis results when man ingests ova shed by a human tapeworm carrier, close personal contact with or food preparation by a tapeworm carrier (Schantz et al., 1998; White, 2000). Therefore, the disease may also occur in humans who neither eat pork nor share environments with pigs (Schantz et al., 1992, 1998). Infection with *T. solium* is of great public health and economic relevance because of its potential to cause neurocysticercosis (NCC), the infection of the central nervous system by the larval stage of the tapeworm (White, 1997). Most morbidity and mortality in human cysticercosis occur when the parasite invades the central nervous system and causes epilepsy, hydrocephalus and other neurological manifestations (Garcia-Garcia et al., 1999).

In Zambia, pig rearing has increased recently because farmers have made a shift from keeping cattle alone to keeping both cattle and other livestock including pigs. This is because of the increase in the deaths of cattle from tick-borne infections such as theileriosis and/or East Coast Fever (ECF) particularly in the Eastern and Southern provinces (MACO, 2003). Besides, farmers have also realised that they would get quicker and more returns on their investment by raising pigs (Phiri et al., 2002, 2003; Boa et al., 2003).

In the pilot study conducted by Phiri et al. (2002), the sample size was small and the analysis of risk factors was inadequate. In the current study, logistic regression was used to analyse the risk factors instead of a descriptive analysis only. Therefore, the current study was conducted to determine the risk factors associated with *T. solium* transmission in humans and pigs using a questionnaire and to establish any association between the risk factors and the prevalence of households with at least one pig positive for cysticercosis. Prevalence of porcine cysticercosis was determined by tongue examination and Ag-ELISA.

2. Materials and methods

2.1. Study areas

The survey was conducted in the rural areas of the Eastern and Southern provinces of Zambia. In the Eastern province, the study was conducted in Petauke and Katete districts. The two districts were selected following a preliminary survey that was conducted by Phiri et al. (2002), which indicated presence of free ranging pigs. In the Southern province, the study was done in Gwembe and Monze districts. These districts were selected following a preliminary visit that was made which indicated the presence of pigs in the area.

2.2. Study design and population

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 is 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.

Households with pigs were identified using the snowballing technique and sampling to redundancy. Snowballing is a technique for developing a research sample where existing study subjects recruit future subjects from among their acquaintances. Once the first household with pigs was identified, sampling of pigs from that household was done before going to the next. We kept on asking until every farmer with pigs in that particular village was covered. Sampling of pigs in the households was done using simple random method. In the villages, the principle investigator explained the purpose of the study to the village headmen and requested permission to conduct the study.

Sows with advanced pregnancy, those that had recently farrowed and piglets less than 6 months old were excluded from the study. This was done so as to avoid stress in sows and the possibility of having false positives due to passive immunity in pigs less than 6 months old (Gonzalez et al., 1999). If the number of pigs in a household was less than five, all the pigs that met our selection criteria from the selected households were examined. Fifty percent of pigs per household were examined if the herd size was greater than five. Households found to have one pig positive for cysticercosis on tongue examination or Ag-ELISA were considered positive.

2.3. Tongue examination

The pig was firmly restrained in lateral recumbency and a hard wooden stick was used to open and maintain the mouth open. Using a mutton cloth for grip, the tongue was pulled out, examined and palpated all along its ventral side for the presence of cysticerci.

2.4. Blood collection for antigen ELISA (Ag-ELISA)

The blood samples were obtained from the cranial vena cava into plain blood collecting tubes and allowed to clot. To obtain serum, the clotted blood was separated by centrifugation. The supernatant (serum) was dispensed into 2 ml aliquots and stored in labelled vials and kept at −20 °C until use.

2.5. Antigen-ELISA for the detection of circulating antigens of T. solium cysticerci

Antigen-ELISA was preferred as a second measure of infection in pigs besides tongue examination because many studies have reported shortfalls with antibody detection (Ab-ELISA) in animals (Harrison et al., 1989; Pinto et al., 2000; Garcia et al., 2001; Dorny et al., 2003). Ag-ELISA has been shown to have high sensitivity of even detecting a pig with one 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 as described by Pouedet et al. (2002) with slight modifications. Briefly, two monoclonal antibodies (MoAb) used in the ELISA were B158C11A10 diluted at 9.5 μg/ml in carbonate buffer (0.06 M, pH 9.6) for coating and a biotinylated MoAb B60H8A4 diluted at 3.2 μg/ml in phosphate buffered saline-Tween 20 (PBS-T20) + 1% new born calf serum (NBCS) as detector antibody. Streptavidin-horseradish peroxidase (Jackson Immunoresearch Lab, Inc.) diluted at 1/10,000 in PBS-T20/1% NBCS was added to act as conjugate. Sera from two known positive pigs (from our previous dissection work – Phiri et al., 2006) were used as positive control. To determine the cut-off, the optical density (OD) of each serum sample was compared with a series of 8 reference negative serum samples at a probability level of 0.1% (Dorny et al., 2000).

2.6. Household questionnaire

A questionnaire was developed and used to collect information on risk factors and other related information from pig farmers. It was administered by veterinary assistants in charge of the respective areas using the native language. Households in each village were selected using the snowballing technique (see Section 2.2) from those farmers willing to participate in the study. Each person from the selected household was approached individually to obtain his or her informed consent before questionnaire administration. The respondent was any adult person living in the household who could understand and answer the questions. The questionnaire was written in English but was translated into a local language, Nsenga or Chewa in the Eastern province and Tonga in the Southern province, and back translated to English. Only after pigs were examined (tongue examination) and sampled (blood collected from them) was the questionnaire administered to any adult member of that particular household. Thus pig sampling and questionnaire administration were done concurrently. Data collected included the number of pigs owned, general pig management system and the main aim of keeping pigs. Also collected was the information about their knowledge on taeniasis and cysticercosis, pork consumption as well as presence and usage of latrines. Specific information regarding medical history related to the presence of taeniasis and/or symptoms suggestive of neural disorders in the family and the entire community in the village was collected.

2.7. Statistical analysis

Data on pig seroprevalence and risk factors were entered and analysed in separate databases using SPSS Version 11 (IL, USA). A multivariate logistic regression analysis was then performed, calculating odds ratios and 95% confidence intervals for risk factors for porcine seropositivity to cysticercosis.

3. Results

3.1. Study population

Pigs were sampled from 800 households. However, we were able to administer questionnaires to only 788 households. Reasons for not interviewing all the farmers were that other farmers refused to be interviewed despite giving us a go ahead to examine their pigs. Others gave the consent to examine their pigs but were not available for interviews because they had to attend to family commitments. In circumstances where we could not interview the farmer, an attempt was made to assess if the latrine was present or not and if present, whether it was used or not.

A total of 156 male respondents and 231 female respondents were interviewed from the Eastern province, whereas 175 male and 227 female respondents were interviewed in the Southern province. In the Eastern province 51 villages were visited, 26 villages in Petauke and 25 villages in Katete districts were visited.

A total of 769 pigs from 388 households were examined; 384 pigs were examined from 168 households in Petauke and 385 pigs were examined from 220 households in Katete. In the Southern province, a total of 104 villages comprising 60 villages in Gwembe and 44 villages in Monze were visited. A total sample of 772 pigs from 412 households was examined. In Gwembe, 385 pigs were examined from 209 households while in Monze 387 pigs were examined from 203 households.

3.2. Prevalence of households with at least one pig with cysticercosis

Prevalence of households with pigs infected with *T. solium* on tongue examination by district ranged from 12.7% to 32.1% (Table 1). On Ag-ELISA, the prevalence by district ranged from 30.0% to 51.7%. Households in Gwembe district had the highest prevalence rates on both tongue examination (32.1%) and Ag-ELISA (51.7%) whereas households in Katete district had the lowest prevalence rate on tongue examination (12.7%) and Ag-ELISA (30.0%). Of the total number of households visited, 18.8% and 37.6% of them had at least one pig positive for porcine cysticercosis on tongue examination and Ag-ELISA, respectively.

3.3. General pig management system

From a total of 788 farmers interviewed, 83.2% kept pigs on free-range and 16.8% practiced semi-intensive management system. About 99.5% ($n = 388$) of the farmers in the Eastern province reared pigs on free-range as compared to 67.7% ($n = 412$) of the farmers in the Southern province. In both provinces, pigs were normally kept in small shelters or kraals during the rainy season and were left to scavenge during the dry season. The pigs were often supplementary fed with agricultural products such as pumpkins, cucumbers, watermelons and by-products consisting mostly of maize bran.

Table 1
Prevalence of households with pigs infected with *T. solium* cysticercosis in Eastern and Southern provinces based on tongue examination and Ag-ELISA

Province	District	*n*	+ve (%)	
			Tongue	Ag-ELISA
Eastern	Petauke	168	22 (13.1)	51 (30.4)
	Katete	220	28 (12.7)	66 (30.0)
Total		388	50 (12.9)	117 (30.2)
Southern	Gwembe	209	67 (32.1)	108 (51.7)
	Monze	203	33 (16.3)	76 (37.4)
Total		412	100 (24.3)	184 (44.7)
Total		800	150 (18.8)	301 (37.6)

Key: n = number of households.

3.4. Purpose for keeping pigs

Most of the pig farmers (98.4%) in the Eastern province kept pigs for both sale and home consumption whereas 86.1% of the farmers in the Southern province kept pigs with the sole purpose of selling them. Less than 1% of the farmers kept pigs only for home consumption in all the districts in both Eastern and Southern provinces.

3.5. Constraints to pig rearing

Farmers reported that diseases and feeding (49.1%) and diseases alone (41.3%) were the most common problems affecting pig rearing in the Eastern province. Feeding (49.5%) was the main problem cited by the pig farmers in the Southern province. Besides disease and feeding, farmers considered lack of market (2.5%) and theft (1.4%) as other problems associated with pig rearing.

3.6. Pig slaughter, pork inspection and consumption

Most of the farmers in both provinces had slaughtered at least one pig at home (82.8%), with 96.7% doing so without inspection. The majority of the farmers (92.3%) consumed pork (Table 2).

3.7. Assessment of sanitary facilities

Generally, 58.0% of all the households visited did not have latrines. In the Eastern province, 52.7% ($n = 388$) of the households did not have latrines. Katete had more (10.9%) households not using latrines even when they had them as compared with 5.4% in Petauke. Of the 412 households visited in the Southern province, 63.1% did not have latrines, either. Non-use of latrines in Gwembe and Monze districts in the Southern province was similar (11.8% and 11.0%, respectively).

Seroprevalence based on porcine seropositivity on Ag-ELISA for households without latrines was 38.9%, those that had latrines but did not use them was 35.4% and those that used latrines was 37.0%.

Table 2
The number and percentage of households in Gwembe, Monze, Petauke and Katete districts with respect to pork consumption, home slaughter and lack of pork inspection

Province	District	*n*	Number (%)		
			Pork consumption	Home slaughter	Lack of pork inspection
Eastern	Petauke	167	164 (98.2)	156 (93.4)	159 (95.2)
	Katete	220	215 (97.7)	217 (98.6)	217 (98.6)
Sub-total		387	379 (97.9)	373 (96.4)	376 (97.2)
Southern	Gwembe	202	178 (88.1)	128 (63.4)	193 (95.5)
	Monze	200	171 (85.5)	152 (76.0)	194 (97.0)
Sub-total		402	349 (86.8)	280 (69.7)	387 (96.3)
Total		789	728 (92.3)	653 (82.8)	763 (96.7)

Key: *n* = number of households.

3.8. Knowledge of taeniasis and cysticercosis

Altogether, 85.5% of the farmers in the Eastern province and 98.0% in the Southern province did not know that there exists a link between human taeniasis and porcine cysticercosis. Of the farmers interviewed, 87.3% in the Eastern province and 98.5% in the Southern province did not know that tapeworm infection in humans was due to eating infected pork. Ninety-seven percent of the farmers did not know that pig cysticercosis was due to pigs eating human faeces contaminated with *T. solium* eggs.

Observation of cysts by farmers was high in all districts, but did not deter them from eating or selling pork. Most of the respondents (83.3%) had observed cysts in pork. Of the respondents that observed cysts in pork, 20.1% ate and 18.3% sold the meat. Of the 659 farmers who observed cysticercosis, 43.8%

Table 3
Factors that were considered in the logistic regression analysis

Factor	Level	Odds ratio (95% CI)	*P*-value
Breed type	Local	0.86 (0.73–1.02)	0.082
	Cross	Ref	
Husbandry system	Free range	1.68 (1.36–2.07)	<0.001*
	Semi-intensive	Ref	
Pork consumption by any family member	Yes	1.07 (0.77–1.48)	0.705
	No	Ref	
Home slaughter	Yes	1.02 (0.08–1.30)	0.889
	No	Ref	
Meat inspection	Yes	1.36 (0.91–2.04)	0.137
	No	Ref	
Presence of latrine	Present	1.03 (0.89–1.20)	0.708
	Absent	Ref	
Knowledge of taeniasis	Yes	1.25 (0.91–1.72)	0.173
	No	Ref	
Knowledge on how man gets infected	Correct	1.00 (0.73–1.43)	0.993
	Wrong	Ref	
Observation of cysts in pork	Yes	0.84 (0.68–1.04)	0.112
	No	Ref	
Knowledge of how pigs get infected	Correct	0.64 (0.35–1.18)	0.156
	Wrong	Ref	
Eating of infected pork	Yes	1.04 (0.80–1.37)	0.762
	No	Ref	
Selling of infected pork	Yes	0.91 (0.69–1.20)	0.513
	No	Ref	

Key: *statistically significant at 95% CI. Households that practiced free-range husbandry system were 68% more likely to have had at least one pig positive for cysticercosis compared to households that practiced semi-intensive husbandry system (OR = 1.68; 95% CI = 1.36-2.07).

acknowledged pork measles as just being a pig disease, 6.5% referred to it as being husks or maize bran (because they think that porcine cysticercosis results from feeding pigs with maize bran or husks) whereas, 48.3% had no idea of what cysts seen in pork were.

3.9. Symptoms related to neurocysticercosis

Out of the 788 respondents, 53.2% acknowledged knowing someone with epilepsy, 38.8% with mental illness (madness) and 55.1% had heard of someone with chronic headache. Those who acknowledged hearing of someone suffering from epilepsy and madness were more in the Eastern province (91.2% and 72.4%, respectively) than in the Southern province (16.5% and 6.5%, respectively). Only 24.3% of the pig farmers in the Eastern and 3.2% in the Southern provinces had heard about someone who had a tapeworm infection.

3.10. Logistic regression analysis

The factors that were considered in the analysis of risk factors associated with prevalence of households with at least one pig with cysticercosis are shown in Table 3. While adjusting for other factors in the model, only husbandry system was significantly associated with porcine cysticercosis. Households that practiced free-range husbandry system were 68% more likely to have had at least one pig positive for cysticercosis compared to households that practiced semi-intensive husbandry system (OR = 1.68; 95% CI = 1.36–2.07).

4. Discussion

This study to our knowledge is the first in Zambia that has quantitatively investigated the risk factors associated with *T. solium* infection in pigs. We have shown that *T. solium* porcine cysticercosis is endemic and highly prevalent in the study areas and that pig keeping has been predominantly of the traditional type, characterized by free-range and semi-intensive management systems. Pigs are slaughtered mainly in the backyard or at illegal slaughter slabs where no meat inspection is performed.

Like the pilot study conducted by Phiri et al. (2002), our study also revealed higher prevalence of *T. solium* cysticercosis in pigs in the Southern province than Eastern province. This finding could be due to the fact that the small villages and limited use of latrines in Southern province (Gwembe in particular) could lead to pigs having more access to human faeces than in Eastern province. It may be anticipated also that the indigenous pigs that are predominant in Eastern province are more resistant to *T. solium* infection than the cross breeds (mostly crosses between Large white and Landrace breeds) found in Southern province (Phiri et al., 2006). In Gwembe district of Southern province, the households are isolated and surrounded by shrub woodlands that provide a conducive environment for open-air defecation compared to households in Monze district where *Miombo* (*Brachystegia Julbernardia*) woodlands predominate. *Miombo* woodland is more open compared to the shrub woodland. This might explain the observed differences in household prevalence of porcine cysticercosis in the two districts.

The fact that 58% of the households had no latrines implied that people from such households could have been using the nearby bush for defecation. This was supported by the observation in the Eastern province where most of the households that were located on the periphery of the villages had positive pigs on tongue examination (data not shown). This could suggest that the pigs in households along the edge of the villages were more exposed to tapeworm eggs than the ones in the interior of the village. The finding that in Eastern province, farmers practised more of free-range husbandry system than in the Southern province and yet had lower prevalence of households with infected pigs was rather a difficult finding to explain.

The keeping of pigs on free-range and humans defecating in the bush may have permitted pigs to have access to human faeces and could account for the high prevalence of *T. solium* infection in pigs found in our study. It has also been shown that pigs from households that practiced free-range husbandry system (OR = 1.68) were at a higher risk of acquiring cysticercosis than those raised on semi-intensive system. Thus free-range method of keeping pigs was a significant risk factor for porcine cysticercosis in the surveyed areas. This agrees with studies by Sarti et al. (1997) and Rodriquez-Canul et al. (1999), which showed that extensively raised pigs (free-range type of husbandry system) had a higher seroprevalence of cysticercosis than intensively raised pigs. Pig husbandry practices within the communities in Mexico and Cameroon were the main risk factors associated with porcine cysticercosis as free-ranging pigs have a much higher access to human faeces in communities with few or no latrines (Sarti et al., 1992; Pouedet et al., 2002). In Tanzania, Ngowi et al. (2004) found that the prevalence rate of porcine cysticercosis was considerably higher in pigs reared in households lacking latrines than in those reared in households that

were using latrines. Similarly in Peru, Diaz et al. (1992) reported a statistically significant association between latrines and households with at least one pig with lingual cysticercosis.

Majority of the farmers slaughtered pigs without inspection and many consumed and sold infected pork. Others did not eat infected pork but sold it to other people within their communities. Some farmers accepted eating infected pork when the carcass was not heavily infected and still others only ate such pork after drying it (data not shown). Though most of these farmers admitted eating infected pork, others could have eaten infected pork unknowingly if the carcasses had light (low cyst burden) infections of *T. solium* cysticerci. The fact that people conducted home slaughter and consumed un-inspected pork increased their chances of eating infected pork and being at great risk of acquiring taeniasis.

In our study, pig farmers did not know porcine cysticercosis *per se* despite giving it many names. The few that knew that tapeworm infection in humans was due to eating infected pork were ignorant of the fact that a person with taeniasis could infect another person. Sanchez et al. (1997) found that the less the population knew about the existence of the parasite, the greater the risk they had of being seropositive.

The finding that most of the farmers had heard of someone suffering from epilepsy and madness in their communities is suggestive that there could be widespread human cysticercosis and/or neurocysticercosis in these surveyed villages. However, up to date, no association between epilepsy and NCC has been reported in Zambia. In regions where cysticercosis in pigs is common, human cysticercosis and epilepsy prevalence are also usually high (Diaz et al., 1992; Sciutto et al., 2000). Birbeck (2000) found that epilepsy and febrile seizures were responsible for a significant burden of disease in the Southern province of Zambia. Patients with epilepsy had significantly less education than their sex-matched siblings and that epilepsy was under reported, under recognized, and under treated in that population (Birbeck, 2000).

The current study confirms that *T. solium* infection poses a public health risk not only to these rural areas but to urban centres as well. Pigs from rural areas eventually get transported to large population centres where the demand for pork is high. Besides, the migration of tapeworm carriers from rural areas to the city poses a higher transmission risk of cysticercosis when poor environmental and social conditions are present (Schantz et al., 1992).

In conclusion, the life cycle of *T. solium* in these surveyed areas is sustained because pigs have access to infected faeces, and cysticercosis-infected pork is available for consumption to both pig owners and to those who do not keep pigs. We, therefore, recommend that a human survey be conducted to verify the human exposure to taeniasis and/or cysticercosis in these surveyed areas. In addition, an education programme needs to be initiated as part of prevention and control.

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