

Aus dem Institut für Parasitologie und Tropenveterinärmedizin  
des Fachbereichs Veterinärmedizin der Freien Universität Berlin  
und dem International Livestock Research Institute



# Assessment of the parasitic burden in the smallholder pig value chain and implications for public health in Uganda

## Inaugural-Dissertation

zur Erlangung des Grades PhD of Biomedical Sciences  
an der Freien Universität Berlin

vorgelegt von

**Kristina Rösel**

Tierärztin aus Karl-Marx-Stadt (jetzt Chemnitz)

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“Overcoming poverty is not a task of charity, it is an act of justice.” (Nelson Mandela)

Dedicated to the smallholder pig value chain actors in Uganda.



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**List of abbreviations**

AIDS	Acquired immunodeficiency syndrome
AAT	Animal African trypanosomosis
ADG	Average daily (weight) gain
ASF	African swine fever
BMZ	Bundesministerium für wirtschaftliche Zusammenarbeit und Entwicklung (German Federal Ministry for Economic Cooperation and Development)
CGIAR	Global research partnership for a food-secure future (formerly Consultative Group for International Agricultural Research)
CI	Confidence interval
CIESIN	Center for International Earth Science Information Network
CRP	CGIAR Research Program
CRP A4NH	CGIAR Research Program on Agriculture for Nutrition and Health
CRP L&F	CGIAR Research Program on Livestock and Fish
CSF	Classical swine fever
DAD-IS	Domestic Animal Diversity Information System
DALYs	Disability Adjusted Life Years
ELISA	Enzyme-linked immunosorbent assay
FAO	Food and Agricultural Organization of the United Nations
FAOSTAT	Statistics Division of the Food and Agricultural Organization of the United Nations
FGD	Focus group discussion
FMD	Foot and mouth disease
GIS	Geographic information system
GIZ	Gesellschaft für Internationale Zusammenarbeit
HAT	Human African trypanosomosis
HIV	Human immunodeficiency virus
IFAD	International Fund for Agricultural Development
IgG	Immunoglobulin G
ILRI	International Livestock Research Institute
IFPRI	International Food Policy Research Institute

KCCA	Kampala City Council Authority
LC	Local government council
MAAIF	Ministry of Agriculture, Animal Industry and Fisheries (Government of Uganda)
NAADS	National Agricultural Advisory Service
NEMA	National Environment Management Agency
NGO	Non-governmental organization
OIE	World Organization for Animal Health
PCV	Porcine circovirus
PE	Participatory epidemiology
PRA	Participatory rural appraisal
PRRS	Porcine Reproductive and Respiratory Syndrome
RR	Rural production for rural consumption (value chain type)
RU	Rural production for urban consumption (value chain type)
SFFF	Safe Food, Fair Food project
SPVCD	Smallholder Pig Value Chain Development project
STH	Soil-transmitted helminths
UBOS	Uganda Bureau of Statistics
UU	Urban production for urban consumption (value chain type)
VEDCO	Volunteer Efforts for Development Concerns
WHO	World Health Organization



## 1 Preface

### 1.1 Introduction and study objectives

Population growth, urbanization and increasing consumers' demand for products of animal origin are fuelling the so-called Livestock Revolution which could provide poor smallholder farmers with income (Delgado et al., 2001; Thornton, 2010) and thus contribute to poverty alleviation. In low-income countries, meat and dairy consumption have grown at average annual rates of 5.1 and 3.6%, respectively, between 1971 and 2007 (Alexandratos and Bruinsma, 2012). Between 2000 and 2050, demand for livestock products is estimated to nearly double in sub-Saharan Africa (Thornton, 2010), while globally, potential improvements in food security and nutrition could benefit directly more than 750 million poor livestock keepers (Food and Agriculture Organization of the United Nations (FAO), 2011). Uganda's population is currently growing at 3.3% annually (Worldbank, 2015a), and has increased seven times since the 1950s. The country is experiencing rapid urbanization – like most developing countries – at currently 4.5% which could result in an increase in the population of Uganda's cities from 6.4 million today to more than 30 million within the next two decades (Worldbank, 2015b), demanding ever more food.

Pork is the most popular meat in the world today accounting for over 36% of the world meat production, followed by poultry and beef (FAO, 2015). In Uganda, pork has only become important over the past 25 years; the Statistics Division of the Food and Agricultural Organization of the United Nations (FAOSTAT) estimated pig numbers have grown rapidly from 716,400 in 1989 to 2.18 million in 2008 (FAOSTAT, 2016). The 2008 national livestock census recorded 3.2 million pigs (Ministry of Agriculture, Animal Industry and Fisheries (MAAIF)/ Uganda Bureau of Statistics (UBOS), 2009), significantly surpassing estimates of the FAO. Whereas pork accounted for barely 1% of the annual per capita meat consumption in Uganda in 1961, it made up 27% of all meat consumed in 2011 (FAOSTAT, 2011). Little information is available regarding the structure and composition of the pig sector in Uganda but it has been documented that almost one fifth of the Ugandan households own pigs, on average three (MAAIF/ UBOS, 2009), while up to 70% of all pork consumed in Uganda is consumed in urban and peri-urban areas (International Livestock Research Institute (ILRI), 2011).

The majority of pigs in Kenya, Tanzania and Uganda are estimated to be reared in smallholder households under extensive systems (Lekule and Kyvsgaard, 2003); e.g. mostly free-ranging with tethering. Conventional knowledge states that traditional production systems are wasteful and unprofitable due to poor feed conversion, low reproductive rates, final products of inferior quality, and losses due to high animal mortality. A high gastrointestinal parasitic burden and direct losses due to infectious animal diseases such as African swine fever, foot and mouth disease or rabies (all endemic in Uganda) could contribute to low productivity (World Organisation for Animal Health (OIE), 2016). Concurrent with growth in smallholder pig keeping and pork consumption, zoonoses such as mycobacterial infections and porcine cysticercosis have increasingly been reported (Muwonge et al., 2010; Waiswa et al., 2009; Phiri et al., 2003). However, traditional production also has advantages compared to intensive systems, for example the small inputs required in traditional production (little to no housing), and the pig's natural scavenging behaviour which allows them to feed on kitchen waste and agricultural waste that would otherwise be wasted.

The surging demand for livestock products provides an opportunity to set poor farmers on pathways out of poverty by increasing production outputs and market access. At the same time,

markets have become more demanding and threaten the continued presence of smallholder farmers: livestock diseases and other factors affect a constant supply while foodborne pathogens may impair the quality and safety of the farmers' products. While the presence of food safety hazards such as bacteria, parasites or drug residues in informally marketed food is high, the risk to human health is mostly unknown and current food safety management is both ineffective and inequitable.

In Uganda, intensification level may affect the parasitic burden of pigs and hence the profitability of pig farming as well as the risk to human health associated with pork borne parasites. The present study aimed to contribute to improving selected smallholder pig value chains in Uganda by increasing the knowledge on prevalent parasitic diseases.

Specific objectives are to:

1. understand whether parasites are perceived as a production constraint by farmers;
2. estimate the burden of selected endoparasites in pigs and pork at farm, slaughter and retail outlet level in three selected value chain types in Uganda. These parasites included common gastrointestinal helminths and protozoa, as well as two major porkborne zoonotic parasites *Trichinella* spp. and *Toxoplasma gondii*;
3. identify risk factors contributing to parasitic infections in pigs and pork, in order to better understand the epidemiology of parasitic diseases in these value chains with emphasis on pork borne zoonoses;
4. identify current practices that increase or reduce risks to public health associated with pork consumption;
5. assess the risk to public health through the consumption of pork infested with parasites.



## 1.2 Structure of the thesis

This cumulative thesis is structured as follows:

**Chapter 1** Preface: introduction, study objectives and structure of the thesis.

**Chapter 2** Literature review: outlines the history of pigs in Africa, describes the current smallholder pig production systems in sub-Saharan Africa, and reviews the current state of knowledge on parasitic pig diseases compromising farm productivity as well as selected parasitic diseases that are potentially transmitted to humans through pork consumption. The review refers to previous research from sub-Saharan Africa, and particularly Uganda.

**Chapter 3** Research context: describes the context of the study and how it fits into overarching research for development on smallholder pig value chains in Uganda. It further describes how study sites were selected, value chain systems defined and characteristics of the area under study.

**Chapter 4** Parasites as a production constraint: shows that gastrointestinal parasites are perceived as an important production constraint by smallholder pig farmers in Central and Eastern Uganda (Publication I) and summarizes baseline information on the prevalence and risk factors of the most common intestinal parasites (Publication II).

**Chapter 5** Parasites with implications for public health: provides baseline information on two pork borne zoonoses of global importance, namely infection with *Trichinella* spp. (Publication III) and *Toxoplasma gondii* (Publication IV), in smallholder pig value chains in Central and Eastern Uganda.

**Chapter 6** Knowledge, attitudes and practices of pork consumers: outlines the findings on common preparation and consumption practices as well as sources of pork for consumption in the study area to potentially draw conclusions on the risk to pork consumers (Publication V).

**Chapter 7** Summarizing discussion: discusses major findings, conclusions and limitations of the study, and generates assumptions for potential interventions and further research.

## 2 Literature review

### 2.1 The history of pigs in Africa

Literature on the domestication of pigs in Africa is scarce and highly debated (Amills et al., 2013; Blench, 2000). The domestic pig (*Sus scrofa domesticus*) was domesticated from its ancestor the wild boar, or Eurasian wild pig (*Sus scrofa*) which is a native to Europe, Asia and North Africa. There are three distinct wild members of the family Suidae in tropical Africa: the bushpig (*Potamochoerus porcus*), the warthog (*Phacochoerus aethiopicus*), and the giant forest hog (*Hylochoerus meinertzhageni*). It is still debated to what extent African wild Suidae were domesticated (Gifford-Gonzalez and Hanotte, 2011; Blench, 2000; Haltenorth and Diller, 1980); but evidence is appearing that bush pigs and domestic pigs have indeed mated (Rothschild et al., 2014).

Genetic and archaeological findings suggest that pig domestication began about 9,000 to 10,000 years ago at multiple sites across Eurasia, followed by their subsequent worldwide spread through increased exploration and commercialization (Amills et al., 2010; Larson, 2005). According to recent findings (Amills et al., 2013; Ramirez et al., 2009; Blench, 2000), it is likely that domestic pigs have been introduced to sub-Saharan Africa from the Near East via Egypt approximately 6,000 years ago, from the Far East during the Indian Ocean trade in the 7<sup>th</sup> century, and during European colonisation from the 15<sup>th</sup> century. By analysing the genetic diversity of African pigs, Ramirez et al. (2009) showed the existence of a clear genetic dichotomy between East and West Africa: While a clear Far Eastern genetic signature was found in pigs from the Indian Ocean coast of Africa, it could not be detected in pigs from the African Atlantic shores. The findings have been discussed in-depth (Gifford-Gonzalez and Hanotte, 2011; Ramirez et al., 2009). It has also been hypothesized that African pig breeds were domesticated locally and independently in the Nile Delta (Gautier, 2002; Houlihan, 1997); however, at present evidence is still weak (Amills et al., 2013). African pig breeds are still poorly characterised but according to the Domestic Animal Diversity Information System (DAD-IS) database hosted by FAO, there are currently 148 pig breeds in Africa (FAO, 2016).

Phenotypically, domestic pigs in Uganda are divided into two major groups, the so-called “local” pigs and the introduced “exotic” pigs. Local pigs are usually black with medium-sized, semi-erect ears, a straight tail and a long snout (Figure 2.1). Exotic breeds such as Large White (Figure 2.2), Landrace and Saddleback have been introduced during the colonial periods. Under local management practices, local and exotic breeds interbred (Figure 2.3). Other pig breeds such as Camborough<sup>®</sup> have recently become of interest to Ugandan pig keepers (Muhanguzi, 2012). The Camborough<sup>®</sup> (Figure 2.4) is a product (material pig line) of a South Africa-based franchise of a commercial pig breeding company called the Pig Improvement Company (PIC International)<sup>1</sup>. The breed is derived from original crosses followed by heavy selection for maternal traits. Approximately 40 Camborough<sup>®</sup> lines have been released by PIC over the past years, and some have been introduced to Uganda by the Ministry of Agriculture (personal communication).

The distribution of religion in Africa is another critical aspect of the history of pigs in Africa (Blench, 2000). Muslims and Ethiopian Christian Orthodox are forbidden to eat pork which is generally interpreted as a prohibition on any sort of contact with pigs. Islam spread to Africa in

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<sup>1</sup> <http://www.picgenus.com/>

the early 7<sup>th</sup> century when Muslims sought refuge in present-day Ethiopia during the Migration to Abyssinia (*hijarat*); in Uganda, they arrived only in the late 19<sup>th</sup> century (David, 2004). Until the spread of Islam, pigs were quite important, especially in the Egyptian and Berber cultures in Northern Africa, already 6,000 years ago (Blench, 2000). All aspects of pig-keeping have been relatively little researched in Africa. Even international research organizations – such as ILRI since its inception in 1974 – have not included pigs in Africa in its research agenda until recently. Blench (2000) argues this may have been related to prejudice against pigs from potential donor agencies but also the belief that – as monogastric animals – pigs may compete with humans for food.



Figure 2.1 Tethered local breed in Wakiso district, Central Uganda (Kristina Roesel).



Figure 2.3 Fully confined cross-breed pig in an intensive production unit in Wakiso district, Central Uganda (Kristina Roesel).



Figure 2.2 Exotic breed (Large White) in a commercial farm in Masaka district, Central Uganda (Kristina Roesel).



Figure 2.4 Ugandan pig farmer tends to her Camborough pigs (Edgar R. Batte in Uganda Daily Monitor, 7 November 2012 (Muhanguzi, 2012)).

## 2.2 Smallholder pig production systems in sub-Saharan Africa

Pig production has emerged as an important agribusiness over the past few decades. Not only in Uganda but in many regions of sub-Saharan Africa, e.g. Nigeria (Machebe et al., 2009; Adesehinwa et al., 2003), Ethiopia (Tekle et al., 2013; Seid and Abebaw, 2010), Tanzania (Karimuribo et al., 2011; Esrony et al., 1997), Namibia (Petrus et al., 2011) and Kenya (Obonyo et al., 2013; Kagira et al., 2010b; Mutua et al., 2010; Wabacha et al., 2004a) to mention but a few. Given population growth and the pressure on the size of farm land, monogastrics seem to provide better production efficiency per unit area of land (FAO, 2009; Wabacha et al., 2004a). In Western Kenya, for instance, the average size of a smallholder pig farmers' land is one acre<sup>2</sup> or less (Mutua et al., 2011a; Kagira et al., 2010b). Pigs are often reared by women (Obonyo et al., 2013; Petrus et al., 2011; Kagira et al., 2010b; Mutua et al., 2010; Nsoso et al., 2006) as one of several income-generating activities, and kept in close proximity to homesteads (Obonyo et al., 2013; Karimuribo et al., 2011; Mutua et al., 2010). Income generation as well as savings and insurance against family emergency is often a more important motivation for pig keeping than providing meat for household consumption (Mbuthia et al., 2015; Tekle et al., 2013; Mutua et al., 2010). The regular smallholder in Kenya and Uganda keeps 3-5 pigs (Kagira et al., 2010b; MAAIF/UBOS, 2009); and herd size increases with proximity to urban centres (MAAIF/UBOS, 2009), with the level of intensification (Mbuthia et al., 2015; Wabacha et al., 2004a), or concentrates in certain areas; this is the case in Ethiopia where pig keeping is increasing but not as commonly practiced due stringent religious laws (Tekle et al., 2013). Most of the pig farmers in traditional production systems in sub-Saharan Africa keep cross-breeds (Kagira et al., 2010b) as they believe local breeds require less feeds and are more tolerant to diseases (Petrus et al., 2011; Lekule and Kyvsgaard, 2003) while exotic breeds grow faster.

Many small-scale farmers sell live animals produced in farrow-to-weaner (piglet) and porker-to-finisher (fattening) units; few keep boars and those who do often rent them out to other farmers for breeding (Muhanguzi et al., 2012; Kagira et al., 2010b), often against in-kind payment with a piglet. Many of the pigs are kept in mixed confinement systems, often tethered, especially during crop season, and otherwise free-roaming, especially during dry seasons when crop fields are fallowing and feeds are scarce (Obonyo et al., 2013; Mutua et al., 2011a; Permin et al., 1999). Housing is difficult to provide for many farmers because it is considered too expensive, and shelter is often just temporary or rarely cleaned (Muhanguzi et al., 2012; Kagira et al., 2010b; Seid and Abebaw, 2010). A study in rural Western Kenya reported that if farmers did not consider housing unnecessary altogether, the main reasons for non-confinement are fear of the pig damaging the houses, lack of food to provide for confined pigs, houses becoming muddy during the rainy season and farmers lacking the time to manage confined pigs (Mutua et al., 2011a). Smallholders' pigs are mostly fed with locally available ingredients such as crop residues (Muhanguzi et al., 2012; Kagira et al., 2010b; Petrus et al., 2011) but in some areas pigs are left grazing/scavenging or fed with household leftovers (Obonyo et al., 2013; Seid and Abebaw, 2010), supplemented with commercial feeds such as sunflower seed cake (Karimuribo et al., 2011), wheat bran (Tekle et al., 2013), or ruminal contents from abattoirs (Muhanguzi et al., 2012). Marketing is often disorganised and economically inefficient because record keeping is not common (Nsoso et al., 2006; Wabacha et al., 2004a); farmers mostly sell when they are in need (e.g. for school fees), and they do not weigh their animals and have therefore no objective basis for negotiation (Mutua et

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<sup>2</sup> one acre equals approx. 4,000 square metres

al., 2011a, 2011b; Petrus et al., 2011). Many other knowledge gaps on pig husbandry such as how to feed monogastrics, regular provision of water and the difference between treatment and vaccination have been documented in Kenya, and some of them seem to depend on the presence and a positive attitude towards pig-keeping of the veterinary extension service in a region (Obonyo et al., 2013; Mutua et al., 2011a; Kagira et al., 2010b).

Pig diseases (especially African swine fever and parasitic infestation), cost of feeds, lack of market access and veterinary extension as well as lack of knowledge on pig husbandry and disease prevention are perceived to be the greatest production constraints in many pig-keeping communities in sub-Saharan Africa (Atuhaire et al., 2013; Tekle et al., 2013; Muhanguzi et al., 2012; Karimuribo et al., 2011; Petrus et al., 2011; Kagira et al., 2010b). Important parasitic diseases (based on signs observed by farmers) are sarcoptic mange, lice and gastrointestinal worms (Karimuribo et al., 2011; Petrus et al., 2011; Kagira et al., 2010b). Different pig production systems in Uganda are illustrated in Figures 2.5-2.10.





Figure 2.5 Pig tethered under a tree and provided with sweet potato vines for feed, Kiboga district in Central Uganda (Kristina Roesel).



Figure 2.6 Adult pig confined but piglets able to roam freely in Kamuli district, Eastern Uganda (Kristina Roesel).



Figure 2.7 One type of confinement in a rural setting in peri-urban Wakiso district, Central Uganda (Kristina Roesel).



Figure 2.8 Pigs confined in Kamuli district, Eastern Uganda (Kristina Roesel).



Figure 2.9 Confined pigs in peri-urban Masaka, Masaka district, Central Uganda (Kristina Roesel).



Figure 2.10 Commercial pig farm with biosecurity protocol in peri-urban Kampala (Kristina Roesel).



Table 2.1 Simplified and selective classification of pig endoparasites. Adapted from Deplazes et al. (2016), Gajadhar (2015), Greve (2012), Lindsay et al. (2012), Kaufmann (1996).

	Order: species	Common name	Location in the pig host	Economic relevance	Zoonotic relevance
Cestoda	Cyclophyllidea: <i>Taenia solium</i> , larval	Pork tapeworm, pork measles	Muscle tissue	At slaughter if infected meat is condemned.	Ingestion of <i>Cysticercus cellulosae</i> in pork causes taeniosis in humans; infection in pigs helps maintaining the life cycle. High risk for humans developing neurocysticercosis when ingesting <i>T. solium</i> eggs (autoinfection).
	Cyclophyllidea: <i>Taenia hydatigena</i> , larval		Omentum, mesentery	Losses through trimming at slaughter.	N/A
	Cyclophyllidea: <i>Echinococcus granulosus s.l.</i> , larval	Hydatid disease	Liver, lungs	At slaughter if infected organs are condemned.	Pigs can be a reservoir for echinococcosis in carnivores (maintain life cycle). Humans are accidental hosts when ingesting eggs of <i>E. intermedius</i> (pig strain G7) passed in dog faeces.
Trematoda <sup>3</sup>	Echinostomida: <i>Fasciola hepatica</i>	Liver fluke	Liver (bile system)	At slaughter if infected livers are condemned.	Accidental infection of humans through ingestion of encysted metacercaria.
	Strigeatida: <i>Alaria alata</i>		Muscle tissue	At slaughter if infected meat is condemned.	Emerging parasitic disease. Human infection through ingestion of mesocercaria.
	Plagiorchiida: <i>Paragonimus</i> spp.		Lungs	N/A	N/A
Nematoda <sup>4</sup>	Ascarida: <i>Ascaris suum</i>	Large intestinal roundworm	Lumen of small intestine	Major growth reduction if piglets are infected.	Especially for children in poor pig-keeping communities.
	Enoplidae: <i>Trichuris suis</i>	Whipworm	Large intestine	Haemorrhagic enteritis in heavy infection; weight losses in chronic infections.	Potential still under debate.
	Rhabditida: <i>Strongyloides suis</i> (syn. <i>ransomi</i> )	Threadworm	Epithelium small intestine	Major impact in suckling piglets.	Larva migrans cutanea (creeping eruption).
	Strongylida: <i>Hyostrongylus rubidus</i>	Red stomach worm	Stomach fundus	Suck small amounts of blood and cause catarrhal gastritis.	N/A
	Strongylida: <i>Metastrongylus apri</i>	Lung worm	Bronchial tubes and bronchioles	Respiratory disease, especially in young pigs; can facilitate infection with pathogens causing pneumonia.	N/A
	Strongylida: <i>Oesophagostomum</i> spp.	Nodular worm	Large intestine	Reoccurring enteritis with every new infection (no protective immunity).	N/A
	Strongylida: <i>Globocephalus</i> spp.	Pig hookworm	Attached to mucosa of small intestine.	Aenemia in growers.	N/A
	Strongylida: <i>Ollulanus tricuspis</i>		Stomach	Weight loss and chronic-catarrhal gastritis.	N/A
Strongylida: <i>Stephanurus dentatus</i>	Swine kidney worm	Peri-renal fat tissue	Reduced growth; trimming at slaughter due to abscesses.	N/A	

<sup>3</sup> less significant trematodes reported in pigs: *Dicrocoelium hospes*, *Gastrodiscus aegyptiacus*, *Postharmostomum suis*, *Schistosoma bovis*, *Schistosoma mathei*, *Eurytrema pancreaticum*, *Gongylonema pulchrum*, *Prosthogonimus cuneatus*<sup>4</sup> less significant nematodes reported in pigs: *Gongylonema pulchrum*, *Setaria congolensis*, *Gnathosoma hispidum*, *Trichostrongylus axei*

Table 2.1 (continued). Simplified and selective classification of pig endoparasites. Adapted from Deplazes et al. (2016), Gajadhar (2015), Greve (2012), Lindsay et al. (2012), Kaufmann (1996).

	Order: species	Common name	Location in the pig host	Economic relevance	Zoonotic relevance
Nematoda <sup>4</sup>	Oligacanthorhynchida: <i>Macracanthorhynchus hirudinaceus</i>	Thorny-headed worms	Attached to mucosa of small intestine	Only if burden is high.	Accidental infection through ingestion of infected arthropod.
	Spirurida: <i>Physocephalus sexalatus</i> ; <i>Ascarops strongylina</i> ; <i>Gnathosoma spinigerum</i> ; <i>Simonsia paradoxa</i>	Thick stomach worms	Stomach	Less important than <i>H. rubidus</i> .	N/A
	Spirurida: <i>Gongylonema pulchrum</i>	Esophageal worm	Epithelium of tongue and esophagus	Minor irritation.	Humans are susceptible but must ingest intermediate host insect to become infected.
	Trichocephalida: <i>Trichinella</i> spp.	Pork worm, muscle worm	Adults in epithelium of small intestines; first-stage larvae encysted in striated muscle.	Not in pig farms but potentially at slaughter if infected carcasses are condemned.	Human trichinellosis mostly caused by ingestion of viable first-stage larvae in infected and undercooked pork.
Protozoa <sup>5</sup>	Cryptosporida: <i>Cryptosporidium</i> spp.		Epithelium of small intestine	Diarrhoea in growers.	<i>C. parvum</i> and <i>C. suis</i> are human-infective.
	Eimeriida: <i>Eimeria</i> spp.		Epithelium of small intestine	Diarrhoea in weaners and finishers.	N/A
	Eimeriida: <i>Isospora suis</i>		Epithelium of small intestine	Diarrhoea in piglets.	N/A
	Eimeriida: <i>Toxoplasma gondii</i>		Muscle and brain tissue	Not in pig farms but potentially at slaughter if infected carcasses are condemned.	Undercooked pork considered source of foodborne infection; occupational exposure to infective bradyzoites.
	Eimeriida: <i>Sarcocystis</i> spp.		Muscle tissue	On farm piglet diarrhoea.	Transient diarrhoea in humans.
	Eimeriida: <i>Neospora caninum</i>		Nervous tissue	On farm piglet losses due to abortions and stillbirth.	N/A
	Piroplasmida: <i>Babesia trautmanni</i>	Porcine piroplasmosis	Erythrocytes	Up to 50% mortality in infected pigs of all ages.	N/A
	Trypanosomatida: <i>Trypanosoma</i> spp.	Nagana	Extracellular blood	Peracute death in pigs caused by <i>T. simiae</i> or <i>T. suis</i> ; reservoir for cattle pathogenic trypanosomes.	Pigs as reservoir for human pathogenic <i>Trypanosoma</i> spp.
	Diplomonadida: <i>Giardia duodenalis</i> <sup>6</sup>		Small intestine	Subclinical	Assemblages A and B causative agent of human giardiasis.
	Vestibuliferida: <i>Balantidium coli</i>		Large intestine	Haemorrhagic enteritis if burden is high.	Faecal-oral transmission route; low pathogenicity but discussed as contributing factor to colitis.

<sup>4</sup> less significant nematodes reported in pigs: *Gongylonema pulchrum*, *Setaria congolensis*, *Gnathosoma hispidum*, *Trichostrongylus axei*<sup>5</sup> less significant protozoa reported in pigs: *Tritrichomonas suis*, *Trichomonas rotunda*, *Tetratrichomonas buttreyi*, *Entamoeba suis* (syn. *polecki*)<sup>6</sup> syn. *lamblia*, syn. *intestinales*

Table 2.2 Common ectoparasites of domestic pigs and their relevance to the pig farm enterprise and public health. Adapted from Deplazes et al. (2016) and Greve and Davies (2012).

	Order: species	Common name	Economic relevance	Zoonotic relevance
Arthropoda	<i>Astigmata: Sarcoptes scabiei</i> var. <i>suis</i>	Sarcoptic mange mite, burrowing mite	Reduction of growth rates, feed efficiency, and fertility.	Transient and self-limiting dermatitis in humans.
	<i>Prostigmata: Demodex phylloides</i> (syn. <i>suis</i> )	Follicular mange mite, follicle mite	N/A	N/A
	<i>Metastigmata: Ornithodoros moubata</i> complex	Soft tick (e.g. eyeless tampan)	Vector for African swine fever virus.	Vector for rickettsiosis.
	<i>Metastigmata: Amblyomma/Rhipicephalus</i> spp.	Hard tick (e.g. bont/blue/brown tick)	Nuisance and hence stress-induced reduced growth rate or anaemia; vector for porcine babesiosis	N/A
	<i>Phthiraptera: Haematopinus suis</i>	Hog louse, sucking louse	Reduced growth and increased susceptibility to disease in piglets due to anaemia; discussed as vector for African swine fever virus.	N/A
	<i>Siphonaptera: Tunga penetrans</i>	Sand flea, jigger flea	Nuisance	Pigs as reservoir for human tungiasis.
	<i>Siphonaptera: Pulex irritans/ Ctenocephalides felis/ Echidnophaga gallinae</i>	Human flea/ cat flea/ sticktight flea	Nuisance	N/A
	<i>Diptera: Musca domestica</i>	House fly	Nuisance	Mechanical vector of pathogens (faecal-oral route).
	<i>Diptera: Stomoxys calcitrans</i>	Stable fly	Nuisance and potential vector of pig pathogens	N/A
	<i>Diptera: Chrysomya bezziana, Calliphora</i> spp. etc.	Screwworm/ blow fly/ carrion fly/ flesh fly	Excavation of primary wounds (myiasis, or fly strike) and exposure to secondary infections	N/A
	<i>Diptera: Glossina</i> spp.	Tsetse fly	Vector for <i>nagana</i> in pigs caused by <i>T. simiae</i> or <i>T. suis</i> ; vector for cattle pathogenic <i>Trypanosoma</i> spp.	Vector for human pathogenic <i>Trypanosoma</i> species for which pigs can be reservoirs.
	<i>Diptera: Aedes</i> spp./ <i>Culex</i> spp.	Mosquito	Mechanical vector for PRRS virus and <i>Mycoplasma (Eperythrozoon) suis</i> .	Potential vector for human pathogenic arboviruses (e.g. Japanese encephalitis, West Nile fever, Tahyna).
	<i>Diptera: Phlebotomus</i> spp.	Sand fly	N/A	N/A
	<i>Diptera: Simulium</i> spp.	Black fly/ gnat	Nuisance	N/A
<i>Diptera: Culicoides</i> spp.	Biting midge	Nuisance	N/A	

### 2.3 Parasites of domestic pigs and their occurrence in East Africa

In general, pig parasites can be divided into two major groups: endo- and ectoparasites. Endoparasites live within the host, either inside the gastrointestinal lumen or organ tissue such as liver, lungs or muscle. Many of them are heteroxenous, an important trait that needs to be considered in diagnostics, management and control. This group of endoparasites in pigs (Table 2.1) includes representatives of major economic importance and the potential to cause disease in humans. Ectoparasites (Table 2.2) are usually found outside of the pig's body, on or in the skin, either temporary or permanently. Many of them are economically important as a nuisance and as a consequence, decreased weight gain due to the animals' pressure to deter them. Others are known vectors for more debilitating diseases, including parasites, such as ticks transmitting the African swine fever virus or tsetse flies carrying *Trypanosoma* species. The following section outlines the biology and occurrence of the endoparasites economic and veterinary public health importance that are subject to this study, with particular reference to East Africa and Uganda. For the denomination of parasitic diseases or infections, the author followed the *Standardized Nomenclature of Animal Parasitic Diseases* (Kassai et al., 1988).

#### 2.3.1 Trematoda (flukes)

*Fasciola (F.) hepatica* and *Fasciola (F.) gigantica*, the common or gigantic liver flukes, are trematodes with a broad range of mammal hosts including ruminants, horses, camels, pigs and humans. Pigs are not very susceptible to infection with *Fasciola* but can be carriers of adult flukes shedding eggs (very rare). The World Health Organization (WHO) estimates that at least 2.4 million people are infected in more than 70 countries worldwide with several million at risk; and that no continent is free from fasciolosis, and it is likely that where animal cases are reported, human cases also exist (WHO, 2016a). In livestock production, the burden is mainly on herbivores, especially ruminants (sheep > goat > cattle), and the result of reduced growth performance, anaemia and condemnation of infected livers if meat inspection takes place and condemnation is enforced. In Kenya, liver condemnation at beef cattle abattoirs due to fasciolosis led to losses worth \$25,000 in 2007 and 2008 (Kanyari et al., 2012). The life cycle is indirect whereby the pigs ingest encysted metacercariae with contaminated food (pasture) or water. Within one day, juvenile flukes penetrate the intestinal wall and migrate to the liver where they migrate the tissue for about six weeks, and then move through the bile system. This can take months to years; from there up to 20,000 eggs per female per day are produced 8 to 10 weeks post infection at the earliest. The eggs are passed in the pig faeces and develop in water where a miracidium hatches and penetrates freshwater molluscs (*Galba trunculata* or *Lymnaea natalensis*) where it produces hundreds of cercariae. These encyst on plants where they develop into infective metacercariae. They can survive up to 10 weeks in a moist environment such as wetlands. In Uganda *F. gigantica* has been reported in bovines from Mt. Elgon (Howell et al., 2012); and both *F. gigantica* and *F. hepatica* were reported from the tropical highlands in Tanzania (Walker et al., 2008). Kagira et al. (2012) reported 3% of the pigs in Western Kenya infected with *Fasciola* spp. using McMaster fecal flotation technique.

#### 2.3.2 Nematoda (roundworms)

*Ascaris (A.) suum*, the large roundworm of the small intestine, is cosmopolitan in domestic and wild Suidae. Infection is particularly important in the pig industry for two reasons: first, the adult worms compete with their pig host for nutrients through disruption of enteral absorption which

reduces feed efficiency and average daily gains (Hale et al., 1985); secondly, once on the pig farm, it is difficult to control because the eggs are extremely resilient and difficult to eliminate. Reproduction follows a direct life cycle: ingestion of the egg with infectious third-stage larvae, which hatches in the lumen of the jejunum and penetrates the intestinal wall, then migrates into the liver via the portal vena where it migrates the tissue for one to two days causing temporary inflammation and the well-known milk spots which disappear again after about 25 days once larval burden decreases. Within the next week larvae are carried to the lungs where migration causes inflammation and, potentially, respiratory signs. In the lungs larvae moult and further move to the bronchioles from where they are coughed up and swallowed. About two weeks post infection the larvae reach the small intestine again where they mature into adults. After another 4-6 weeks females start laying eggs, sometimes more than 200,000 per day for about six months. The un-embryonated eggs have a thick shell, which enables them to survive in moist soil for up to five years. Most chemicals do not have an effect on the egg and they have a sticky coat which enables them to be easily carried from farm to farm by live vectors (i.e. flies, beetles, cats or dogs but also children) or other vectors (i.e. boots, vehicles, tools). Also, they can stick to the sow's coat from where nursing piglets ingest the eggs when suckling. Infection and clinical signs occur mostly in pigs younger than five months, with age there is increased immunity though adult pigs may still harbour worms that produce eggs intermittently. *A. suum* can infect humans, and recent research including the application of molecular diagnostics increasingly suggests the possibility that *A. lumbricoides* may be closely related or even be the same species as *A. suum* (Iñiguez et al., 2012; Leles et al., 2012; Liu et al., 2012; Peng and Criscione, 2012; Zhou et al., 2012).

***Trichuris (Tr.) suis***, due to its whip-like oesophagus, is commonly known as the whipworm and primarily located in the large intestine where it permanently penetrates the epithelium. This causes minimal lesions which are considered to offer an entry point for swine dysentery complex (Beer, 1973; Greve, 2012); heavy infections are associated with haemorrhagic enteritis while chronic infection may cause reduced growth. In experiments, pigs infected with *Tr. suis* required up to 33% more feeds (Hale and Stewart, 1979). Reproduction follows a direct life cycle, whereby the egg with an infectious first-stage larvae is ingested and penetrates the walls of the smaller intestine and caecum. Four moults inside the wall follow over the next two weeks and six to seven weeks post infection, eggs are shed intermittently over the five month life span of the adult worm (Beer, 1973). It takes the first-stage larvae two months to reach infectivity which makes herd infection better manageable. However, the eggs can remain viable on the ground for many years. The results of genetic analysis of *Trichuris* species (*Tr. suis* and *Tr. trichuria*) obtained from naturally infected pigs and humans in Uganda suggest cross-infections of humans with *Tr. suis* (Nissen et al., 2012).

***Strongyloides (S.) ransomi*** is a very small threadworm (3-5 mm) that is found worldwide but especially in the tropical and subtropical areas where temperature and humidity are favourable. This roundworm is particularly pathogenic in nursing piglets (within first 10-14 days) and causes poor weight gains (stunting) due to the destruction of the small intestines' epithelium where the adult worms are located. Malabsorption, diarrhoea, dullness, hypo-albuminaemia, and alterations of organs (i.e. skin, liver, kidney, spleen, urinary bladder etc.) and even death may occur if the burden is high. Reproduction follows a direct life cycle, and infection of the pig host with third-stage larvae occur most often either orally through ingestion or by skin penetration. Through penetration of the buccal mucosa or thin skin (i.e. hoof skin, belly) they reach the lymph vessels and migrate to the heart and lungs from where they are coughed up and swallowed (tracheal migration). By the time they reach the small intestines (6 to 10 days), they have developed into

parthenogenic adult females laying embryonated eggs within a shell (up to 2,000 per day) that appear in the faeces. The larvae hatch within a few hours, and develop into an infective third-stage larvae within 3 to 4 days (and 2 moults). Besides that homogonic cycle, some of the larvae develop into free-living males and females who sexually produce infective larvae after a few generations (heterogonic cycle). A less common but potential infection of nursing piglets occurs via colostrum as infective larvae can accumulate in the sow's mammary fat for over 2.5 years; in the piglet, larvae develop directly into adult worms causing diarrhoea only a few days after infection. Acquired immunity is strong and therefore, older pigs are not usually presenting with clinical signs in case of infection.

***Hyostrongylus (H.) rubidus*** (red stomach worm) is a nematode specific to swine (Deplazes et al., 2016) whose adults are found lodged in the stomach where they suck small amounts of blood. At higher burdens infection causes catarrhal gastritis and eventually, the potential erosion of the gastric mucosa may cause significant weight losses (Stewart et al., 1985) as well as anaemia, lack of milk and fertility problems. Reproduction of the roundworm follows a direct life cycle, whereby the pig ingests the infective third-stage larvae which moults twice in the stomach's fundus glands and matures into adults. The first eggs with the characteristic strongyle morula appear in the faeces after 16 to 22 days. Within one week, a first-stage larvae has developed inside the egg, hatches and migrates from the faeces onto the grass from where it moults twice into infectious third-stage larvae and is eventually ingested by pigs feeding on pasture. In older animals, hypobiosis is possible after the first moult in the stomach fundus glands and they are potentially reactivated under peripartum stress.

Lung worms of porcines, the collective term for members of the genus ***Metastrongylus*** include ***Metastrongylus (M.) apri***, ***M. pudendotectus***, and ***M. salmi*** in pigs, and mixed infections are common. The adult worms (approx. 50 mm) are found in the pigs' bronchi and bronchioli, live for six to nine months and reproduce via indirect life cycle: the pigs ingest the infective third-stage larvae lodged in earthworms on pasture. Within the pig, larvae penetrate the intestinal wall and migrate through the lymph vessels to the right heart and lungs. Four to five weeks post infection, adults have developed and start producing eggs which are coughed up, swallowed and then passed in the faeces. They are thick-shelled with a rough coat and carry the embryonated first-stage larvae. Earthworms ingest them and the larvae hatch and moult within the intermediate host. After about ten days, the larvae in the earth worm has become infective for the pig. Clinical signs are only found in heavy infection but lung tissue may be compromised due to the larval migration and leaves the pig susceptible to secondary infection, e.g. ***Mycoplasma hyopneumoniae*** which then presents with coughing. In young pigs up to six months, clinical signs such as coughing, bronchitis, running nose, emphysema, anorexia and weight loss are more common; piglet mortality and stunting may be high (Deplazes et al., 2016; Greve, 2012). Prevalence rates vary across East Africa and are reported later in this section.

Two ***Oesophagostomum (Oe.) spp.***, ***Oe. dentatum*** and ***Oe. quadrispinulatum***, are found in pigs worldwide. All ages can be affected, but higher prevalence is usually found in older pigs where the worms accumulate with age due to lacking acquired immunity. Infection causes enteritis that may worsen with every new infection. The life cycle is direct: third-stage larvae are ingested by the pig and enter the mucosal glands of the large intestine where they undergo moulting in the lamina propria over the next two weeks causing the worm that gives rise to the common name of the worm (nodular worm). The adults persist in the lumen of the large intestine for up to six



months, eggs are shed three to six weeks post infection. Once excreted, the third-stage larvae develops in the faeces within one week and eventually, it moves away from the faeces onto the grass from where it is ingested by a pasture-fed animal. Infection with *Oesophagostomum* spp. is reported to extend the excretion of *Salmonella* Typhimurium in case of co-infection (Steenhard et al., 2002).

In East African pigs, infection with gastrointestinal helminths has been frequently reported over the past few years. Studies in Kenyan extensive and semi-intensive pig farms show that infection with strongyles dominate, and where larvae were cultured *Oesophagostomum* spp. are the most common followed by *H. rubidus* and a small fraction of *Trichostrongylus* spp. Infections with *S. ransomi* are common, too, while eggs of *Ascaris* spp., *Trichuris* spp., *Metastrongylus* spp. and spiruid eggs are found at varying levels (Obonyo et al., 2013; Kagira et al., 2012; Nganga et al., 2008). The same studies reported that mixed infections are very common but findings on risk factors diverged, especially with regards to the association between gastrointestinal worm prevalence and deworming history (incl. frequency), housing and type of feeds. Interestingly, in commercial farms in Central Kenya which supply formal pork processors and have a median of 60 animals, 94% of farms also showed infections with *Oesophagostomum* spp., *A. suum*, *T. suis* and *S. ransomi* despite 48% reporting to regularly treat their animals with dewormers as well as routine dung removal and disinfection (Kagira et al., 2008). In Tanzania, overall prevalences over 50% have been reported but infection with *Oesophagostomum* spp., *A. suum*, *S. ransomi*, and *T. suis* were documented (Esrony et al., 1997), and significantly higher egg counts were found in tropical highlands compared to the semi-arid zones. In Ethiopia, at least 25% of extensively managed pigs were infected with at least one species of gastrointestinal helminth: *A. suum* (25.9%), *F. hepatica* (1.8%), and *T. suis* (0.3%); moreover, 1.7% of the pigs carried oocysts of *Eimeria* spp. and 7% of *Cryptosporidium* spp., and 72% of the soil samples in the study sites were contaminated with *A. suum* eggs (Tomass et al., 2013). In growing pigs in Western Uganda, 89% of the animals showed strongyle infections, 40% infection with *A. suum*, 17% with *T. suis*, and 48% spiruroid eggs (Nissen et al., 2011). In the Eastern parts of the country, in Kamuli and Iganga districts, 94.8% were infected, mainly with strongyles (55.2%), *Metastrongylus* spp. (49%), *Coccidia* oocysts (68.6%), *Ascaris* spp. (42.2%), hookworms (18.6%), and *Trichuris* spp. (6.2%); 80.3 had mixed infections (Waiswa et al., 2007). Other helminths of the gastrointestinal system are less common and less pathogenic, except when burdens are very heavy (Table 2.1).

The nematode genus *Trichinella* is found everywhere except the Antarctica and comprises eight species and four genotypes: five have been reported from Africa. Figure 2.11 shows the distribution based on recent reviews (Mukaratirwa et al., 2013; Murrell and Pozio, 2011; Pozio, 2007; Pozio et al., 2005). While a wide range of host species can be infected (mammals, birds and reptiles), only humans show clinical signs (Gottstein et al., 2009); infection is strictly related to the consumption of raw or undercooked infected meat (Pozio, 2007). The life cycle is direct and starts with the ingestion of infectious first-stage larvae in infected muscle tissue. Within the pig, the larvae develop into adults (1-4 mm) within two days and lodge in the villi of the small intestine's epithelium. Females give birth to larvae in the lamina propria five days after mating which may cause subclinical enteritis. The larvae are then carried away in the blood stream into skeletal muscle cells which they transform into a nurse cell and where they mature into infective and encysted first-stage larvae within two weeks. If the larvae do not enter muscle cells but other organ tissue, they die in granulomas. The encysted muscle larvae is very resilient and survives many years, even if the cyst starts calcifying or the infected carcass starts decaying. Transmission

to pigs can occur in case of tail biting within an infected herd, scavenging carcasses of wild, infected animals (i.e. rodents) as well as feeding on garbage or kitchen swill that contains infected meat scraps. Humans get infected in the same way, by ingesting infectious first-stage larvae in raw or undercooked meat. The infection dose depends on the *Trichinella* species. *Trichinella* (*T.*) *spiralis* is the most important and most researched *Trichinella* species in veterinary public health and the ingestion of 500 first-stage larvae may result in diarrhoea, vomiting, high fever as well as headache and periorbital oedema. At two to three weeks after infection muscle pain occurs and death may result from cardiomyopathy and paralysis of respiratory muscles.

In 1962, Kozar presented a detailed and up-to-date account of the known distribution of *Trichinella*. At that time, apart from the Mediterranean region, the map of Africa was a complete blank (Nelson, 1970). Two years later, several outbreaks of the disease in humans in Kenya lead to the discovery of an enzootic sylvatic cycle (Nelson, 1970), and since then wild animals have increasingly been reported to carry *Trichinella* in Africa (Figure 2.11) (La Grange et al., 2013, 2010, 2009; Marucci et al., 2009; Pozio et al., 2007, 1997). Human outbreaks date back several decades and were documented in Egypt (Sayed et al., 2010), Northwest Ethiopia (Kefenie and Bero, 1992; Kefenie et al., 1988), Kenya (Kaminsky and Zimmerman, 1977) and Tanzania (Bura and Willett, 1977). Studies on domestic pigs are rare but increasing: *Trichinella*-specific antibodies have been found in 11-40% of surveyed pigs in Nigeria (Adediran et al., 2015; Momoh et al., 2013) and in 0.8% of a study cohort of pigs in Madagascar (Rakotoharinome et al., 2014). Attempts to isolate muscle larvae in pig abattoirs were unsuccessful in Tanzania and Ghana (Ngowi et al., 2004; Permin et al., 1999) but *T. spiralis* has been isolated by means of artificial digestion from domestic pigs in Egypt (Sayed et al., 2010; Morsy et al., 2000).

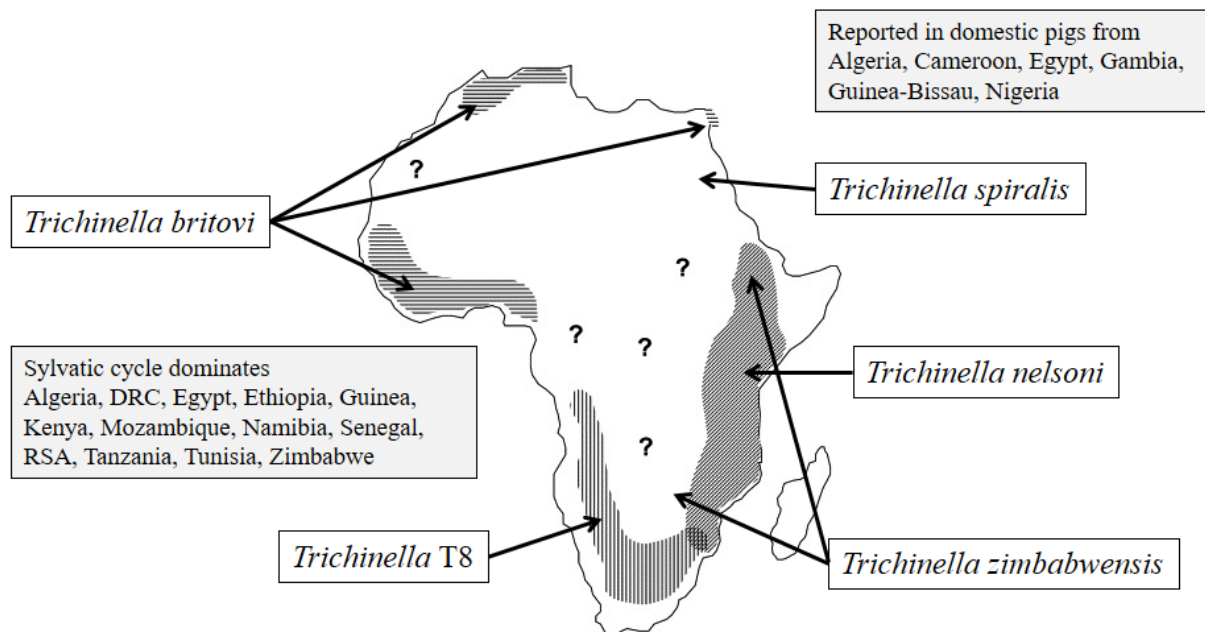


Figure 2.11 The distribution of *Trichinella* spp. in Africa reviewed by Mukaratirwa et al. (2013), Murrell and Pozio (2011), Pozio (2007), and Pozio et al. (2005). Map adapted from Pozio et al. (2005).

### 2.3.3 Protozoa

**Coccidia** are very host specific protozoa, and pigs are susceptible to (mixed) infection with *Eimeria* and *Isospora* (Levine and Ivens, 1986). Infection with *Eimeria* is usually considered subclinical; however, while experimental infection with *E. deblickei* did not cause clinical disease in suckling pigs or growers (Lindsay et al., 1987), infection with *E. spinosa* has indeed been reported to cause clinical disease in weaners and finishers (Lindsay et al., 2002; Yaeger et al., 2003). Infection with *Isospora (I.) suis* on the other hand can cause serious diarrhoea in nursing piglets (piglet coccidiosis) and is of much higher significance to the pig industry. *I. suis* is found globally, but mostly where pigs are kept in confinement. Infected suckling piglets appear healthy until they suddenly develop a yellowish-grayish diarrhoea (caused by a catarrhal-necrotizing enteritis), most often between the first and second week postpartum; morbidity in the litter is high, and mortality moderate but it can increase rapidly with concurrent bacterial, viral or parasitic infections (Lindsay et al., 2012). Piglets one to two days old at infection develop much more severe disease than if infected at two to four weeks of age (Koudela and Kučerová, 1999). After pigs ingest sporulated oocysts from their environment, they are excysted when passing the stomach and small intestines. Here, the sporozoites are activated, leave the sporocyst and penetrate the enterocytes. Endogenous development through asexual and sexual reproduction follows in the cytoplasm of the enterocytes of the small intestine. Five days post infection, pigs pass unsporulated oocysts with their faeces; these are not yet infectious but sporogony into environmentally very resistant oocysts follows rapidly after, if ambient temperature ranges between 20-37°C (Lindsay et al., 1982). Pigs develop partial immunity but small amounts of oocysts can be shed intermittently, and colostral antibodies do not prevent clinical coccidiosis in piglets. Studies have shown that sows are apparently not the source of infection for suckling pigs but that improved sanitation (including regular cleaning, disinfection, restricted farm access, pets not entering farm, rodent control, sanitizing after every farrowing) has been the most successful method for reducing losses due to piglet coccidiosis (Lindsay et al., 2012). In Eastern Uganda, 68.6% of the pigs have been found to pass coccidia oocysts (Waiswa et al., 2007); however, oocysts were not sporulated and the prevalence was determined at animal level, not herd level. Much lower levels of coccidia oocysts have been reported from Ethiopia (Tomass et al., 2013) and infection with *Eimeria* spp. has been recorded in 33% of pigs studied in Western Kenya (Kagira et al., 2010a).

**Balantidium spp.** is found in pigs and humans worldwide, but more commonly in tropical and subtropical regions (Thompson and Smith, 2011). It has direct life cycle similar to *Giardia*, except that infection is usually located in the large intestine. Infection in pigs and humans is usually subclinical, but it is discussed as a contributing factor to colitis. Infection in pigs increases with age but has not been found in pigs younger than four weeks (Schuster and Ramirez-Avila, 2008). If the burden is high, then haemorrhagic enteritis may occur as the protozoa invade mucosal tissue. Higher infection rates are sometimes associated with extensive pig keeping (reviewed by Schuster and Ramirez-Avila, 2008) but the extent of this is not yet known. In West Africa, *B. coli* has been found in pigs (Yatswako et al., 2007; Permin et al., 1999), raw vegetables (Ogbolu et al., 2009) and non-human primates. In Western Kenya, 64% of pigs in extensive systems have been found infected with *B. coli* (Kagira et al., 2010a).

Toxoplasmosis is a major zoonosis with a global distribution; it is caused by *Toxoplasma (T.) gondii*. Infection is common where susceptible hosts have access to food, water, or soil

contaminated with sporulated oocysts passed in cat faeces. The life cycle is indirect with pigs as intermediate host: pigs pick up sporulated oocysts from cat faeces or bradyzoites from infected carcasses in the environment when scavenging. The oocysts survive passage through the stomach and invade the lamina propria of the small intestine where they transform into quickly multiplying tachyzoites. From there spread into the body through the blood stream and form cysts in brain and muscle tissue for which they have a particular tropism. However, they can be found in virtually all organ tissue. When forming tissue cysts, tachyzoites turn into bradyzoites that keep multiplying, perhaps for the whole life of a food animal, but very slowly. If infected tissue of the intermediate host is ingested by the definitive feline host, asexual and sexual multiplication takes place in the cat's intestines from where oocysts are shed into the environment again. These oocysts sporulate within five days and can persist in moist and cool soil for up to one year. Tissue cysts have been shown to be viable for up to 2.5 years and can be rendered nonviable by freezing at  $-12^{\circ}\text{C}$  for three days (Dubey, 1988) or salting/curing meat (Hill et al., 2004). Clinical toxoplasmosis is rare in pigs (Dubey, 2009, 1986), but reproductive signs can occur such as abortion, stillbirth and foetal mummification in females infected for the first time during gestation (Feitosa et al., 2014); surviving piglets may be uncoordinated and show signs of tremor and coughing (Lindsay et al., 2012). However, this is highly unlikely in settings where extensive production systems dominate. Postnatal infection is more likely and may occur due to the ingestion of large numbers of oocysts, e.g. in pig feeds, and clinical signs may include anorexia, fever, dyspnoea, limb weakness and even death (Dubey, 2009; Weissenböck and Dubey, 1993). The presence of infected cats, rodents, or birds is suggested to be the main source of infection for pigs; and herd size (e.g. risk for cannibalism), source of water, disposal of dead pigs on the farm, garbage or swill feeding, cats used for rodent control in pig feed stock have been reported to be risk factors (Feitosa et al., 2014; Dubey, 2009; Weigel et al., 1995). Experimental studies also showed that dogs can be mechanical vector of oocysts but sporulation does not occur in dog fur (Frenkel et al., 2003). Attempts to develop pig vaccines are underway (Garcia, 2009).

Most human infections are asymptomatic in otherwise healthy people; approximately 25-30% of the world's human population is infected (Montoya and Liesenfeld, 2004) varying in geographical regions, also depending on dietary habits, and increasing in resource-poor communities with limited access to safe water (Bahia-Oliveira et al., 2003). The severity of toxoplasmosis depends on the infection dose (Dubey, 1986) and the virulence of the genotype. A recent review by Robert-Gangeux and Dardé (2012), found that in Africa the predominant genotypes are I and III (cosmopolitan), coexisting with the Africa haplogroups 1 to 3. The synthesis of data in the review also showed a higher rate of retinochoroiditis in Africa than in Europe. In Kampala, Uganda, all three lineages have been reported in HIV-patients but also in chickens (Lindström et al., 2008, 2006) as well as one recombinant isolate of type II/III (Bontell et al., 2009).

Recent advances in molecular epidemiology suggest that there are no species-specific strains of *T. gondii*, and that pigs can be infected with at least nine different genotypes (reviewed by Dubey, 2009). Nevertheless, studies on the prevalence of *T. gondii* infection in pigs in Africa is still limited. In Nigeria, ELISA-based prevalences ranged from 25% on farms to 45.2% in slaughtered pigs (Ayinmode and Abiola, 2016; Ayinmode and Olaosebikan, 2013) and 19.8% in dogs, which are often used for food (Ayinmode et al., 2015). In Malagasy pigs, a seroprevalence of 22.8% was reported (Rakotoharinome et al., 2014) while in Ghana 40.6% of 641 pigs were ELISA-positive (Arko-Mensah et al., 2000), and in Ethiopian pigs the proportion was 32.1% (Gebremedhin et al., 2015). Serological surveys in Zimbabwe showed that 9.3-26.8% of domestic pigs were ELISA-

positive, and IgGs have also been reported from wildlife and small ruminants (Hove, 2005; Hove et al., 2005; Hove and Dubey, 1999); mouse-pathogenic *T. gondii* was isolated from a slaughtered pig in Zimbabwe (Hove and Mukaratirwa, 2002). In Uganda, there is only one study on *T. gondii* infection in livestock. It has investigated the IgG prevalence in goats and found 31% of the 784 animals originating from all parts of the country sero-positive; herd prevalence was 100% and infection rates were significantly higher in urban locations (Bisson et al., 2000). Another sero-survey on dogs kept near conservation areas showed very high levels of infection with *T. gondii* and other, potentially zoonotic, agents (Millán et al., 2013).

In high-risk groups in the human population, namely sero-negative pregnant women or HIV-patients, infection with any strain may result in severe clinical disease including foetal death (abortion), congenital toxoplasmosis, encephalitis, or retinochoroiditis (Montoya and Liesenfeld, 2004). An estimated 50% of human toxoplasmosis can be attributed to foodborne transmission where consumers ingest viable bradyzoites from raw or undercooked meat (Torgerson et al., 2014). Estimates of the global burden have been limited to congenital toxoplasmosis for which the burden is high at 1.2 million DALYs (Torgerson and Mastroiacovo, 2013). *T. gondii* is considered an opportunistic pathogen and suggested to cause meningitis in HIV/Aids patients (Wright and Ford, 1995). A post mortem survey in Côte d'Ivoire showed cerebral toxoplasmosis in 21% of Aids patients (Lucas et al., 1993). A previously subclinical infection can be reactivated and become clinical if people's immune system is suppressed at a later time in life (Robert-Gangneux and Dardé, 2012), for instance through a disease or medication. Reactivation results from the rupture of tissue cysts, a continuing process that is considered responsible for the continuous immune response in a competent healthy human, which ensures dynamic control of the infection (Robert-Gangneux and Dardé, 2012). Apart from the above-mentioned infection routes, people working with infected meat have also been shown to be at higher risk to infection (Jones et al., 2009); potentially due to accidentally ingesting bradyzoites from tissue cysts ruptured during slaughter or processing.

### 3 The context for researching smallholder pig value chains in Uganda

Value chains resemble intricate webs and include all the links that start with the idea for a product and continue through to the consumption of the product, often beyond to the disposal of the product itself and its remainders. In the case of food products of animal origin, value chains include farm-level inputs and services that enable its production (e.g. feeds, breeding services, animal health services), transporting, processing, marketing and consumption of animal sourced foods and related products (Figure 3.1). Moreover, value chains include institutional and governance arrangements that facilitate the operation of these systems (e.g. Animal Disease or Meat Acts). Value chain analysis considers how and by whom the value is utilized. Past livestock research in low-income countries has focused on specific aspects of value chains, for instance on increased farm output (e.g. by implementing interventions that reduce livestock disease burden but which usually come at additional cost to the farmer) without considering if markets and purchasing power are readily available.

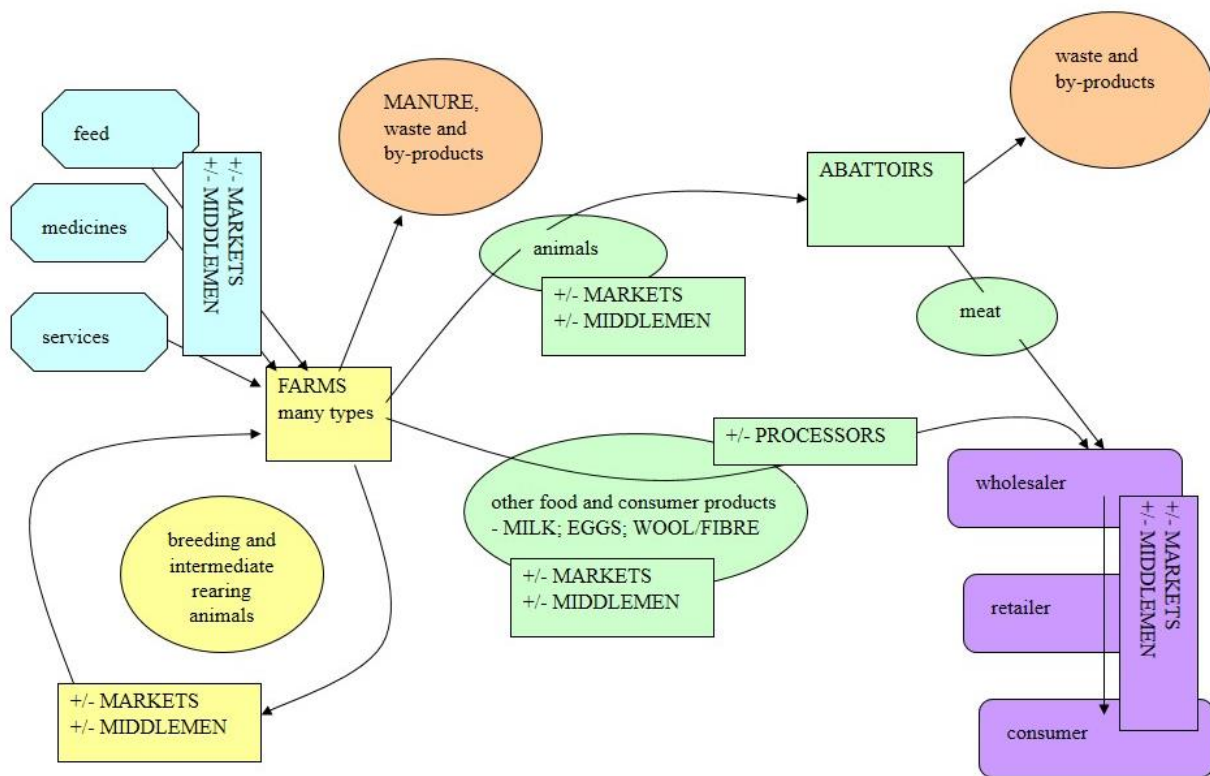


Figure 3.1 A livestock value chain and its actors and beneficiaries. Source: Nick Taylor, University of Reading, and Jonathan Rushton, Royal Veterinary College, In: Roesel and Grace (2014).

#### 3.1 CGIAR Research Framework

In January 2012, the CGIAR launched research programs (CRP), in which different CGIAR centres work in multi-centre and multi-disciplinary teams to increase research impact. The CRP

on Livestock and Fish (L&F), led by ILRI, has the overall goal to sustainably increase the productivity of small-scale livestock and fish systems in order to make animal sourced foods more available and more affordable for poor consumers, and in doing so, to reduce poverty and poverty-related impact through greater participation (ILRI, 2012a).

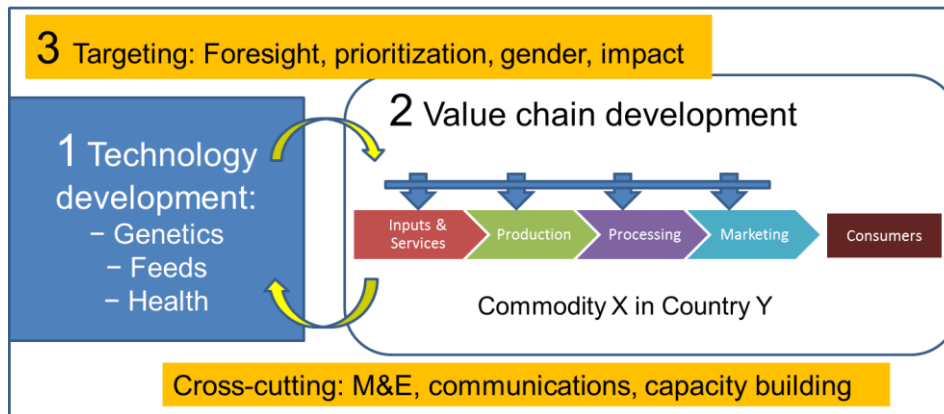


Figure 3.2 Delivering CGIAR Research Program on Livestock & Fish (CRP L&F). Structure: Three integrated components. Source: CRP L&F, 2012.

The program follows an integrated approach (Figure 3.2) to transform selected value chains in targeted livestock and fish commodities in nine countries, one of them being the smallholder pig value chain in Uganda. In 2012/13, as an initial step, the value chain was described, its actors and products mapped and constraints and opportunities for growth of the pig sector in Uganda assessed. In 2013/14, in-depth surveys quantified disease prevalence and other elements previously established as important constraints. The results and implications have been discussed with stakeholders and potential solutions to constraints tested in pilot studies from 2014 until today.

ILRI is also leading one of four components in the first phase of the CRP on Agriculture for Nutrition and Health (A4NH), led by the International Food Policy Research Institute (IFPRI). The rationale for this program is based on the assumption that agriculture provides poor people with nutritious food and income from agricultural products (IFPRI, 2012). However, agricultural activities, especially in livestock production, bear potential health risks. The Food Safety and Zoonoses program of ILRI leads an A4NH research component on the *prevention and control of agriculture-associated diseases* which has four major research areas: emerging infectious diseases, food safety, neglected zoonoses, and One Health/Ecohealth (ILRI, 2012b).

The thesis presented here was aligned with both of the programs outlined above and part of the research project *Safe Food, Fair Food: Risk-based approaches to improving food safety and market access in smallholder meat, milk and fish value chains in four African countries* under A4NH (ILRI, 2012c). The project's overall aim is to generate evidence on risks from animal sourced food products of which the majority in sub-Saharan Africa is sold in informal markets. Figure 3.3 outlines why food safety should be one of the components considered in a livestock value chain analysis and why this research was aligned with L&F.



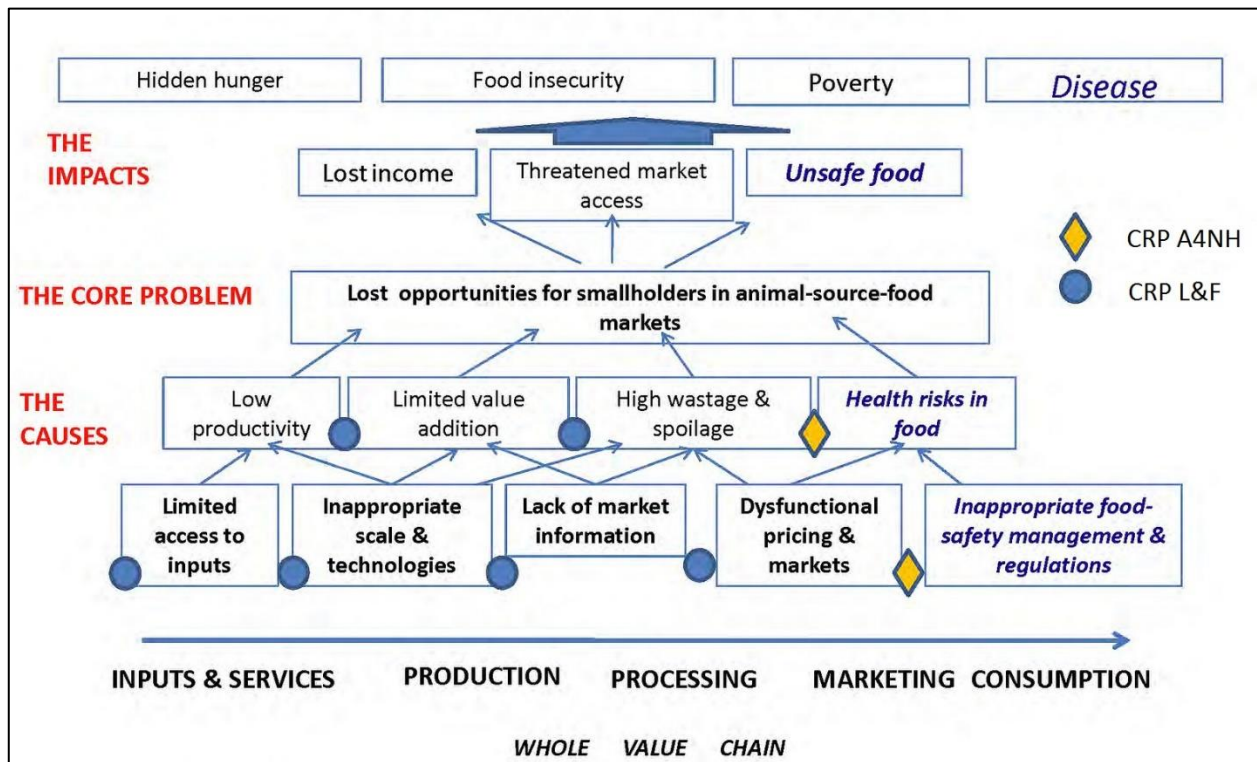


Figure 3.3 Problem tree showing food safety and related market access in the context of whole value chain development (ILRI, 2011).

### 3.2 Study area

Uganda is located in Equatorial Africa, west of Kenya, south of South Sudan, east of the Democratic Republic of the Congo and north of Tanzania and Rwanda. It is situated in the heart of the Great Lakes region and much of its borders being freshwater lakeshore; however, Uganda is landlocked with no access to the sea. The country covers a total surface area of 241,551 square kilometres of which about 17.3% are open water and wetlands (National Environment Management Agency (NEMA), 2009). Uganda’s climate is tropical but mostly temperate due to its location on the African Plateau with average altitudes between 900 and 1,500 metres above sea level (NEMA, 2009). While the semiarid north-eastern part of the country is characterized by an intense hot and dry season (November to March) followed by a single rainy season from April to August, the Central and southern parts of the country have seasonal rains with peaks in April/May and October/November but rainfall of lower intensity throughout the year (NEMA, 2009). Given the favourable climatic conditions, mixed crop-livestock systems are most common and covering just over 75.0% of the land (Robinson et al., 2011), 14% is under grasslands supporting agro-pastoral livestock production in the North-eastern part of the country, and mixed irrigated systems do not cover even one percent of the surface land area.

Uganda has one of the youngest and most rapidly growing populations in the world; from 2011 to 2015, it increased by 13.1% from 34.5 million to 39.05 million (Worldbank, 2016). For comparison, in 1991 the population counted 16.7 million (Twinomujini et al., 2011). More than 50% are younger than 15 years, well above sub-Saharan Africa’s average of 43.2%, and about



500,000 people are expected to enter the labour market every year (Worldbank, 2015c). Despite good economic progress over the past few decades, about 62.0% and 78.5% of the population still live on less than US\$ 1.25 and US\$ 2 per day, respectively (Center for International Earth Science Information Network (CIESIN), 2011; Wood et al., 2010). Since 1993, when the Ugandan Parliament enacted the Local Governments Statute, functions, powers and services were gradually transferred from the central government to local governments (local government councils, LC). By 2011, there were 112 districts spread across four administrative regions: Northern, Eastern, Central and Western. Each district (LC5) is further divided into countries (LC4), sub-counties (LC3), parishes (LC2) and zones/villages (LC1).

Pig keeping is practiced across the whole country with concentrations around Kampala, in the Central and Western regions and to the east in the Soroti-Mbale area (Figures 3.4 and 3.5). Per capita consumption of pig meat is 3.4 kg (FAOSTAT, 2011) with the highest consumption levels in urban areas and lowest in pastoral rangeland areas (CIESIN, 2011) (Figure 3.6).

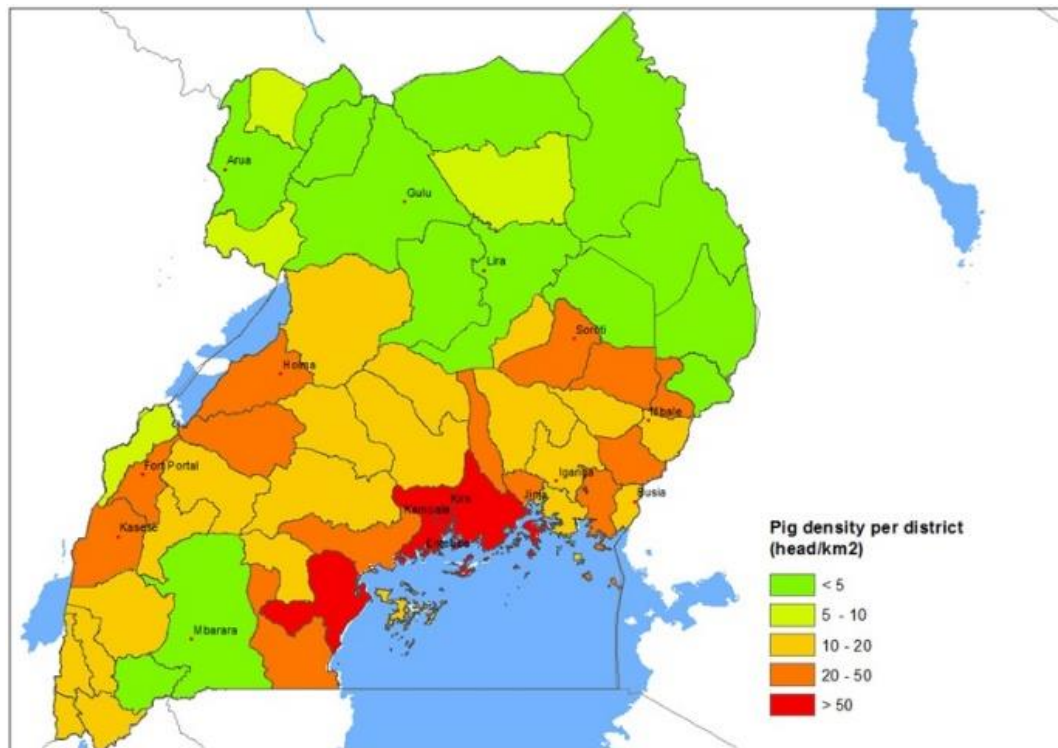


Figure 3.4 Pig density by district shows a higher concentration in the Central, Western and Eastern regions of Uganda. Source: Poole et al. (2015). Category thresholds are as accurate as 14 decimals.

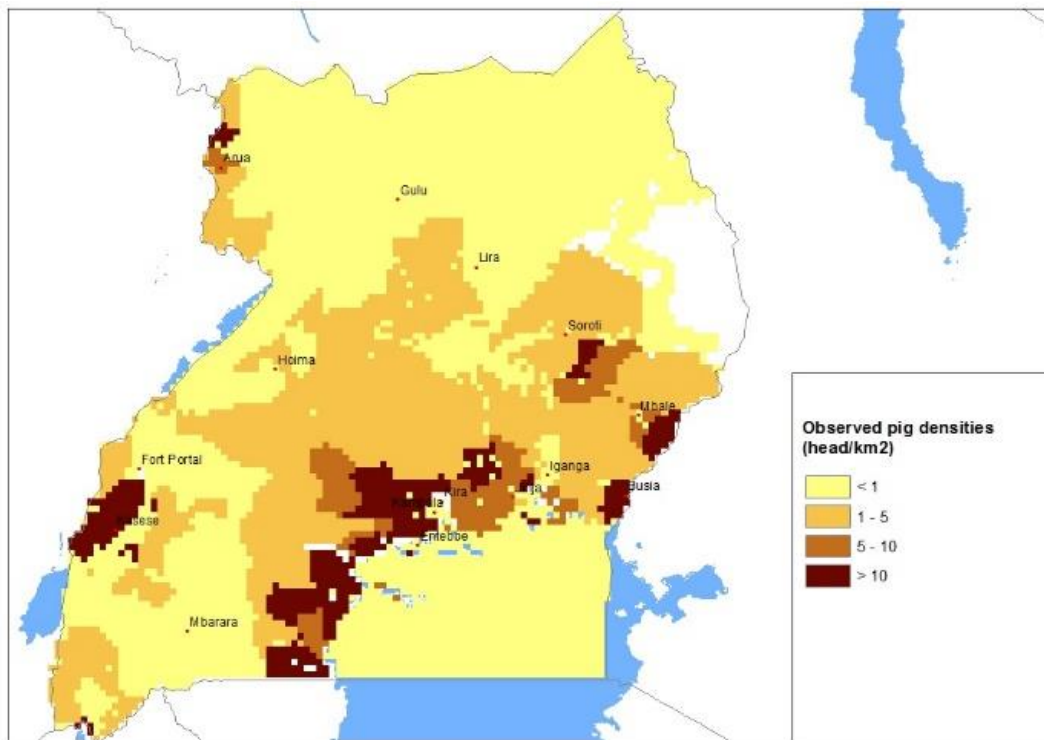


Figure 3.5 Average pig densities in Uganda show production specific production foci in Mbale, peri-urban Kampala, Kasese and Masaka. Source: Robinson et al. (2014). Category thresholds are as accurate as 14 decimals.

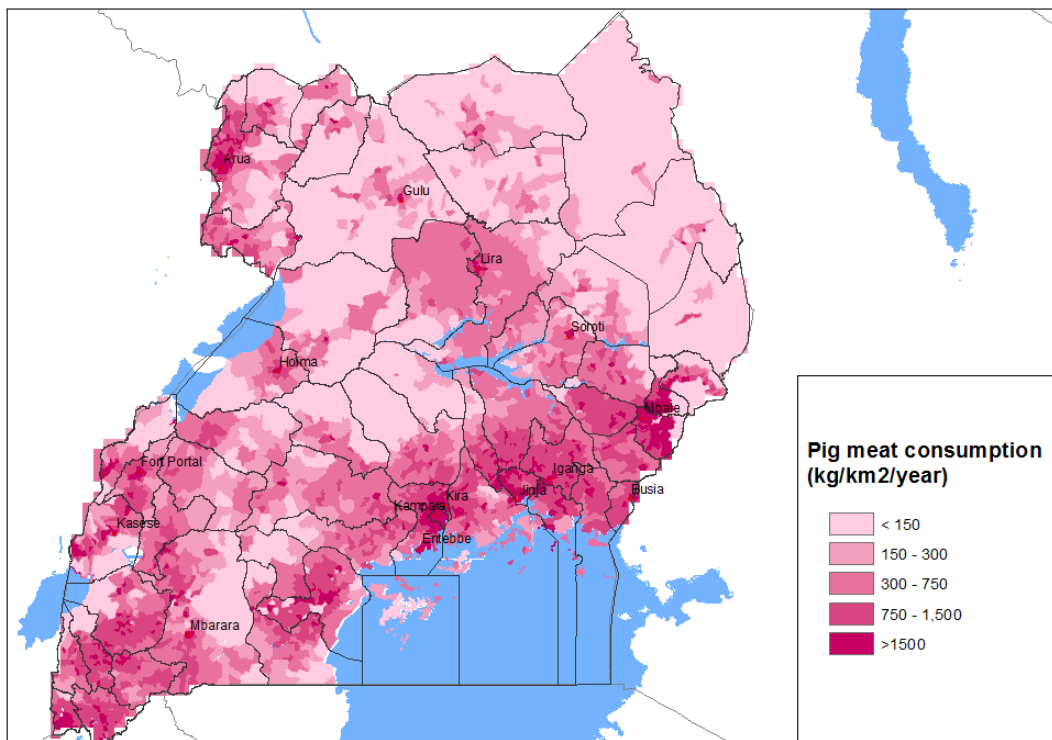


Figure 3.6 Spatial distribution of per capita pig meat consumption, based on population density. Source: Center for International Earth Science Information Network (CIESIN), 2011. Category thresholds are as accurate as 14 decimals.

### 3.3 Study site selection and characteristics

While in the long term, research findings are to be extrapolated and interventions scaled up and out to the whole of Uganda, the CRP L&F selected three districts for initial smallholder pig value chain scoping, assessment and piloting interventions. The process was completed in 2012 and involved

- 3.3.1 Geographical targeting** using GIS characterization by utilizing existing spatial data (Van De Steeg et al., 2011). Candidate districts were established and classified into value chain domains. The pre-selection of districts was based on data overlays of pig population density and poverty levels. The time needed to reach the nearest urban centre served as a proxy for market access and played an important role in classifying the districts into different value chain domains. These are based on the time needed to reach the nearest urban centre with a human population of at least 50,000 and describe the spatial dimensions of production and consumption of pork: rural production for rural consumption (rural-rural, RR), rural production for urban consumption (rural-urban, RU) and urban production for urban consumption (urban-urban, UU).
- 3.3.2 Stakeholder consultation** of step 3.3.1 and definition of soft selection criteria (based on local knowledge) during a site selection workshop (Ochola, 2012). Stakeholders critically discussed how poverty levels in some districts may be biased because there are great gaps between rich and poor. For Masaka district, for instance, the overall poverty levels seem to be low but it was emphasized that many farmers in the rural areas are poor but the average poverty levels are raised by the number of wealthy people in the urban areas. Other criteria such as the number of female-headed households, levels of HIV/Aids, cross-border trade, market demand, seasonality of market access and partnerships were thought necessary to be considered in site selection. From the discussion, some criteria were added: potential partnerships, disease burden in pigs, presence/access of input and service providers, and geographical access all year round. At the end of the workshop the participants were asked to vote by scoring the relation suitability of each candidate district established in 3.3.1 against the four soft criteria; voting excluded ILRI and other CGIAR staff or affiliates.

Masaka, Mukono and Wakiso districts were voted the top three. Wakiso and Mukono districts are both in close proximity to Kampala and are both considered peri-urban (UU value chain domain); the participants therefore agreed to select Mukono, while Wakiso was omitted to include Kamuli district which represents the typical RR value chain domain. The final districts selected are shown in Figure 3.7. A **Minimum validation** checklist was administered in villages across the selected districts between November and December 2012 (ILRI, 2012d). The objective was to validate if preselected sites fit into the defined value chain domains as assumed in the steps above, and to gather more information on sub-county and village data that could inform the finalization of the value chain assessment tools. The final sites selected are listed in Table 3.1. Table 3.2 outlines major characteristics of the selected districts.

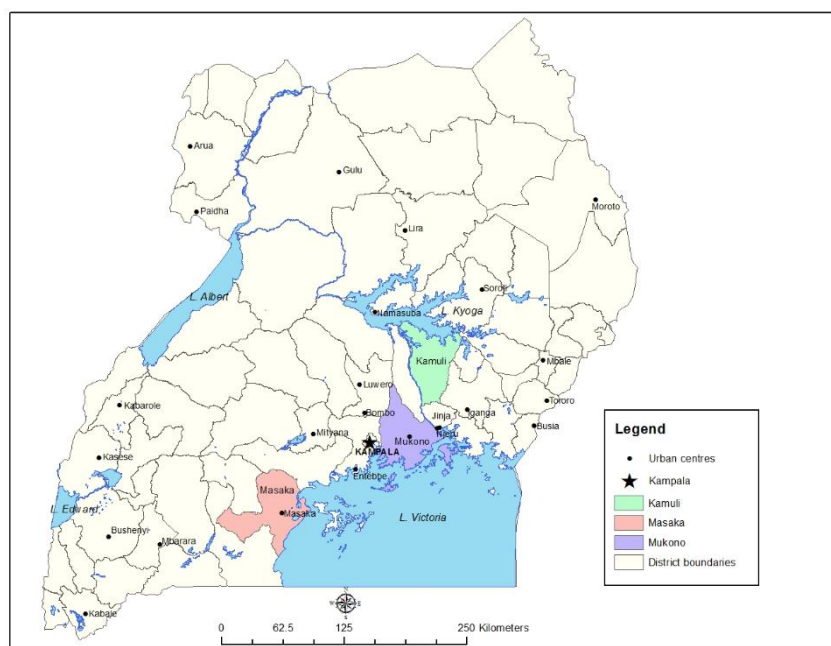


Figure 3.7 The CRP L&F study sites selected in 2012: Masaka and Mukono districts in the Central region; Kamuli district in the Eastern region of Uganda (ILRI/Pamela Ochungo).

Table 3.1 The final sites selected for the pig value chain assessment under the CRP L&F. Source: Poole et al. (2015).

District	Sub-county	Dominant value chain domain	No. of villages selected
<b>Masaka</b>	Kkingo	RR	3
	Kyanamukaka	RR	3
	Kabonera	RU	3
	Kimanya-Kyabakuza	UU	2
	Katwe-Butego	UU	2
	Nyendo-Ssenyange	UU	2
<b>Kamuli</b>	Kitayunwa	RR	2
	Namwendwa	RR	2
	Bugulumbya	RR	4
<b>Mukono</b>	Mukono town council	UU	2
	Goma	UU	2
	Kyampisi	RU	4
	Ntenjeru	RR	4

Table 3.2 Characteristics of the sites selected by the CRP L&amp;F. Sources: MAAIF/UBOS (2009); Twinomujini et al. (2011).

District	<b>Kamuli</b>	<b>Masaka</b>	<b>Mukono</b>
Region	Eastern	Central	Central
Area in sq km	1,154	2,196 <sup>7</sup>	4,125
Human population estimate (density per sq km)	281,800 (181)	239,700 (109)	498,500 (120)
Estimated number of pigs	55,989	236,150 (highest in Central region)	172,428
Livelihood activities	<p>Mainly agriculture-based:</p> <ul style="list-style-type: none"> <li>• Food crops: Soya bean, maize, sorghum, cassava, sweet potatoes, finger millet, rice, groundnuts, simsim (sesame) and sunflower.</li> <li>• Cash crops: Cotton, coffee and sugar cane.</li> <li>• Vegetables: Tomatoes, onions and cabbage.</li> </ul> <p>Industry:</p> <ul style="list-style-type: none"> <li>• Manufacture of jiggery, brick making, maize milling and cotton ginning.</li> </ul>	<p>Mainly agriculture-based:</p> <ul style="list-style-type: none"> <li>• Food crops: Sweet potatoes, bananas, beans, maize, cassava, groundnuts, soya bean, sorghum and finger millet.</li> <li>• Cash crops: Coffee, cotton and maize.</li> <li>• Fruits and vegetables: Pineapples, tomatoes, onions and cabbage.</li> <li>• Livestock farming.</li> <li>• Fishing on Lake Victoria.</li> </ul> <p>Industry:</p> <ul style="list-style-type: none"> <li>• Manufacture of cassava starch, curry powder, garments, animal feeds, soft drinks, footwear, laundry soap, metal work, furniture, jiggery, processing of tea, coffee, hand weaving, glass making and cotton ginning.</li> </ul>	<p>Mainly agriculture-based:</p> <ul style="list-style-type: none"> <li>• Food crops: Cassava, sweet potatoes, beans, maize, finger millet, groundnuts, soya bean, bananas, sorghum, simsim (sesame), cow peas, pigeon peas and yams.</li> <li>• Cash crops: Cotton, coffee, sugar cane and tea.</li> <li>• Fruits and vegetables: Tomatoes, onions, pineapples, vanilla, chillies, passion fruit and cabbage.</li> <li>• Dairy farming.</li> <li>• Fishing on Lake Victoria.</li> </ul> <p>Industry:</p> <ul style="list-style-type: none"> <li>• Processing of coffee, tea, cocoa, sugar; manufacture of textiles, animal feeds, beer, boats, furniture, metal products and grain milling.</li> </ul>

<sup>7</sup> While the map in Figure 3.7 represents the old district boundaries, numbers in Table 3.2 are based on the 2010 district reform whose implementation overlapped with the launch of the research.

#### 4 Parasites as a production constraint in smallholder pig production systems (Publications I & II)

This chapter characterizes the smallholder pig production systems in the selected study sites, documents husbandry practices and describes major constraints to pig health as perceived by the pig farmers (publication I). It also summarizes base line information on the prevalence of major gastrointestinal helminths and protozoa and factors correlating with increased presence (publication II).

The first publication shows that expensive feeds and pig disease were reported to be the major constraints to more productivity at farm as perceived by the farmers; disease causing 45-90% of losses across the sites. Major diseases described by the farmers were based on clinical signs visible to the farmers and included ASF and infestation with parasites, both gastrointestinal helminths and sarcoptic mange. Other diseases causing losses mainly due to skin alterations and weight loss were ectoparasites such as flies, lice, ticks and tungiasis (in Kamuli only). Piglet diarrhoea was also widely distributed. With regards to parasite control, the majority of the farmers (93%) stated to deworm their animals at least once, usually when the piglet enters the farm and a few days before it is sold. For treatment they either call a local veterinary professional or buy the drugs and administer them themselves. The most common anthelmintics used were albendazole and ivermectin. Spraying with acaricides is practiced by 37% of the farmers. Cross-cutting constraints to pig health at the production sites were lack of knowledge on pig management, especially on managing disease prevention and treatment; how to formulate suitable feed rations; limited access to animal health services; poor quality drugs and lack of capital to invest in improved housing.

Participatory methods were used for the assessments and included qualitative and semi-quantitative tools such as proportional piling, listing, ranking, scoring, seasonal calendars, as well as pair-wise comparison and problem-opportunity matrices. Two facilitators fluent in the local languages Luganda (spoken in Masaka and Mukono districts) and Lusoga (spoken in Kamuli district) were trained on the tools, and after pre-testing them with a group of farmers, they conducted the group sessions in all 35 selected villages. Pig health was only one component researched during the value chain assessments; other aspects covered were pig feeding and breeding, marketing as well as food safety and nutrition. These have been published separately.

The second publication reports findings from a cross-sectional survey in 21 of the 35 villages where the participatory value chain assessments were held approximately five months prior to the sampling. A random sample was taken between April and July 2013, and investigated the prevalence of common nematode and trematode eggs, and coccidian oocysts in faecal samples from 932 pigs by means of combined sedimentation-flotation method. The majority (61.4%) of all pigs were found infected with at least one species of gastrointestinal helminths: 57.1% strongyles, 7.6% *Metastrongylus* spp., 5.9% *Ascaris suum*, 4.2% *Strongyloides ransomi*, and 3.4% *Trichuris suis*. Coccidia oocysts were found in 40.7% of the sampled pigs, with significantly higher levels in the more urban Mukono district. None of the animals showed infection with *Balantidium coli* or *Fasciola hepatica*. A logistic regression model showed that routine management factors had a greater impact on the prevalence of infection than regular treatment or the level of confinement. Factors negatively correlating with gastrointestinal helminth infection were routine manure removal, and the routine use of disinfectants.

Full details of the scientific findings of this chapter have been subjected to peer review and published as original research papers in *Preventive Veterinary Medicine and Parasitology Research*. In compliance with the copyright transfer statement to Elsevier and Springer-Verlag Berlin Heidelberg, copies of the published articles appear overleaf. The final publications, together with the supplementary material published electronically, are available online:

Dione, M. M., Ouma, E.A., Roesel, K., Kungu, J., Lule, P., Pezo, D. 2014. Participatory assessment of animal health and husbandry practices in smallholder pig production systems in three high poverty districts in Uganda. *Prev Vet Med.* 2014 Dec 1;117(3-4):565-576. doi: 10.1016/j.prevetmed.2014.10.012. Erratum in: *Prev Vet Med.* 2015 May 1;119(3-4):239.

Roesel, K., Dohoo, I., Baumann, M., Dione, M., Grace, D., Clausen, P.-H. 2017. Prevalence and risk factors for gastrointestinal parasites in small-scale pig enterprises in Central and Eastern Uganda. *Parasitol Res.* [2016 Oct 26; Epub ahead of print] 116: 335-345. doi: 10.1007/s00436-016-5296-7

**Participatory assessment of animal health and husbandry practices in smallholder pig production systems in three high poverty districts in Uganda**

Michel M. Dione, Emily A. Ouma, Kristina Roesel, Joseph Kungu, Peter Lule, Danilo Pezo

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Corrigendum

**Corrigendum to “Participatory assessment of animal health and husbandry practices in smallholder pig production systems in three high poverty districts in Uganda”**

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<http://dx.doi.org/10.1016/j.prevetmed.2015.03.010>

## Prevalence and risk factors for gastrointestinal parasites in small-scale pig enterprises in Central and Eastern Uganda

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Michel Dione, Delia Grace, Peter-Henning Clausen

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<http://dx.doi.org/10.1007/s00436-016-5296-7>

## 5 Parasites with potential implications for public health (Publications III & IV)

This chapter presents the findings on the presence of two pig pathogens that are of global veterinary public health importance: *Trichinella* spp. and *Toxoplasma (T.) gondii*. These two parasitic infections have never been studied in pigs in Uganda, and we hope that our findings will inform the pig industry and the planning of future in-depth investigations. The prevalence data refers to the same cohort of pigs for which gastrointestinal parasite infection rates were presented in chapter 4.

The overall prevalence of *Trichinella*-specific antibodies was 6.9% out of 1,125 sampled animals but showed significantly higher levels in value chain types with a rural origin of production ( $p < 0.05$ ). Kamuli district, where the RR value chain type dominates showed a significantly higher ELISA-prevalence than the more urban Masaka and Mukono districts ( $p < 0.001$ ). For the first time, a study in pigs in sub-Saharan Africa was able to confirm 31 % ELISA-positive samples by Western Blot. Isolation of infective first-stage larvae from 499 samples in ELISA clusters to identify the dominating *Trichinella* spp. was not successful. Due to the large sample size, we are confident that even if domestic pigs are infected, the larval burden would be too low to pose a major risk to consumers for developing trichinellosis.

*T. gondii*-specific antibodies were found in 28.7% of the animals using a commercial ELISA kit; village herd prevalence was 100%. Significantly higher levels were shown for Masaka and Mukono districts compared to rural Kamuli districts; and univariate analysis showed that pigs raised in urban production settings (UU value chain types) showed significantly higher levels of seropositivity than animals of a rural production origin (RR and RU value chain types) ( $p < 0.007$ ).

Full details of the scientific findings of this chapter have been subjected to peer review, the work on *Trichinella* was published as original paper in PLOS ONE (publication III). In compliance with the copyright transfer statement to the Public Library of Science (PLOS), a copy of the article appears overleaf. The final publication, together with the supplementary material published electronically, is available online:

Roesel, K., Nöckler, K., Baumann, M.P.O., Fries, R., Dione, M.M., Clausen, P.-H., Grace, D. First Report of the Occurrence of *Trichinella*-Specific Antibodies in Domestic Pigs in Central and Eastern Uganda. 2016 Nov 21;11(11):e0166258. doi: 10.1371/journal.pone.0166258

The work on the occurrence of porcine *T. gondii* infections and related risk factors (publication IV) has been accepted for presentation to a peer audience by the scientific committee of the First Joint Conference of the Association of Institutions for Tropical Veterinary Medicine (AITVM) and the Society of Tropical Veterinary Medicine (STVM). An author-created version of the abstract appears overleaf and is available online:

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## RESEARCH ARTICLE

# First Report of the Occurrence of *Trichinella*-Specific Antibodies in Domestic Pigs in Central and Eastern Uganda

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**Data Availability Statement:** Data are available from Figshare: DOI:10.6084/m9.figshare.4219635; URL [https://figshare.com/articles/Trichinella\\_Uganda\\_data\\_for\\_PLOS\\_ONE\\_final\\_xls/4219635](https://figshare.com/articles/Trichinella_Uganda_data_for_PLOS_ONE_final_xls/4219635).

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## Abstract

Previous research on trichinellosis in Africa focused on isolating *Trichinella* from wildlife while the role of domestic pigs has remained highly under-researched. Pig keeping in Uganda is historically recent, and evidence on zoonotic pig diseases, including infection with *Trichinella* species, is scarce. A cross-sectional survey on *Trichinella* seroprevalence in pigs was conducted in three districts in Central and Eastern Uganda from April 2013 to January 2015. Serum from a random sample of 1125 pigs from 22 villages in Eastern and Central Uganda was examined to detect immunoglobulin G (IgG) against any *Trichinella* spp. using a commercially available ELISA based on excretory-secretory antigen. ELISA positive samples were confirmed using Western Blot based on somatic antigen of *Trichinella spiralis* as recommended in previous validation studies. Diaphragm pillar muscle samples (at least 5 g each) of 499 pigs from areas with high ELISA positivity were examined using the artificial digestion method. Overall, 78 of all 1125 animals (6.9%, 95% CI: 5.6–8.6%) tested positive for antibodies against *Trichinella* spp. in the ELISA at significantly higher levels in Kamuli district compared to Masaka and Mukono districts. Thirty-one percent of the ELISA positive samples were confirmed IgG positive by the Western Blot leading to an overall seroprevalence of 2.1% (95% CI: 1.4–3.2%). The large proportion of ELISA positive samples that could not be confirmed using Western blot may be the result of cross-reactivity with other gastrointestinal helminth infections or unknown host-specific immune response mechanisms in local pig breeds in Uganda. Attempts to isolate muscle larvae for species determination using the artificial digestion method were unsuccessful. Due to the large number of muscle samples examined we are confident that even if pigs are infected, the larval burden in pork is too low to pose a major risk to consumers of developing trichinellosis. This was the first large systematic field investigation of *Trichinella* infection in domestic pigs in Uganda

Development, Germany (grant no.: 81141843) in cooperation with the CGIAR Research Program on Livestock and Fish (<https://livestockfish.cgiar.org/>) through the Smallholder Pig Value Chains Development project in Uganda (funded by the International Fund for Agricultural Development – European Commission, grant no. COFIN-ECG-63-ILRI). We also acknowledge the CGIAR Fund Donors (<http://www.cgiar.org/who-we-are/cgiar-fund/fund-donors-2>). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

**Competing Interests:** The authors have declared that no competing interests exist.

and its results imply that further studies are needed to identify the *Trichinella* species involved, and to identify potential sources of infection for humans.

## Introduction

Human trichinellosis is acquired through the ingestion of the first-stage larvae of the nematode *Trichinella* in raw or undercooked meat from domestic animals, mainly pigs, and game. Clinically relevant infections can initially cause diarrhoea and abdominal pain; subsequently, migrating larvae and their metabolites may provoke fever, facial oedema and myalgia, or even fatal myocarditis. The severity of the symptoms may depend on the infection dose and the *Trichinella* spp. causing the infection [1].

The nematode *Trichinella* is cosmopolitan and found everywhere except the Antarctica. Eight species and four genotypes have so far been documented, five of them in Africa: *T. britovi* in North and West Africa, *Trichinella* T8 in Namibia and South Africa, *T. nelsoni* in Eastern and South Africa, *T. zimbabwensis* in Southern and Eastern Africa and *T. spiralis* in Egypt [2–5]. While a wide range of host species can be infected (mammals, birds and reptiles), only humans show clinical signs in case of infection [1]. While human infection is strictly related to the consumption of raw or undercooked infected meat [3], the major risk factors associated with infection in pigs are the ingestion of infectious first-stage larvae by scavenging, or feeding on scraps from pig slaughter or game hunting [6].

Most previous research on trichinellosis in Africa focused on isolating *Trichinella* from wildlife [7–13], and it was long believed that in Africa, *Trichinella* infection essentially affects wild carnivores [14] and that pigs are not infected with *Trichinella* in East Africa. Outbreaks of human trichinellosis in Africa date back several decades and were documented in Egypt [15], Northwest Ethiopia [16,17], Kenya [18] and Tanzania [19]. Other cases have been diagnosed in France in travellers returning from Senegal [20], a patient in Japan after travelling to Kenya [21], and travellers returning to the United States of America from Africa [22]. These cases of human trichinellosis have been mostly associated with the consumption of game meat, especially bush pig (*Potamochoerus* sp.) and warthog (*Phacochoerus* sp.). The prevalence in wildlife by regions and countries in Africa has been synthesized in recent reviews [5,8] but the real prevalence of human trichinellosis in Africa in general, and Uganda in particular, remains unknown.

Only two studies on *Trichinella*-specific antibodies in domestic pigs in sub-Saharan Africa have been published, both from Nigeria, and reporting seroprevalences of 40% (48/120) [23] and 11% (49/450) [24]. Attempts to directly detect muscle larvae from potentially naturally infected domestic pigs have been made in Northern Tanzania [25] and Ghana [26], but unsuccessfully. However, in Egypt, where *T. spiralis* had previously been documented, muscle larvae could be isolated by means of artificial digestion [15,27].

In sub-Saharan Africa, human population is growing rapidly and urbanization is increasing while most of the food is produced in the rural areas by smallholder farmers. Pig keeping has become a popular income-generating activity across Eastern Africa. However, pigs are not a traditional livestock species in the region and, in Uganda, evidence on zoonotic pig diseases has been scarce and limited to pigs as a reservoir for *Trypanosoma* species as well as the natural intermediate host for *Taenia solium* [28]. The presence of porcine *Trichinella* infection in Uganda was reported to the World Organisation for Animal Health (OIE) in 2008 but without any further provision of details [29].

The objectives of the present survey were to investigate if domestic pigs in Uganda are exposed to *Trichinella* by means of indirect methods in order to estimate if consumers are at risk of contracting trichinellosis through pork consumption.

## Materials and Methods

### Ethics statement

The research involved obtaining information from pig farmers and sampling of live animals as well as meat sampling from butcheries. Approval was obtained from the Research and Ethics Committee at the College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University, Kampala (Ref.: VAB/REC/13/103) and from the Uganda National Council for Science and Technology (Ref.: A 525). Informed consent was obtained from all individual participants included in the study.

### Study area

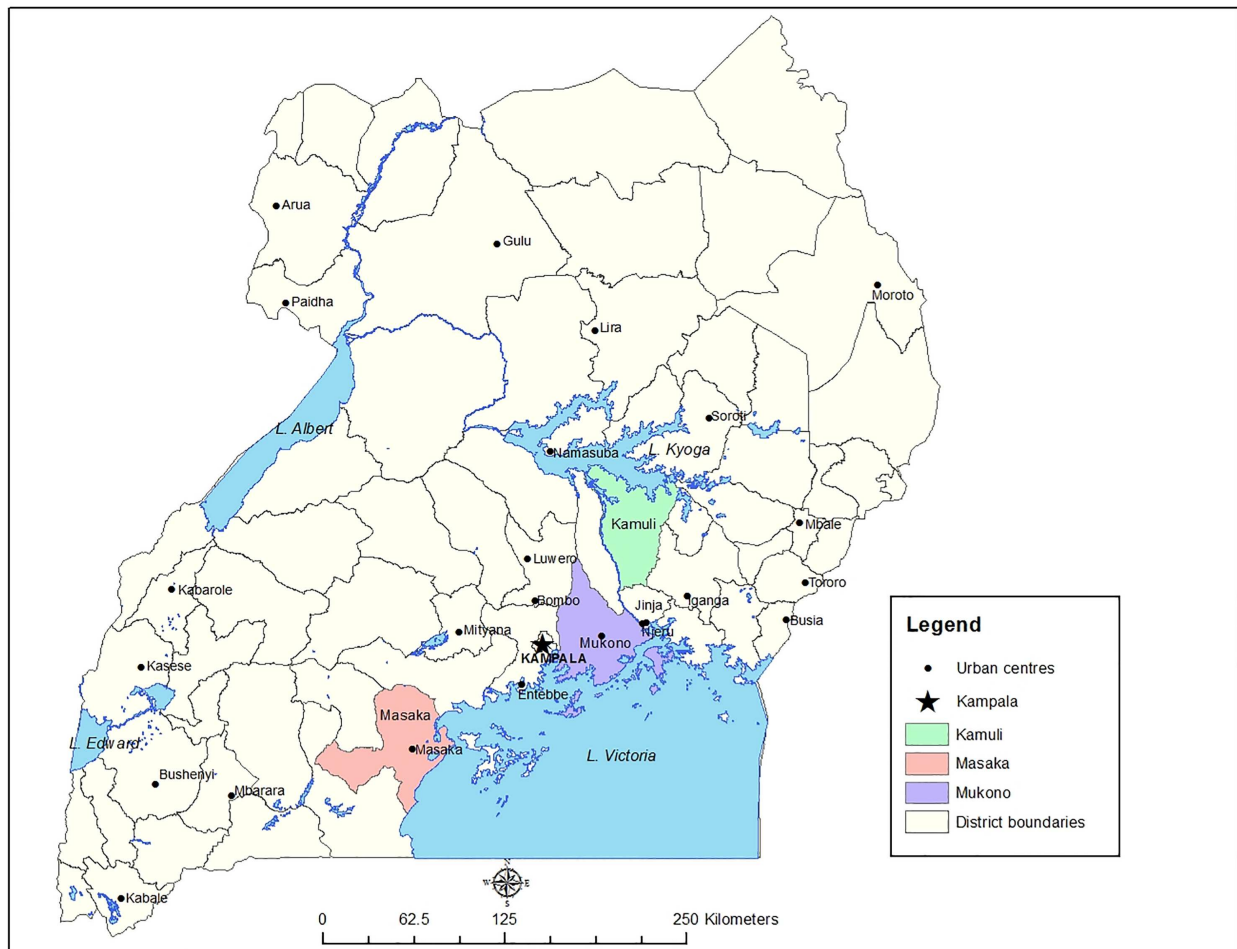
A cross-sectional survey was conducted in three districts in Central and Eastern Uganda from April 2013 to January 2015. Uganda is located between 4°N and 1°S of the equator in East Africa at the northern shores of Lake Victoria. Due to its proximity to the equator, the climate is tropical savannah in most parts of the country with abundant vegetation and rainfall. Despite its small size, the country's fauna is extremely diverse [30] and located in more than 60 protected areas, including ten national parks, and more than 500 forest reserves.

### Site selection

As part of a research for development project led by the International Livestock Research Institute (ILRI) and partners, Kamuli, Mukono and Masaka districts were selected with a range of stakeholders. Details of the process and criteria are described elsewhere [31,32]. Based on the 2008 livestock census [33], four to six sub-counties with a high pig population were purposively selected, and villages were further categorized into value chain domains by means of a checklist and consultation with local partners. Value chain domains describe the spatial dimensions of production and consumption of pigs: rural production for rural consumption (rural-rural, RR), rural production for urban consumption (rural-urban, RU) and urban production for urban consumption (urban-urban, UU) [31]. In each district, two sub-counties were purposively selected, and within each sub-county, two to three villages were randomly selected. From a total of 35 villages [31], 22 villages were purposively selected for the present prevalence study (Fig 1), based on available financial and logistic resources.

### Sample size calculation

The original sample size was calculated to estimate district-level prevalence and considering an infinite population using  $n = [Z^2P(1-P)]/d^2$ ; where  $n$  is the required sample size;  $Z$  is the multiplier from a standard normal distribution (1.96) at a probability level of 0.05;  $P$  is the estimated prevalence which is most conservatively estimated to be 50% considering there is no reference data from pigs in the area under study;  $(1-P)$  is the probability of having no disease and  $d$  is the desired precision level (5%). Therefore, a sample size of 384 pigs per district was required for the study. To increase precision, a sample size of 400 pigs in each district was planned.



**Fig 1.** Twenty-two villages were selected for a multi-pathogen survey in pigs in three districts of Central and Eastern Uganda, April to July 2013. (Pamela Ochungo/ ILRI)

doi:10.1371/journal.pone.0166258.g001

### Selection of the individual animals

In each of the 22 selected villages, a list of all households keeping pigs was generated in collaboration with the local government. From that list, households were randomly selected and invited to participate. Per household, one animal was selected for blood collection if it was aged three months or older, not weak or emaciated, not pregnant, and did not have a litter less than two months old.

Pigs were restrained using a snare and bled from the cranial *vena cava* using BD Vacutainer® needles and BD Vacutainer® plain tubes. The blood samples were kept standing in an ice box at 4°C to avoid haemolysis. At the field laboratory, blood was centrifuged and serum harvested into barcoded vials that were stored at -20°C until processed at Makerere University in Kampala, Uganda. Prior to shipment to Friedrich-Loeffler-Institute, Greifswald, Germany, the samples were heat-inactivated at 56°C for 20 minutes.

### Enzyme-linked immunosorbent assay (ELISA)

A commercially available ELISA based on excretory-secretory (E/S) antigen was used to detect immunoglobulin G (IgG) against any *Trichinella* spp. in pig sera (PrioCHECK® Trichinella Ab, Prionics AG, Wagensstrasse 27A, CH-8952 Schlieren). According to the manufacturer, the PrioCHECK® Trichinella Ab ELISA showed a sensitivity and specificity of 100% in an in-house validation study [34].

The assay was performed according to the manufacturer's instructions in a four-step protocol: serum was diluted 1:50 and incubated on plates coated with E/S antigen of *Trichinella spiralis*; a peroxidase-labelled anti-pig IgG was used as a secondary antibody bound to the antigen; a chromogen (TMB) substrate was added and the optical density of the sample was measured by reading the test plate at 450 nm wavelength (Sunrise™ Tecan, powered by Magellan™ software). The results were calculated in percent positivity (PP) in reference to the positive control sample. Results at or above the manufacturer's cut-off of 15% PP were considered ELISA positive.

### Confirmatory testing

It is recommended that ELISA positive samples are tested by Western Blot for confirmation of the presence of IgG antibodies against *Trichinella* spp. [35,36]. While the ELISA was based on E/S antigen, the Western Blot used for confirmatory testing in this study was based on somatic antigen from crude worm extract (CWE) of *Trichinella spiralis* first-stage larvae [37]. In addition to all relevant protein fractions of the E/S antigen (43–45 and 66–67 kDa), it includes fractions 47, 61, and 102 kDa which are *Trichinella*-specific [36]. In validation studies evaluating CWE- against E/S-Western Blot and using artificial digestion as 'gold standard', the CWE--Western Blot showed a sensitivity ranging from 95.8–91.1% and a specificity ranging from 99.5–100% [35,36]. In this study, it was performed on all ELISA positive samples, and a randomly selected subset of ELISA negative samples (n = 16) for internal quality control. Sera that showed an antibody reaction to any of the five immunogenic protein bands mentioned above were considered Western Blot positive.

### Artificial digestion

In an attempt to isolate *Trichinella* muscle larvae for species determination, 499 pig diaphragm samples from four clusters with a high antibody prevalence based on the ELISA test were collected from October 2014 to January 2015. Diaphragm muscle is the major predilection site for *Trichinella* spp. in domestic swine [38,39]. Artificial digestion according to OIE instructions [40] was carried out at the Central Diagnostic Laboratory at the College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University in Kampala, Uganda. A positive control of *Trichinella spiralis* was provided by the National Reference Laboratory for *Trichinella* hosted by the Federal Institute for Risk Assessment in Berlin, Germany.

Muscle samples of at least 5 g per animal were tested to increase sensitivity of the artificial digestion method [37,39,41]; 102 samples weighed 5 g; 396 samples 10 g and one sample 20 g. Up to 20 samples with a total weight of 100 g were examined in a pool by artificial digestion method [40].

### Household survey data

Using a structured questionnaire, information on self-reported husbandry and consumption practices was collected at each household from where a pig was sampled.



## Data management

ELISA results were exported to Microsoft Excel, Version 2010, from the ELISA reader using Magellan™ software. Laboratory data from Western Blot and artificial digestion were entered into Microsoft Excel, Version 2010. Household data were entered using Census and Survey Processing System, Version 4.1. (U.S. Census Bureau), and subsequently exported to MySQL for data validation. Datasets were merged for cleaning and descriptive analysis in STATA 13.1 (StataCorp).

## Results

### Antibody prevalence

Overall, 78 of 1125 animals (6.9%, 95% CI: 5.6–8.6%) tested positive for antibodies against *Trichinella* spp. in the ELISA (Table 1), with significant differences across districts ( $p < 0.001$ ), sub-counties ( $p < 0.05$ ) and value chain types ( $p = 0.05$ ) (Fig 2). Most ELISA positive samples were found in Kamuli district, which is dominated by an RR value chain type, compared to Mukono and Masaka districts.

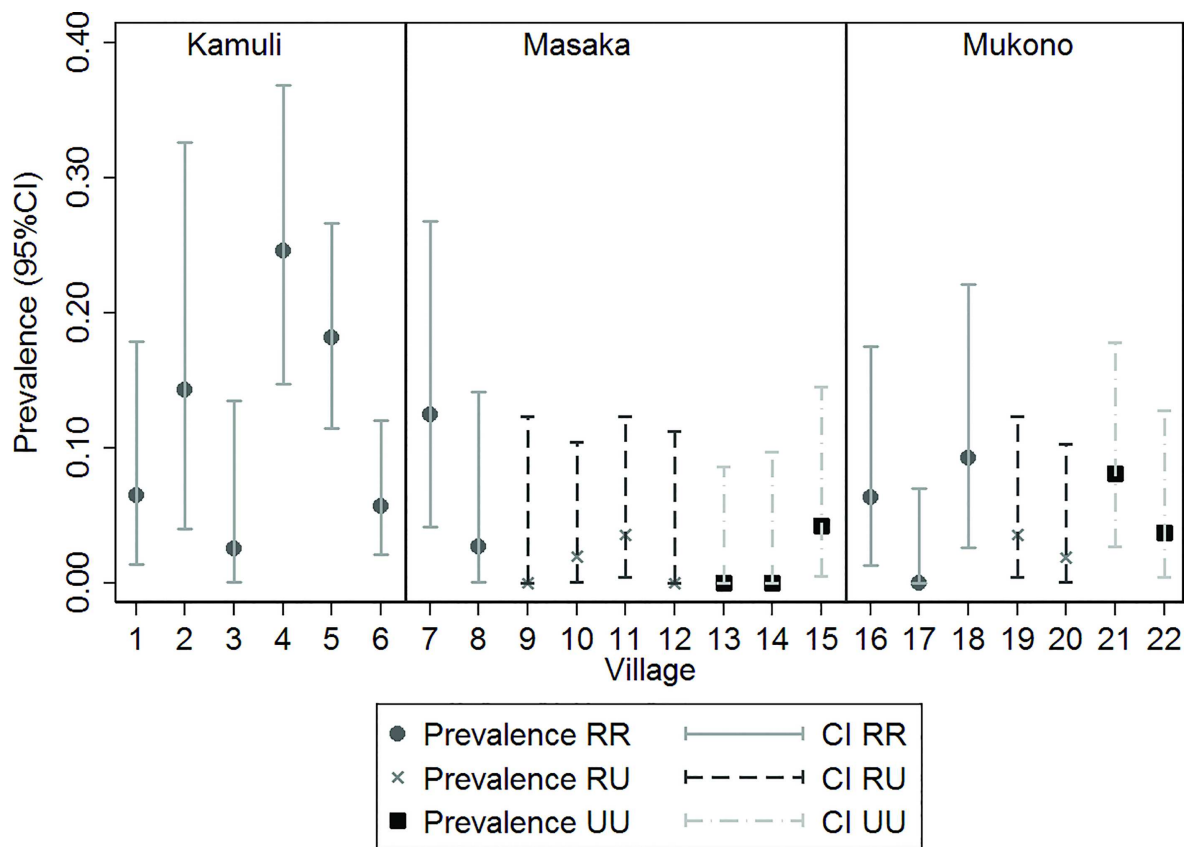
**Table 1. Prevalence estimates for anti-*Trichinella* IgG (PrioCHECK® *Trichinella* Ab) in three districts in Central and Eastern Uganda, sampled between April and July 2013.**

District	Village	Value chain type <sup>a</sup>	Sample size (n)	ELISA+	Prevalence estimate <sup>b</sup> (confidence limits)	
Kamuli	(1) Baluboinewa	RR	46	3	6.5% (2.1–18.6%)	
	(2) Bukyonza B	RR	28	4	14.3% (5.4–32.9%)	
	(3) Kantu zone	RR	110	20	18.2% (12.0–26.6%)	
	(4) Ntansi	RR	105	6	5.7% (2.6–12.2%)	
	(5) Butabala	RR	39	1	2.6% (0.4–16.5%)	
	(6) Isingo A	RR	65	16	24.6% (15.6–36.6%)	
<b>Total</b>			<b>393</b>	<b>50</b>	<b>12.7% (9.8–16.4%)</b>	
Masaka	(7) Kyamuyimbwa-Kikalala	RU	31	0	0%	
	(8) Butego	RU	28	0	0%	
	(9) Kijjabwemi	UU	41	0	0%	
	(10) Kyabakuza B	UU	36	0	0%	
	(11) Kisoso	RU	56	2	3.6% (0.9–13.4%)	
	(12) Ssenya	RR	37	1	2.7% (0.4–17.3%)	
	(13) Kanoni-Bukunda	RU	51	1	2.0% (0.3–12.9%)	
	(14) Lukindu	RR	40	5	12.5% (5.2–27.0%)	
	(15) Ssenyange A	UU	47	2	4.3% (1.1–15.7%)	
	<b>Total</b>			<b>367</b>	<b>11</b>	<b>3.0% (1.7–5.3%)</b>
	Mukono	(16) Dundu	RU	56	2	3.6% (0.9–13.4%)
		(17) Kyoga	RU	52	1	1.9% (2.7–12.7%)
		(18) Joggo	RR	62	5	8.1% (3.4–18.1%)
		(19) Kitete	RR	54	2	3.7% (0.9–13.8%)
		(20) Bugoye-Kabira	RR	47	3	6.4% (2.1–18.2%)
(21) Kazo-Kalagala		RR	51	0	0%	
(22) Nsanja-Gonve		RR	43	4	9.3% (3.5–22.5%)	
<b>Total</b>				<b>365</b>	<b>17</b>	<b>4.7% (2.9–7.4%)</b>
<b>Grand Total</b>			<b>1125</b>	<b>78</b>	<b>6.9% (5.6–8.6%)</b>	

<sup>a</sup>Spatial dimensions of production and consumption of pigs [31]: rural production for rural consumption (RR); rural production for urban consumption (RU); urban production for urban consumption (UU)

<sup>b</sup>Calculated at  $p = 0.05$  and  $CI = 0.95$

doi:10.1371/journal.pone.0166258.t001



**Fig 2. Prevalence estimates and confidence levels for anti-*Trichinella* IgG (PrioCHECK *Trichinella* Ab) in three districts in Central and Eastern Uganda, sampled between April and July 2013.**

doi:10.1371/journal.pone.0166258.g002

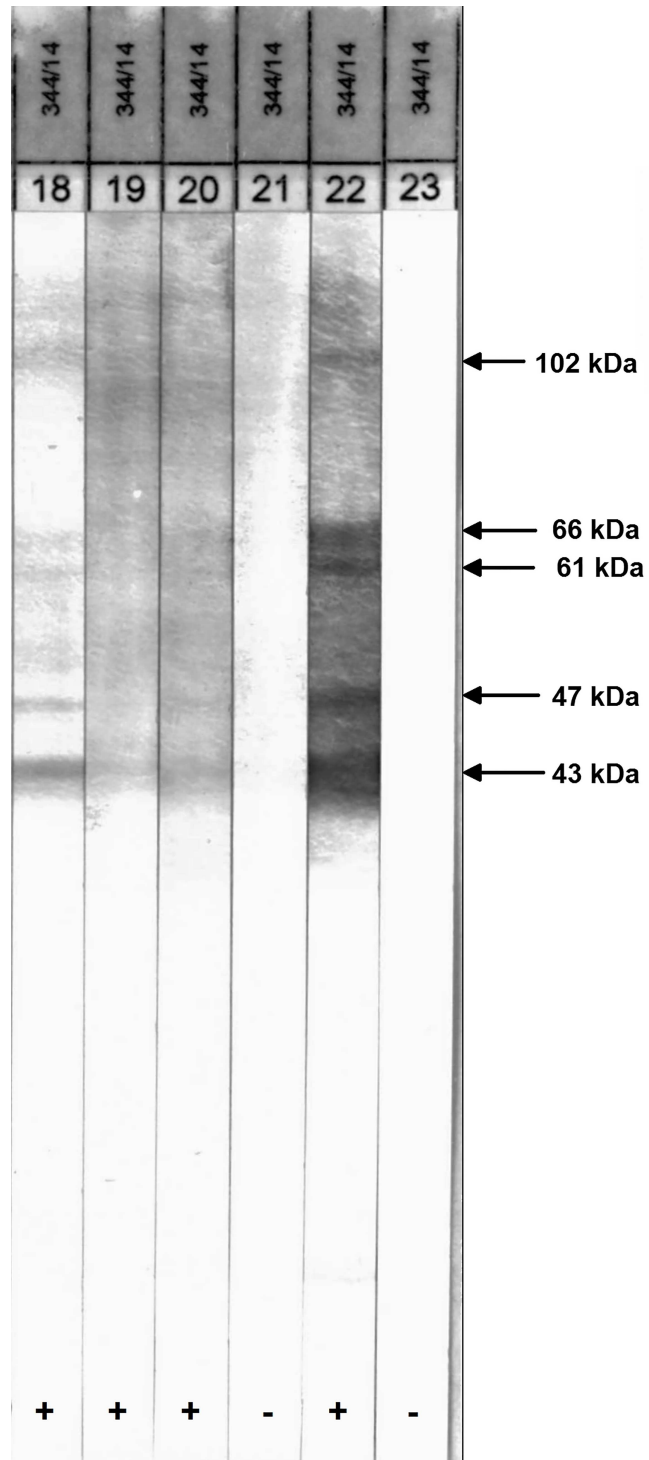
Twenty-four (30.8%) of the 78 ELISA positive samples were confirmed IgG positive by the Western Blot leading to an overall seroprevalence of 2.1% (95% CI: 1.4–3.2%) (Fig 3 and Table 2). There were no significant differences in the Western Blot prevalence between districts (Fig 4). However, the proportion of confirmed samples showed a reverse order with most ELISA positive samples confirmed in Mukono district (47%), followed by Masaka (36%) and Kamuli (24%) districts. All ELISA negative samples used for internal quality control tested negative in the Western Blot.

### Artificial digestion

A total of 499 diaphragm samples were collected from 32 butcheries, representing all butcheries operating in the four sub-counties with the highest ELISA prevalence at the time of sampling. *Trichinella* muscle larvae were not detected in any of the samples.

### Descriptive analysis

Two-thirds of the households participating in the survey were male-headed (67.0%) with a more equal ratio (60:40) in Masaka district. The majority of the households in the study were



**Fig 3. Four of the 78 ELISA positive samples tested by Western Blot based on somatic antigen (strip 18–21).** Strip 22 represents the positive control and strip 23 the negative control.

doi:10.1371/journal.pone.0166258.g003

members of the Christian faith (94.0%) and four pig keeping households (0.4%) in Masaka and Mukono districts were headed by Muslims. In all districts, the most commonly found herd size was two to five pigs (39.4% of households); households keeping only one pig were more common in rural Kamuli (28.5%), while in Masaka and Mukono districts, keeping six to ten pigs was more frequent. The most commonly reared breeds (58.3%) were crosses between the so-called local (black) and exotic (i.e. Large White, Camborough) breeds.

Most of the animals (76.7%) were raised in rural settings. According to the ILRI value chain classification, 30.5% of the pigs kept in rural areas were destined for sale at urban markets (RU) while the rest were intended to be marketed locally (RR). More than half of the households kept their pigs tethered or confined (53.5% and 52.7%, respectively), and only a small fraction left their pigs roam free (2.1%). Tethering was the preferred method of restricting the movement of pigs in Kamuli and Mukono districts, while in Masaka almost three-quarters of pig farmers kept their pigs confined. Overall, all pig farming households fed mixed pig rations, utilizing crop residues (98.3%) and commercial feeds (70.9%), while 35.0% practised swill feeding (e.g. kitchen or restaurant leftovers and wasted bread). More than half of the pig farming households (52.9%) performed routine pest/rodent control.

Direct and indirect contact with wild animals was proxied by reports of wildlife sightings in the village (34.9% of pig farmers). However, only 4.1% of pig farmers reported that they consumed game meat. Those who did ate meat from antelopes such as Sitatunga (*Tragelaphus spekkii*) or Ugandan Kob (*Kobus kob thomasi*), but also rodents such as cane rats (*Thryonomys* spp., locally referred to as “edible rats”), or *Canidae* including stray dogs (*Canis familiaris*) and civet cats (*Viverridae*) as well as wild pigs (*Potamochoerus* spp.), buffaloes (*Syncerus caffer*), hares (*Leporidae*) and guinea fowls (*Numididae*).

Slaughtering pigs at home was more frequent in Masaka and Mukono districts. Overall, only 12.6% of pig farmers slaughtered at home at least once a year; the majority (71.2%) never slaughtered their own pigs (Table 3). Of those farmers who slaughtered at home, more than three-quarters reported that the meat had never been inspected. At slaughter, viscera and intestinal contents were usually buried, thrown into the bush or given to dogs. Most of the pig farmers in the study area were also pork consumers and ate pig meat at least once a month (57.8%), while 21.7% never consumed pork and 15.4% ate it only occasionally. The most frequent mode of processing the meat was frying (62.9%) or boiling (37.7%) (Table 3).

## Discussion

The present survey is one of the few in sub-Saharan Africa and the first in Uganda to investigate the occurrence of infection with *Trichinella* spp. in domestic pigs. To the authors’

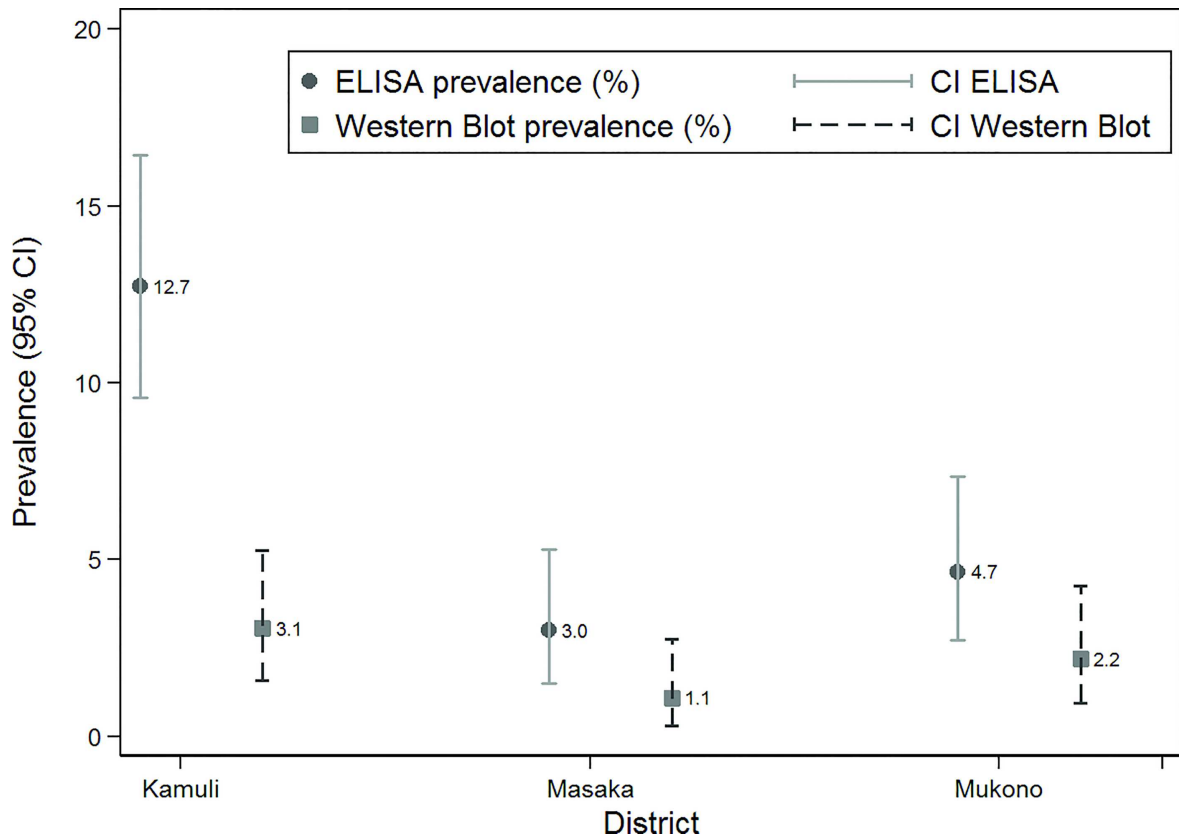
**Table 2. Western Blot (WB) confirmation of ELISA positive samples (anti-*Trichinella* IgG), collected between April and July 2013 in 22 villages from three districts in Central and Eastern Uganda.**

District	Sample size (n)	ELISA+	WB-	WB+	Total confirmed (%)	WB prevalence estimate <sup>a</sup> (confidence limits)
Kamuli	393	50	38	12	24.0	3.1% (1.7–5.3%)
Masaka	367	11	7	4	36.4	1.1% (0.4–2.9%)
Mukono	365	17	9	8	47.1	2.2% (1.1–4.3%)
Total	1125	78	54	24	30.8	2.1% (1.4–3.2%)

WB: Western Blot

<sup>a</sup>Calculated at  $p = 0.05$  and  $CI = 0.95$

doi:10.1371/journal.pone.0166258.t002



**Fig 4. Proportion of ELISA positive samples confirmed by Western Blot based on somatic antigen of *Trichinella spiralis* in three districts in Central and Eastern Uganda, sampled between April and July 2013.**

doi:10.1371/journal.pone.0166258.g004

knowledge, it is also the first investigation in Africa confirming ELISA positive results by Western Blot that resulted in an overall seroprevalence of 2.1%.

Two farm surveys in Nigeria found a prevalence of *Trichinella* IgG of 10.9% [24] and 40% [23]. Both surveys used the same commercial ELISA based on E/S antigens as in the present survey but reported higher prevalence rates. In previous studies, this ELISA detected antibodies in samples with larval densities of 0.025 larvae per gram (LPG) [42]. The authors of those validation studies were also able to detect antibodies in pigs infected with different *Trichinella* species, like *T. spiralis*, *T. britovi* and *T. pseudospiralis*, but showed no cross-reactivity with other pig pathogens such as *Trichuris suis*, *Oesophagostomum*, *Strongyloides*, *Hyostromylus*, *Oesophagostomum/Hyostromylus* and *Salmonella typhimurium* [34]. *T. nelsoni*, a *Trichinella* species previously reported in game animals in Eastern Africa, has not been included in those validation studies.

ELISAs of the preceding generation were based on somatic antigen of *Trichinella spiralis*. Since then, the specificity of the ELISA has been improved by utilizing E/S antigens obtained from metabolites of *Trichinella* muscle larvae incubated *in vitro* [37]. While the manufacturer of the commercial kit used in this study reports a sensitivity and specificity of 100% [34], a comparable in-house ELISA for the detection of *Trichinella*-specific antibodies had a test

**Table 3. Pig slaughter, processing and consumption practices in Central and Eastern Uganda (2013).**

Variable	Totals n (%)
<i>Frequency of home slaughter</i>	
≥ Once a month	26 (2.74%)
≥ Once a year	71 (7.49%)
≤ Once a year	22 (2.32%)
Never	675 (71.20%)
Cannot remember/don't know	9 (0.95%)
Missing	145 (15.30%)
Total	948 (100.00%)
<i>Frequency of meat inspection at home slaughter (if practiced)</i>	
Always	7 (5.88%)
Sometimes	9 (7.56%)
Cannot remember/don't know	2 (1.68%)
Never	93 (76.15%)
Missing	8 (6.72%)
Total	119 (100.00%)
<i>Disposal of viscera &amp; intestinal contents</i>	
Throw away outside compound	17 (1.79%)
Throw away inside compound	2 (0.21%)
Manure	0
Feed the live pigs	0
Other: buried	28 (2.95%)
Other: given to dogs	12 (1.27%)
Other: given to people	2 (0.21%)
Other: not specified	81 (8.54%)
Missing	806 (85.02%)
Total	948 (100.00%)
<i>Frequency of pork consumption</i>	
≥ Once a month	548 (57.81%)
≥ Once a year	146 (15.40%)
≤ Once a year	32 (3.38%)
Never	206 (21.73%)
Missing	16 (1.69%)
Total	948 (100.00%)
<i>Pork preparation</i>	
Boiling	357/948 (37.66%)
Frying	596/948 (62.87%)
Barbeque	95/948 (10.02%)
Other	7/948 (0.74%)
Missing	213/948 (22.47%)
<i>Consumption of game meat</i>	
	39/948 (4.11%)

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sensitivity and specificity of 96.8% and 97.9%, respectively [36]. As shown in other studies, unspecific cross-reactions may not be fully excluded; therefore, a Western Blot was used for confirmatory testing. Reportedly, the combined use of both serological methods shows a sensitivity that is 31.4 times higher than that of the digestion assay [43], especially in naturally infected wild pigs with a lower larval burden. Western Blot based on somatic antigen allows

discriminating ELISA positive samples further because it includes all relevant protein fractions of the E/S antigen (43–45 and 66–67 kDa) and fractions 47, 61 and 102 kDa which are *Trichinella*-specific [35,36]. The obvious discrepancy between ELISA positives and Western Blot positives may be the result of cross-reactivity with other gastrointestinal helminth infections as found in this study for co-infection with strongyle species (data not shown). It could also be caused by unknown host-specific immune response mechanisms in local pig breeds from Uganda because the validation studies involved European pig breeds.

We were not able to determine the *Trichinella* species infecting domestic pigs in the investigated study areas of Uganda as artificial digestion of a large number of muscle samples did not result in the isolation of muscle larvae. Infectivity and larval burden in domestic pigs largely depends on the *Trichinella* species. While *T. spiralis* has a very high reproductive capacity resulting in 427.4 LPG five weeks p.i., *T. nelsoni* and *T. britovi* result in a larval burden of 52.2 and 38.0 LPG, respectively [44]. This may explain why potentially low larval burdens of a non-*T. spiralis* infestation in the muscle could not be detected by artificial digestion. For future studies, testing larger amounts (up to 50–100g) of muscle tissue may compensate for a possible decrease in sensitivity [40] due to unknown larval burden in predilection sites. In addition, future sampling studies to detect larvae should concentrate in areas where seroprevalence was highest by Western Blot. This was not applicable in this study due to logistics and timing of sampling and testing.

While the Nigerian survey by Momoh et al. [23] reported potential risk factors, we deliberately waived statistical risk factor analysis in our study. We performed Western Blot as a confirmatory test which reduced the number of positive samples by 69% and left us with too few positives to conduct sound univariate analysis. Confirmation by Western Blot has not been done in previous studies in domestic pigs in Africa where ELISA positivity was used as the outcome variable and the number of observations was much higher. In the present study, the outcome variable would ideally be the result of the Western Blot. In this study, 78 samples tested ELISA positive and 24 were confirmed as Western Blot positive; 1047 samples tested ELISA negative but an unknown number could be Western Blot positive. Since we had only sufficient funding to carry out Western Blot on a subset of 16 ELISA negative samples, and although all were negative in Western Blot, it is not possible to definitively identify which (if any) of the untested 1031 ELISA negative samples would have tested positive or negative in Western Blot. Thus, conducting a risk factor analysis on the small subset of ELISA positive samples was not considered meaningful as the relation between ELISA positive, Western Blot positive, ELISA negative and Western Blot negative samples was not known, and risk factor analysis would then only apply to a small subset of pigs of unknown epidemiological status.

From the information gathered in this survey, potential reservoirs for *Trichinella* sp. such as rodents and stray dogs should be investigated in the future. They may act as potential links between the sylvatic and domestic life cycle as shown by researchers in Egypt who isolated muscle larvae from dogs and rodents in urban settings and in the proximity of abattoirs [45,46].

Information gathered on slaughter and consumption practices are indicative but not conclusive. However, previous reports in the same geographical area suggest that pig farmers do not consume raw pork but thoroughly heat meat products [47]. Results on slaughter practices, for instance, disposal of remains, or the consumption of game meat, are likely to be underreported by the participants of the present survey. The frequency of wildlife sightings, too, may be underreported as wildlife is very common in some areas and often only subconsciously noticed as it is such an integral part of daily life.

Previous reviews suggest that *Trichinella* infection in pigs and people plays a negligible role in Africa because the people of sub-Saharan Africa consist mainly of Bantu origin who do not

consume meat [6,48]. Personal observation of an increasing number of roasted meat outlets (both pork and beef) and Food and Agriculture Organization of the United Nations statistics on per capita consumption of meat [49] strongly disagree with this argument. In Uganda, pig numbers increased more than tenfold from 200,000 to more than 3 million since the 1980s [33], while per capita consumption is highest in Eastern Africa at 3.4 kg [49] at increasing trends. In general, the history of the domestic pig in Africa is highly controversial and has been heavily neglected in research [50]. Uganda in particular is a very diverse country with Muslims and Christians intermarrying and a substantial inflow of migrants, especially from China and India [51], who traditionally consume a lot of pork and may contribute to an increased demand in the near future.

In conclusion, we were able to demonstrate that domestic pigs in Kamuli, Masaka and Mukono districts in Uganda have been infected with *Trichinella* spp. This was the first large systematic investigation of *Trichinella* infection in domestic pigs in Uganda, potentially identifying high-risk areas through the confirmation of ELISA positive samples using Western Blot. We were not able to identify the species; however, this was not the objective of the study. Due to the large number of diaphragm muscle samples examined at higher sample weight (at least 5 g), we are confident that even if pigs are infected, the larval burden in pork is too low to pose a major risk to consumers of developing trichinellosis. Further studies are needed to identify the *Trichinella* species infecting domestic pigs and to identify potential sources of infection and modes of transmission.

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## **The occurrence of porcine *Toxoplasma gondii* infections in smallholder production systems in Central and Eastern Uganda**

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### **Abstract**

Pig production is an emerging agribusiness in Eastern Africa but baseline information on pig diseases including zoonoses is still scarce. Infection with *Toxoplasma gondii* does not usually present with clinical signs in pigs, yet it is considered an important source of human infection when pork containing tissue cysts is poorly handled or consumed raw or undercooked.

In a cross-sectional survey between April and July 2013, we sampled 932 pigs between three months to three years of age in 22 villages at smallholder farms. The sera were tested for the presence of antibodies to *T. gondii* using a commercial ELISA (PrioCHECK Toxoplasma Ab porcine) and an in-house assay (TgSAG1 p30). The overall seroprevalence based on the commercial ELISA was 28.7% (95% CI: 25.8-31.7%). Seropositive animals were found in all villages with significant differences across the three districts ( $P < 0.05$ ) and 12 sub-counties ( $P < 0.01$ ) in the survey area. Cohen's kappa statistic showed a very good level of agreement ( $\kappa = 0.7637$ ) between the two serological assays.

Preliminary univariate analysis suggests a significant association between seropositivity and pig age, value chain type, feeding of crop residues, source of drinking water, keeping cats on farm compound, and frequent sightings of wildlife (especially antelopes, hares, wild and stray dogs) near the village. The present report is the first survey documenting the seroprevalence of *T. gondii* in domestic pigs in the East African Community (Burundi, Kenya, Rwanda, Tanzania and Uganda) and investigating potential risk factors that may need attention when promoting smallholder pig keeping as a livelihood activity in Central and Eastern Uganda. The research was carried out with the financial support of the Federal Ministry for Economic Cooperation and Development (BMZ/GIZ), Germany, the CRP A4NH, led by IFPRI through the SFFF project at ILRI as well as the CRP L&F at ILRI as part of the SPVCD project.

## **6 Knowledge, attitudes and practices of pork consumers (Publication V)**

It is not possible to assess livestock value chains comprehensively without considering the consumers of the principal output, in this case the meat. Not only is the consumers' demand for animal sourced food a main driver of livestock production, and secondly, if the edible end product is infected with pathogens it is the consumer whose health may suffer. On the other hand, if the consumer demands certain quality standards he or she can be critical in helping to improve the entire value chain. We therefore wanted to find out what the attitudes of pork consumers are towards the quality of meat and pork in particular, what they believe and know about pig and pork zoonoses, how pork is usually prepared and how frequently it is consumed. Due to time and financial restrictions we were not able to develop a separate sampling frame representative of all pork consumers in the selected sites; therefore, we started by studying the knowledge, attitudes and practices of the smallholder pig producers who are potentially pork consumers.

Tools from participatory epidemiology (PE) were used to discuss questions such as: Who eats pork, when and why? What are reasons not to eat pork? What is the role of pork in farmers' diets? Are pig keepers pork eaters? How accessible is pork? Do pig feeds compete with human food? How does knowledge, attitudes and practices increase or reduce the risk of pork-borne diseases?

Some of the key findings include that pork is widely popular among pig farmers, in rural areas mostly during seasonal festivities and whenever cash is available, in urban areas much more frequently. Pig keepers are pork eaters; however, they are not usually consuming their own animals but buy pork at local (and informal) butcheries and pork joints. Reasons for not eating pork are mainly religious and traditional beliefs, or sometimes taste preferences. According to pig farmers, pig feed production does not compete with human food production; some festivals when pork is consumed even coincide with times when food is not abundant. The consumption of raw pork is considered unsafe, and at home meat is cooked for at least one hour.

Full details of the scientific findings of this chapter have been subjected to peer review and presented as a conference paper (publication V) at the 6<sup>th</sup> All African Conference on Animal Agriculture held in Nairobi, Kenya, from 27-30 October 2014. An author-created version of the published article appears overleaf. The abstract is available online:

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**Smallholder pig producers and their pork consumption practices in Kamuli, Masaka and Mukono districts in Uganda**

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**Abstract:**

Pig production is thriving in Uganda and the demand for pork is increasing, therefore offering potential for increased income through small-scale pig production and marketing. A multi-disciplinary value chain assessment conducted by the International Livestock Research Institute and partners aimed to identify constraints and opportunities for value chain actors as well as shortcomings in the safety of pork products in three districts in Uganda. Prior to quantitative surveys and biological sampling at various nodes of the chain, participatory rural appraisals and focus group discussions were held with about 1,400 smallholder pig farmers to map out qualitative aspects including the various actors involved in pig rearing (e.g. input and service providers) and marketing (e.g. marketing channels and pricing). One particular aspect covered pork consumption habits as well as knowledge, attitudes and practices on pork safety among 294 participants, considering that they are also food consumers. Pork is widely popular, mostly consumed well-cooked or thoroughly pan-fried. The frequency of consumption follows seasonal patterns and increases from rural to more urban settings. Practices such as roasting may lead to the ingestion of undercooked pork, and accompanying dishes such as raw vegetables may lead to cross-contamination of the meat causing foodborne diseases. The scarcity of data on zoonotic pig pathogens, such as erysipelas, salmonellosis, brucellosis and pork-borne parasites requires further research.

**Key words:** participatory research, pork safety, quality attributes, risk, women



In the session on food safety and nutrition, qualitative and semi-quantitative data on pork consumption patterns, preparation methods as well knowledge, attitudes and practices on pork safety was gathered from 294 randomly selected pig farmers (103 men and 191 women) in 34 villages in the above mentioned districts. Generic discussion guides were used with all groups and included tools from Participatory Epidemiology such as focus group discussions, ranking and scoring methods, Venn diagrams and seasonal calendars. These activities were used to answer a set of research questions, specifically: Who eats pork, when and why? What are reasons not to eat pork? What is the role of pork in farmers' diets? Are pig keepers pork eaters? How accessible is pork? Do pig feeds compete with human food? How does knowledge, attitudes and practices increase or reduce the risk of pork-borne diseases?

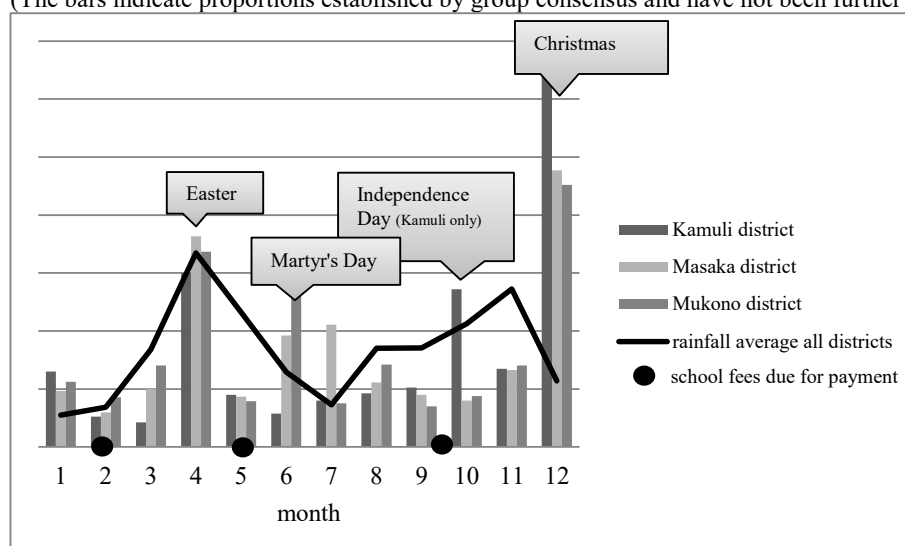
Table 1. Pork consumption assessment tools used by district.

Tools used	PRA guide producers	PRA guide consumers	FGD with mothers
Kamuli district	4	4	5
Masaka district	14	0	14
Mukono district	6	6	8
<b>Total</b>	<b>24</b>	<b>10</b>	<b>27</b>

The data was entered into MS Excel for basic descriptive analysis and visualization.

**Results:** Pig producers are generally pork eaters; 80% of the pig farmers in the survey (n=234/294) eat pork, whereas the proportion of male consumers is marginally higher (89%) than the female (74%). Consumption is mainly driven by festivals as shown in Figure 2. More pork is consumed when cash is available, for instance after the coffee harvest in Masaka during June/July. Less pork is consumed at the beginning of new school terms when pigs are sold to pay for school fees.

Figure 2. Seasonality of pork consumption among 292 pig farmers in Kamuli, Masaka and Mukono districts. (The bars indicate proportions established by group consensus and have not been further quantified.)



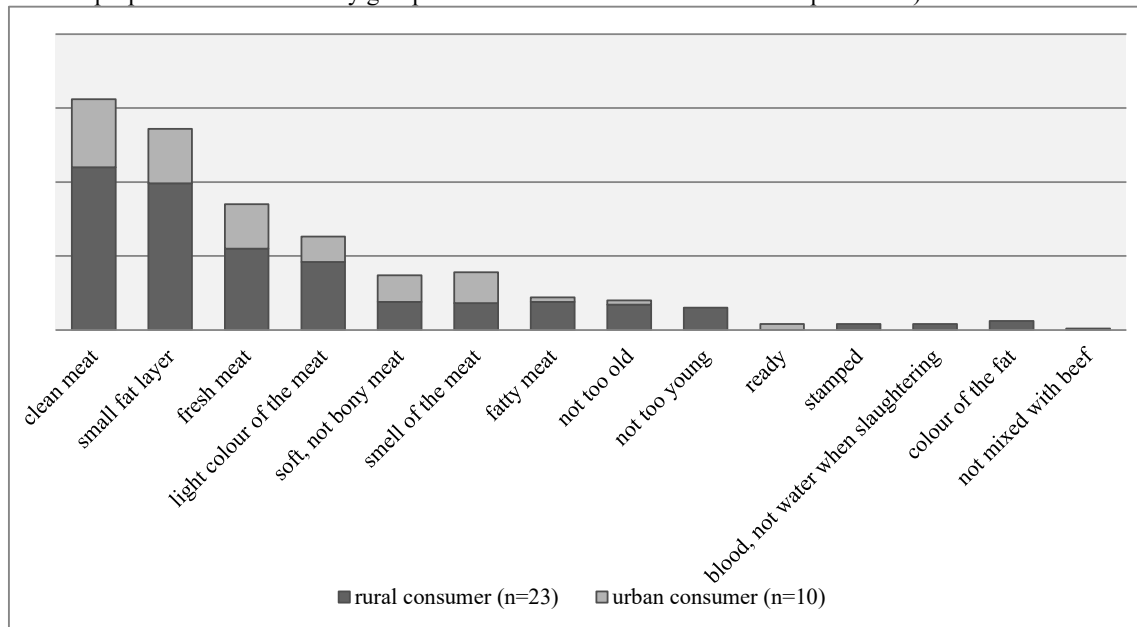
Pork ranks second after chicken in terms of taste and is occasionally given to children under five years as food for good growth. In rural sites, pork is believed to clear the skin, cure



measles and make “strong bones” whereas in the urban areas pork is sometimes believed to cure HIV/AIDS (Ejobi et al, forthcoming). In the rural study sites, pig farmers rarely slaughter their own animals as they use the money generated from sales of live pigs for meeting family needs such as school fees. The closer to urban centres, the more frequently pork and other animal sourced food is eaten, e.g. weekly to daily, and pigs are kept for both sale and home consumption. The biggest constraint for eating more pork in rural areas is low income; other factors include religion or traditional beliefs. Some of the women who do not eat pork claimed that they were raised at times when women were denied pork because men believed that eating it makes women too strong and outspoken. Moreover, according to local tradition in rural Masaka district, elderly women are not supposed to eat pork, chicken and red meat. They are given eggs, fish and even bone marrow as this is believed to keep them strong. Almost all (93%) of the mothers emphasized that nobody eats offal, referring to white offal, partly because pigs eat anything including faeces and snakes. Farmers across all sites, especially in rural settings, indicated they may eat meat from diseased pigs if they cannot find a market for their animals. Food for people does not compete with pig feeds as the animals are fed with leftovers or fattened during “times of plenty”, shortly after the seasonal rains.

The main quality criteria that pig farmers consider as important when purchasing pork are presented in Figure 3. Cleanliness of the meat ranked first and was defined as meat free from visible dirt and flies; a moderate fat layer (ranked second) is important because in the view of the discussants too thick may imply human disease (e.g. high blood pressure) and too thin could indicate pig disease (e.g. it did not gain weight because it was sick). Unbalanced feeding of pigs was not associated with the thickness of the subcutaneous fat layer.

Figure 3. Quality attributes as perceived by pig farmers in Kamuli, Mukono and Masaka district. (The bars indicate proportions established by group consensus and have not been further quantified.)

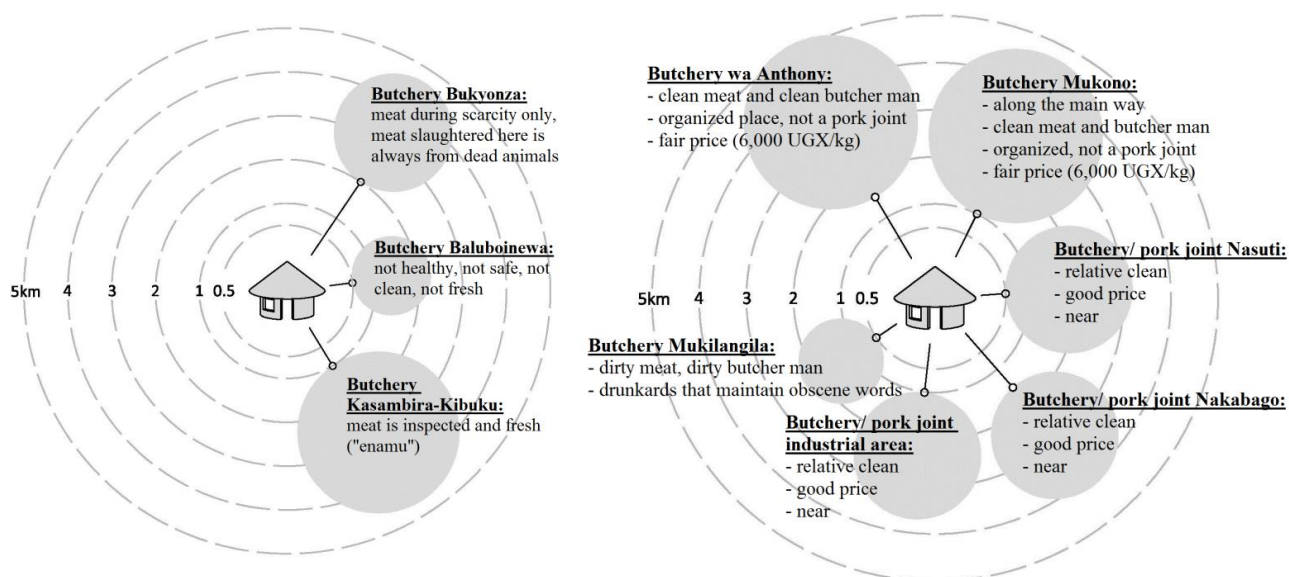


Raw pork is considered unsafe for human consumption and a potential source of diseases across all sites. Pork ranked fifth after chicken, fish, beef and eggs in terms of perceived safety; milk ranked last. All pig farmers agreed that they can contract disease from pigs; symptoms described were worms (26%), stomach pain (20%), diarrhoea (16%) and fever (13%). It is believed that undercooked pork can cause madness or epilepsy in humans. Fifty percent of the participants have heard about food borne diseases in their community and 31%

of them agreed that children are most affected. Food borne diseases are not considered fatal but weaken the person affected and reduce his or her ability to concentrate or work. At home, pork is thoroughly cooked for at least one hour and attempts are made to preserve the shelf-life of raw pork, for instance by smoking and roasting. When eaten outside of the homes, fried or roasted meat is usually consumed with raw vegetables such as tomatoes, cabbage and onions.

People have access to pork in all study sites, and about 70% is consumed outside the homes in pork joints. In the rural areas, consumers have less choice of butchers and the retailers are reported to slaughter diseased animals or sell products in a dirty and unsafe environment (Figure 4).

Figure 4. Venn diagrams developed by the group discussants to describe availability, accessibility and preferences for sources of pork. (left: Baluboinewa village in rural Kamuli district; right: Kitete village in urban Mukono district; the size of the circle represents the relative importance of the source)



### Discussion and conclusion:

Pork is consumed widely and consumers have access to pork in all study sites but quality seems to be neglected in the rural sites where consumers reported less sources of pork and more meat sold from unsanitary outlets. Pork consumption on the occasion of Easter and Christmas often coincides with times of food scarcity. A safe product can therefore contribute to the protein supply of poor farmers and their families during seasons of food shortage.

The consumption of offal is not commonly accepted in the study sites and may need promotion as it provides a source of protein to those who cannot afford to buy pig meat. Offal refers to the internal organs and entrails of an animal slaughtered for meat. While the farmers in the study sites cook red offal (heart, tongue, lungs, spleen and kidneys), white offal are only partly used. Meat is scraped off of feet and heads, and in Masaka district bone marrow is sometimes given to old people to help them maintain their body strength. Brains and genital parts including the uterus and teats are not eaten at all and people do not know about the existence of the pancreas (sweet breads) which is usually discarded in the open by the

butchers together with the slaughtered animal's stomach, intestines, blood and faeces. There they are left for scavengers, potentially contributing to spread of diseases and environmental pollution.

While the meat is consumed hot, the preparation of pork dishes with raw vegetables on the side may lead to cross-contamination with foodborne pathogens. The pathognomonic signs of diamond skin disease (*Erysipelothrix rhusiopathiae*) were reported in Kamuli district and pose a risk to people handling raw pork like butchers and house wives preparing the meat. The common misperception of the life cycle of *Taenia solium* causes inefficient management of the disease risks. More research is required and currently conducted on pork parasites such as trichinosis and both porcine and human cysticercosis due to the common practice of roasting pork which may lead to the ingestion of parasitic larvae.

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## 7 Summarizing discussion

The aim of the present study was to contribute to the evidence base on parasitic infections in pigs raised in smallholder production settings in Uganda. It was hypothesized that the intensification level may affect the parasitic burden of pigs and hence potentially compromise farm productivity and public health, whereby the value chain domain classification established by ILRI and partners (chapter 3) served as a proxy for the level of intensification in smallholder pig value chains in Uganda. While prevalence and potential risk factors for selected parasites have been discussed in-depth in chapters 4 and 5, in this chapter, we will put the key findings in the overall context of research in smallholder pig value chains in Uganda and how they contribute to the overarching goals of the research programs led by ILRI.

### 7.1 Value chain domains as a suitable proxy for the classification of heterogeneous smallholder pig production systems

Pig production in sub-Saharan Africa in general, and Uganda in particular, is increasing (see chapter 2). The fact that numerous governments, including that of Uganda do not proactively promote pig-keeping (Tatwangire, 2013), yet pig numbers are rising, shows that growth of this agribusiness may be intrinsic, perhaps motivated by both the demand for pork driven by urbanization (Robinson et al., 2014) and the preference for a livestock species that can generate returns quickly with limited resources in the absence of access to financial infrastructure. The various assessments of the sector and our research in Uganda show that there are many constraints to the sector as a whole, such as poor organization and lack of governmental support, but also particular constraints, especially to rural producers who face high disease burdens and high costs of investments but also lack many basic skills both entrepreneurial (e.g. record keeping) and in pig husbandry (e.g. biosecurity or pig nutrition) which makes them less competitive. But because pigs are resilient they manage to survive under basic conditions; smallholder pig farmers could potentially make (much more) profit if the range of knowledge gaps could be closed. While the smallholder pig production systems in the selected study sites have been well described in chapter 4 (Dione et al., 2014) and by Ouma et al. (2014); the findings also show the heterogeneity of these systems. All had in common that pigs are mostly kept for income generation, but important determinants for pig health could not be categorized easily. An important example is the level of confinement as pigs that roam and scavenge are more exposed to pathogens, yet confinement types showed a wide variety defying rigid classification. Some farmers let pigs roam during the day and tether or house them during the night. Others leave them roaming when fields are fallow and have them tethered or housed during planting and harvesting season. For instance, the publication in chapter 4 by Dione et al. (2014) showed that during the fallow season when the participatory rural appraisals were conducted, 17.5% of the pigs were free ranging in rural production settings and only 1% in urban production settings. A few months later, during the prevalence survey which happened during the planting season, only a very small fraction of the pigs were free-ranging (Roesel et al., 2017, 2016b). Yet others confine their animals if their farm is located in a neighbourhood where pigs are not tolerated or if they have the resources to feed and house them, which can fluctuate within a short time, for instance if the farmer experienced a poor coffee harvest (this also illustrates the vulnerability of smallholder livestock farming systems). However, the structured targeting process described in chapter 3 and the classification of production systems into value chain domains offered a very suitable way to determine the level of intensification at production and at the same time allowed to us draw conclusions on the location of the end

consumer and therefore, the population at risk for pork-borne parasitic infection. While there was no significant difference in gastrointestinal infection between value chain types, the serological findings on pork-borne diseases showed that the rural population is more exposed to infection with *Trichinella* spp., though our findings suggest that the risk for developing trichinellosis is very low (Roesel et al., 2016b). Antibodies against *T. gondii* tachyzoite antigen on the other hand were found at significantly higher levels in the UU settings showing increased exposure to infection of urban butchers and consumers.

## 7.2 Pig parasites as a constraint to farm productivity

Gastrointestinal parasites are common in the Ugandan production systems as shown by our survey (chapter 4) but their impact is often underestimated because infection does not cause high mortality and may be subclinical. Losses may go unnoticed as changes in pig performance are subtle and in small daily amounts. Studies from Western Kenya showed that average daily gains in pigs kept in rural areas were 110 grams versus 150 grams in peri-urban pig units (Carter et al., 2013). Reasons hypothesized were malnourishment (i.e. poor feed availability or poor rations), high energy expenditure for maintenance (i.e. as roaming pigs expend significant calories through movement), high parasite prevalence, other diseases, and, low genetic potential (Carter et al., 2013). The same study reported that the average body weight at marketing age (5-10 months) was 30 kg; this is very low compared to market weight of exotic breeds in industrialized countries (more than 90 kg at 5 months age) or 40-60 kg in rural Nigeria (Machebe et al., 2009) and 50-70 kg in periurban Nigeria (Adeshinwa et al., 2003). Feed costs account for 70-85% of smallholders pig production costs and given that cost and availability of pig feeds are a major production constraint, parasite control could potentially lead to a much higher feed efficiency. Experiments have shown that growers infected with one or more species of gastrointestinal parasites, in particular *A. suum*, *T. suis* or *Oesophagostomum* spp., experienced reduced growth rates of 31-33% (Kipper et al., 2011; Stewart et al., 1985; Hale and Stewart, 1979) as well as changed body composition, e.g. heavier plucks and less meat (Knecht et al., 2011; Roepstorff et al., 2011). However, these studies were carried out in industrialized countries with potentially improved breeds and in a much more controlled environment (e.g. an otherwise balanced diet and vaccination), and there is need to repeat them in local settings and with local breeds.

Our study showed that husbandry practices such as regular removal of faeces from pig pens or regular cleaning and disinfection correlate with a lower parasite burden in the pig population, which was, perhaps surprisingly, not the case for the regular use of dewormers. However, we have to be aware that findings presented here are the results of a cross-sectional study as in previous assessments from Uganda (Waiswa et al., 2007). From the results we are thus not able to draw conclusions on causal relationships between exposure to a pathogen and actual infection; rather they show statistical correlation between those two, which may be suggestive of causation. More experimental and longitudinal studies are needed to probe findings from cross-sectional studies and to quantify gains and losses. These could include the effect of regular deworming (including the frequency), and of sanitary and hygiene measures as well disease incidence and mortality. A particular focus should be given to pre-weaning and grower pigs as these are the periods of fastest growth (Carter et al., 2013) and when morbidity and mortality seems high in the East African smallholder production systems (Ikwap et al., 2014; Wabacha et al., 2004b). Our study (chapter 4, publication II), too, showed an increase of gastrointestinal helminth burden from 3 to 18 months

(pigs younger than that were not included into the study), which then fell between 18 and 36 months of age.

### 7.3 Parasite infections with public health implications

#### 7.3.1 Gastrointestinal parasites as soil-transmitted zoonotic helminths

In addition to direct performance losses, internal parasite infection in pigs, especially growers, can generally compromise vigour and may act synergistically with other endemic potential pathogens (Greve, 2012). In humans, gastrointestinal, or soil-transmitted helminths (STHs) are among the most common infections worldwide and considered a neglected tropical disease by the WHO (2016b). Infection with STHs impact the wellbeing of the infected person with disorders ranging from general malaise and stomach pain to nutritional impairment through weight or blood loss, diarrhoea or dysentery, loss of appetite or reduced absorption of macro- and micronutrients. Children may experience delay in physical, intellectual, and cognitive development (Bethony et al., 2006). In some cases, STHs cause severe complications such as intestinal obstruction or rectal prolapse, and can be fatal if unattended. As outlined in chapter 2, *A. suum* and *T. suis* are infective for humans (Iñiguez et al., 2012; Leles et al., 2012; Liu et al., 2012; Nissen et al., 2012; Peng and Criscione, 2012; Zhou et al., 2012), and infection with both species has been shown to be prevalent, especially among school children in all regions of Uganda (Brooker et al., 2009; Standley et al., 2009; Kolaczinski et al., 2006; Kabatereine et al., 2005).

#### 7.3.2 Pork-borne zoonoses

A long anticipated report which, for the first time, estimated the global burden attributed to foodborne diseases, found that the overall global burden of foodborne disease (including waterborne disease) is comparable to those of the major infectious diseases (HIV/Aids, malaria and tuberculosis). In the African subregion that includes Uganda (Table 7.4), foodborne parasitic diseases play a smaller role than foodborne disease caused by bacteria but a bigger role than foodborne disease caused by viruses (Havelaar et al., 2015). The data in Table 7.4 also show that parasitic infections cause more chronic burden than acute illness, and that *T. gondii* and *T. solium* infections have the greatest impact. While trends are perhaps accurate, the actual burden of foodborne diseases attributed to biological hazards may be underestimated due to a lack of differential diagnostics and thus lack of attribution data in low-income countries. Also, the presence of parasitic diseases in food producing animals such as pigs is still underresearched in sub-Saharan Africa (Alonso et al., 2016).

Of all the pork-borne parasites, “the three T” parasites have been responsible for most pork-borne illness throughout history (Djurković-Djaković et al., 2013). While the research presented in chapter 5 investigated the occurrence of infection with *Trichinella* and *Toxoplasma gondii*, the “third T”, *Taenia solium*, was researched in the same cohort by a different scientist in the team. A comparative literature analysis on the disease situation and predisposing factors in selected countries including Uganda showed that there is limited knowledge on life cycle of the tapeworm by key players including farmers, butchers, veterinary staff and consumers; poor husbandry and sanitation practices contributing to the maintenance of the life cycle are very common; and that prevalence data on porcine infection is scanty and reports on human infection levels are very scarce (Kungu et al., 2015). A prevalence survey in the same pig cohort subject in chapters 4 and 5 showed lower levels of infection with *T. solium* in rural production settings (10.8%) than in urban

settings (17.1%) (Kungu et al., 2016). This was also true for pig infections with *T. gondii* but the opposite case for infections with *Trichinella* spp.

FAO recommends the daily per capita consumption of 20 g animal protein per person (7.3 kg per person per year) (FAO, 2015); the average daily protein intake in Uganda was 12.38 g from animal sourced foods including milk, meat, eggs, fish, and offal (FAOSTAT, 2011), hence moderate pig meat consumption could contribute to improved nutrition security. Some examples presented in detail in chapters 4-6 show how smallholder farmers do indeed consume pork but only occasionally, especially during festivals such as Christmas, but the frequency of consumption increases towards more urban settings to an average of twice a week in Kampala (chapter 6 and Heilmann et al., 2016). Only 10% of the pig producers practice home slaughter (chapters 5 and 6), which is lower than the 33% in rural Western Kenya (Githigia et al., 2005), and most pigs are sold for consumption in urban centres (RU, UU value chains) or local rural eateries (RR value chain), often via middlemen. It is worrying that there is little or no meat inspection, a scenario similar to rural Western Kenya (Githigia et al., 2005), and a characteristic trait of informal markets in low-income countries. In the whole of Uganda, there is only one formally recognized pig abattoir where meat inspectors are employed but a descriptive survey showed that inspection is not structured, solely based on macroscopic lesions (e.g. *Cysticercus cellulosae*), and not risk-based. Pigs are not traceable, data on diseases prevalent in the production areas is lacking and neither laboratory diagnostics nor condemnation policies are enforced (Roesel et al., 2016a). On the other hand, raw pork consumption is not commonly practiced and pork, if prepared and consumed at home, is cooked for a long time (chapter 6) likely to kill parasitic stages in the meat. However, infection with infectious parasitic stages in pork (e.g. ruptured cysts with *T. gondii* bradyzoites) may occur during pork handling (occupational disease). In addition, methods such as roasting that are more common at pork joints may lead to the ingestion of undercooked pork, and accompanying dishes such as raw vegetables may lead to cross-contamination of the meat causing foodborne diseases (Heilmann et al., 2016, 2015).

Across all sites, raw pork is generally considered unsafe for human consumption and is seen as a potential source of disease (chapter 6); health aspects associated with visible meat quality (e.g. cleanliness, colour or fat layer) are important to the pork-consumers in this study and have been confirmed to be important criteria influencing the buying decision of urban pork consumers in Kampala (Heilmann et al., 2016). As most pork is produced by rural farmers and consumed by urban dwellers, organizing and educating urban consumers instead of trivializing (Figures 7.1 and 7.2), paired with increased income in the cities could potentially stimulate greater demand not only for quantity but also for quality of meat products (Adesehinwa et al., 2003).

Table 7.1 The median global numbers of foodborne illnesses, deaths and Disability Adjusted Life Years (DALYs) compared to the median rates of DALYs in the African sub-region E (AFR E) which includes Uganda. Source: Havelaar et al. (2015).

<b>Hazard</b>	<b>Foodborne illness, global (%)</b>	<b>Foodborne deaths, global (%)</b>	<b>Foodborne DALYs<sup>8</sup>, global (%)</b>	<b>Foodborne DALYs, AFR E<sup>9</sup> (%)</b>
<b><i>Protozoa</i></b>	<b>75,124,588 (13.08)</b>	<b>5,913 (1.49)</b>	<b>1,290,360 (4.11)</b>	<b>37,700 (3.45)</b>
<i>Cryptosporidium</i> spp.	8,584,805 (1.50)	3,759 (0.95)	296,156 (0.94)	1,200 (0.11)
<i>Entamoeba histiolytica</i>	28,023,571 (4.88)	1,470 (0.37)	138,863 (0.44)	5,000 (0.46)
<i>Giardia</i> spp.	28,236,123 (4.92)	-	26,270 (0.08)	700 (0.06)
<i>Toxoplasma gondii</i> <sup>10</sup>	10,280,089 (1.79)	684 (0.17)	829,071 (2.64)	20,000 (1.83)
<b><i>Helminths</i></b>	<b>12,924,132 (2.25)</b>	<b>44,897 (11.31)</b>	<b>5,752,017 (18.31)</b>	<b>181,819 (16.62)</b>
<i>Echinococcus granulosus</i>	43,076 (0.01)	482 (0.12)	39,950 (0.13)	800 (0.07)
<i>Echinococcus multilocularis</i>	8,375 (<0.01)	7,771 (1.96)	312,461 (0.99)	-
<i>Taenia solium</i>	370,710 (0.06)	28,114 (7.08)	2,788,426 (8.87)	176,000 (16.08)
<i>Ascaris</i> spp.	12,280,767 (2.14)	1,008 (0.25)	605,278 (1.93)	5,000 (0.46)
<i>Trichinella</i> spp.	4,472 (<0.01)	4 (<0.01)	550 (<0.01)	1 (<0.01)
<i>Clonorchis sinensis</i>	31,620 (0.01)	5,770 (1.45)	522,863 (1.66)	-
<i>Fasciola</i> spp.	10,635 (<0.01)	-	90,041 (0.29)	10 (<0.01)
<i>Intestinal flukes</i> <sup>11</sup>	18,924 (<0.01)	-	155,165 (0.49)	-
<i>Opisthorchis</i> spp.	16,315 (<0.01)	1,498 (0.38)	188,346 (0.60)	-
<i>Paragonimus</i> spp.	139,238 (<0.02)	250 (0.06)	1,048,937 (3.34)	8 (<0.01)
<b><i>Bacteria</i></b>	<b>347,415,818 (60.50)</b>	<b>263,925 (66.47)</b>	<b>19,632,153 (62.48)</b>	<b>785,380 (71.77)</b>
<b><i>Viruses</i></b>	<b>138,513,782 (24.12)</b>	<b>62,660 (15.78)</b>	<b>3,849,845 (12.25)</b>	<b>94,000 (8.59)</b>
<b><i>Chemicals and toxins</i></b>	<b>216,270 (0.04)</b>	<b>19,682 (4.96)</b>	<b>895,128 (2.85)</b>	<b>6,200 (0.57)</b>
<b>Total:</b>	<b>574,194,590 (100.00)</b>	<b>397,077 (100.0)</b>	<b>31,419,503 (100.00)</b>	<b>1,094,299 (100.00)</b>

<sup>8</sup> Disability Adjusted Life Years (DALYs) are used as a measure of population health and combine the years of life lost due to premature death and the years lived with disability from a disease or condition, for varying degrees of severity (Murray and Lopez, 1997)

<sup>9</sup> Subregion AFR E: Botswana; Burundi; Central African Republic; Congo; Côte d'Ivoire; Democratic Republic of the Congo; Eritrea; Ethiopia; Kenya; Lesotho; Malawi; Mozambique; Namibia; Rwanda; South Africa; Swaziland; Uganda; United Republic of Tanzania; Zambia; Zimbabwe. The term subregion does not refer to the WHO member states, it combines the geographical region child and adult mortality levels as described by Ezzati *et al.* (2002)

<sup>10</sup> Numbers presented here refer to the burden of congenital toxoplasmosis

<sup>11</sup> Includes families Echinostomatidae, Fasciolidae, Gymnophallidae, Heterophyidae, Nanophyetidae, Neodiplostomidae and Plagiorchiidae





Figure 7.1 An article published by the newspaper Uganda Daily Monitor on 6 June 2012 referring to a report that suggests high contamination of pork in Kampala. On inquiry, the Kampala City Council Authority's (KCCA) senior veterinary officer explained that there is no report. Statements on pork safety were based on misperceptions not evidence. Source: Safe Food, Fair Food blog post (ILRI, 2012c).



Figure 7.2 One week later, on 13 June 2016, the Ugandan populist newspaper Red Pepper declared a war on informal pork butcheries operating in Kampala, referring to the same misleading “expert report”. Source: Safe Food, Fair Food blog post (ILRI, 2012c).

Improving pig husbandry practices on farm has emerged as a critical factor for (zoonotic) disease control from various sub-activities in the program (Roesel et al., 2017; Dione et al., 2015a; Kungu et al., 2015), and many good hygiene practices have been shown to be successful and well-documented in other countries, for instance by FAO (2010). Knowing what works best is vital for improving smallholder production systems; however, implementation of best-practices (e.g. housing to prevent exposure to zoonotic pathogens) requires initial financial resources that the smallholder farmer will have to bear. But currently, pig-keeping is particularly attractive to resource-poor smallholders (Lekule and Kyvsgaard, 2003; Phiri et al., 2003; Verhulst, 1993) who will not have the means to adapt technology or practice unless they are given incentives, for instance if consumers are willing to pay a premium for higher quality which reaches the producer. On the other hand, many practices have been known to contribute towards the control of a problem, for instance *T. solium*, and the use of latrines has been promoted for decades. However, open defecation is not only linked to the physical access of water or latrines; a recent study conducted by Thys et al. in Zambia (2015) showed that although latrines were perceived to contribute to good hygiene, because they prevent pigs from eating human faeces. Nonetheless people (especially men) expressed reluctance to abandon the practice of open defecation due to cultural taboos. These are especially found in matrilineal cultures such as the Bantu people, who also dominate large parts of Uganda.

#### 7.4 Researching smallholder pig value chains through an integrated approach and participatory methods

Mbuthia et al. (2015) acknowledged that for smallholder pig farming improvement programmes to be successful, there is the need for appropriate understanding of these systems so as to holistically address their constraints. In the Ugandan smallholder pig value chains, the CRPs have been the first to also include nodes to their assessments that were not only linked to inputs increasing farm productivity (feeds, drugs) but also the value chain system at post-harvest (market access, butchers, consumers) (Figure 7.3 and Ouma et al., 2014). The multi-pathogen prevalence survey, of which the parasitic assessment was one component, has provided information on pig pathogens that have never been documented from Uganda before (e.g. *Brucella suis*, *Erysipelothrix rhusiopathiae*, *Trichinella* spp., *T. gondii*, *Yersinia enterocolitica*), and pathogens known to be present in Uganda (e.g. African swine fever virus, *Taenia solium*). Moreover, testing for more parasites (e.g. *Sarcoptes scabiei* var. *suis*, *Trypanosoma* spp.) as well as bacterial and viral pathogens in the same cohort of pigs is still in progress. Joint resources from two CRPs allowed collection of data from a big cohort, which increases statistical power of the results and in the future will facilitate more analysis from the sample, for instance on multimorbidity or intra-cluster correlation. These data will support scientists in designing follow up studies and target limited resources for research.

Participatory methods have proven useful in identifying priority constraints to pig health in smallholder production systems, and in describing production systems and husbandry practices as well as in scoping diseases that present with clinical signs and can be seen macroscopically. These include specific gastrointestinal worms, ectoparasites or diseases that present with pathognomonic signs. For example, in Kamuli district, farmers reported the diamond-shaped skin lesions in their pigs that are characteristic for acute swine erysipelas caused by *Erysipelas rhusiopathiae* (chapter 6). The pathogen was not on the initial list to investigate but its presence has then been confirmed in the laboratory, showing a high level of infection among animals, meat and raw pork handlers (Musewa, 2016). For other conditions such as disease syndromes, symptoms such as fever, diarrhoea, and vomiting can be described, however, a conclusive etiologic diagnosis cannot be made and findings have to be interpreted with care and supported by laboratory findings. This is the case for ASF which has been identified a priority disease among farmers, yet in their pigs they are only able to observe signs such as haemorrhage, fever, shivering, huddling, and sudden death which are commonly seen in other diseases such as classical swine fever, salmonellosis, erysipelas, infections with *T. simiae* among others. In the pig cohort for which laboratory results are presented here, no antibodies to ASF were detected using INGENASIA protocols; however, one animal tested positive for virus antigen using real time PCR (Akol et al., 2015).

Infections that are usually asymptomatic in pigs, for instance with *T. solium*, *Trichinella* spp. or *T. gondii*, cannot be observed by the farmer and will therefore not be mentioned in participatory epidemiology exercises. Nevertheless, through participatory methods we are able to get indications on good and poor practices at production, processing and consumption level that allow better targeting of resources towards in-depth assessments or interventions; these include husbandry practices on farm, preparation of meat, frequency of consumption, modes of pork preparation and knowledge on zoonotic pig diseases in smallholder pig farming communities. The use of Venn Diagrams illustrated sources of pork, their distance from the homesteads and quality attributes that are important to smallholder pig producers outside of the big urban centres (chapter 6, Figure 4).

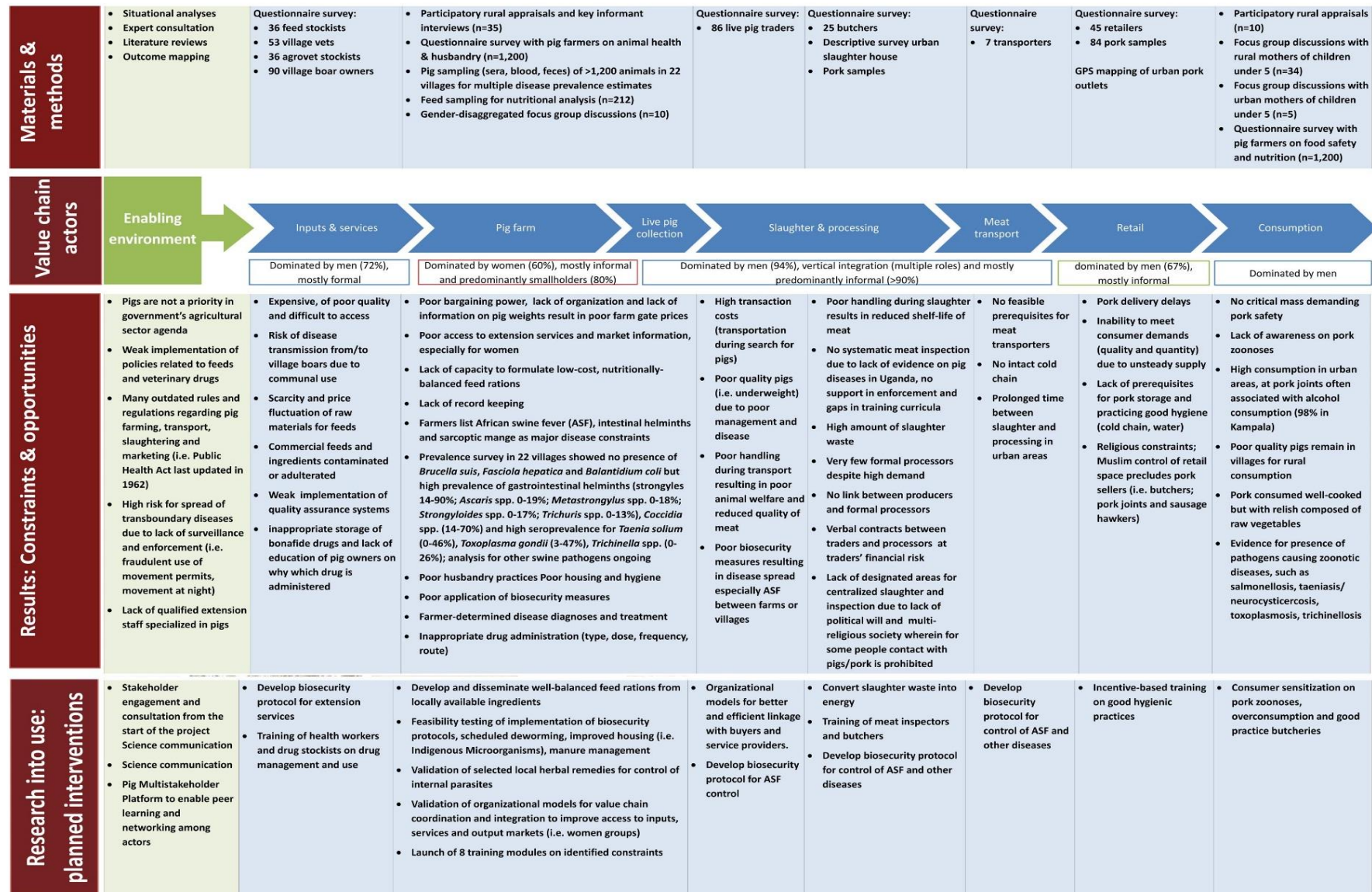


Figure 7.3 Synthesized results of the Ugandan smallholder pig value chain assessment including porcine parasites. The work was presented by the author at Tropentag conference in Prague, September 2014. Source: Ouma et al. (2014).



Participatory methods can indeed be a helpful tool in assessing exposure to food safety risks in informal markets of low-income countries as described earlier (Roesel et al., 2014).

Feedback workshops, where the scientific findings of each sub-activity were discussed with the value chain stakeholders, have been held since the program's inception (chapter 3), and are not only inclusive but help communicating evidence-based findings and solutions in a more bottom-up approach. Based on the assessment findings, seven training modules including parasite control were developed and are tested and validated in the field with smallholder pig producers (Dione et al., 2015b), existing materials have been updated for use in the local setting in Uganda (ILRI and Medical Research Council, 2005), and researchers have been invited to present findings at pig farmer trainings organized by the private sector (ILRI, 2014a). Moreover, training workshops have been held with different stakeholders at the various nodes of the pig value chain, i.e. veterinary public health (ILRI, 2014b). The downside of the intensive interaction with all stakeholders through the application of participatory methods is the increased pressure on scientists to compensate for the big gap in public veterinary extension systems by not only generating data and developing intervention strategies but to implement them at large scale with research resources.

## 7.5 Concluding remarks, limitations of the survey and potential future research

The present study was able to contribute to the increasing evidence base on prevalent pig parasites that may compromise farm productivity but also pork safety in smallholder pig production settings in Uganda.

### 7.5.1 Some of the major research findings are presented below:

- Results from participatory rural appraisals show that pig keeping systems are dominated by tethering and scavenging in rural areas, while in peri-urban and urban areas, confinement in pens is more common. High disease burden, poor housing, poor feeding, poor veterinary services, ineffective drugs and a general lack of knowledge on piggery management were identified as the main production constraints by smallholder pig farmers. Second only to ASF, parasites, especially gastrointestinal worms and sarcoptic mange, were perceived to contribute largely to market losses due to stunting across all value chain domains and study sites.
- The majority (61.4%) of all pigs were found to be infected with at least one species of gastrointestinal helminths: 57.1% strongyle species, 7.6% *Metastrongylus* spp., 5.9% *A. suum*, 4.2% *Strongyloides ransomi*, and 3.4% *Trichuris suis*. There were no statistically significant differences across the different value chain types but across the different study districts. Infection with *A. suum* and *Metastrongylus* spp. were significantly more common in rural Kamuli districts ( $p < 0.01$ ) than in the other two districts while *S. ransomi* was more commonly found in urban Mukono district ( $p < 0.01$ ). Coccidia oocysts were found in 40.7% of the sampled pigs, with significantly higher levels in the more urban Mukono district. None of the animals showed infection with *Balantidium coli* or *Fasciola hepatica*. A logistic regression model showed that routine management factors had a greater impact on the prevalence of infection than regular treatment or the level of confinement. Factors negatively correlating with gastrointestinal helminth infection were routine manure removal, and the routine use of disinfectants.

- The overall prevalence of *Trichinella*-specific antibodies was 6.9% out of 1,125 sampled animals but showed significantly higher levels in value chain types with a rural origin of production (RR and RU value chains) ( $p < 0.05$ ). Kamuli district, where the RR value chain type dominates showed a significantly higher ELISA-prevalence than the more urban Masaka and Mukono districts ( $p < 0.001$ ). For the first time, a study in pigs in sub-Saharan Africa was able to confirm 31 % ELISA-positive samples by Western Blot. Isolation of infective first-stage larvae from 499 samples in ELISA clusters to identify the dominating *Trichinella* spp. was not successful.
- *Toxoplasma*-specific antibodies were found in 28.7% of the animals using a commercial ELISA kit; village herd prevalence was 100%. Pigs raised in urban production settings (UU value chain types) showed significantly higher levels of seropositivity than animals of a rural production origin (RR and RU value chain types) ( $p < 0.007$ ); and significantly higher levels were shown for Masaka and Mukono compared to rural Kamuli district ( $p > 0.05$ ). Moreover, univariate analysis suggested a significant association between seropositivity and pig age, feeding of crop residues, source of drinking water, keeping cats on farm compound, and frequent sightings of wildlife (especially antelopes, hares, wild and stray dogs) near the village. The present report is the first survey documenting the seroprevalence of *T. gondii* in domestic pigs in the East African Community.
- Findings on pork consumption practices from participatory rural appraisals showed that pork is widely popular among pig farmers. In rural areas it is consumed mostly during seasonal festivities and whenever cash is available, in urban areas much more frequently. According to the pig farmers, pig feed production does not compete with human food production; some festivals when pork is consumed even coincide with times when other food is not abundant. The consumption of raw pork is considered unsafe, and at home meat is cooked for at least one hour.

### 7.5.2 Major conclusions that can be drawn from this assessment are:

- Smallholder pig production systems in Uganda are highly heterogeneous and complex, and the level of intensification cannot be characterized by a single variable such as the level of confinement. Due to the informal setting of the pig sector, even a fully confined pig may be exposed to parasites introduced to the farm, e.g. feeds contaminated with rodents or cat faeces. Using a proxy of value chain domains that comprised different aspects, in this case pig population density, poverty levels and the time to reach the nearest market, was useful for comparing pig populations in different villages.
- Infections with gastrointestinal parasites are common but weight loss, a consequence of worm infection as perceived by pig farmers, could not be attributed to a particular parasite species. Other factors causing malnutrition due to poor pig diets, or potentially a combination of these two, have not been investigated in this cohort of pigs. Regular treatment with anthelmintics alone is not useful in containing the burden of infection at acceptable levels if efficient hygiene farming practices such as regular removal of faeces, cleaning and disinfection of pig pens are not followed.

- The *Trichinella* species infecting domestic pigs needs to be identified by isolation of muscle larvae, which was not successful in this study. Subsequently, serological assays developed for the diagnostics of *Trichinella* infection in domestic pigs in high-income countries need to be adapted for use in the local settings of smallholder pig production in the tropics.
- The findings on the seroprevalence of antibodies to *T. gondii* in domestic pigs imply that more research should be carried out to determine the genotype infecting pigs as well as the tissue cyst burden to be able to re-assess the risk for human raw pork handlers and the vulnerable population, e.g. pregnant women and HIV/Aids patients. Consequently, in-depth research on transmission routes for domestic pigs and humans is warranted.
- The high prevalence of gastrointestinal parasites, some of them zoonotic, may expose pig farmers and their children, who often tend to the pigs, to disease. While, according to the findings of this study, the risk for developing trichinellosis is low, the risk for infection with *T. gondii* from raw pork handling or other practices on farm (e.g. handling feeds, or drinking contaminated with oocysts) is higher.

Constraints to the parasitic assessment were mostly related to the design of the overall pig value chain assessment which was cross-sectional and could therefore suggest but not prove causal relationships between exposing variables and parasite infection. Despite the outweighing benefits described above, a shortcoming of the prevalence survey for parasitic assessment, was the exclusion of pigs younger than three months from the sample population as this is the age when they are most susceptible to infection with gastrointestinal parasites. Moreover, the tight field sampling schedule and limited time, space and human resources did not allow for the collection and analysis of urine to determine the prevalence of *Stephanurus dentatus* (kidney worm), to perform larval culture and coccidia oocyst sporulation, or to collect deep skin scrapings and ectoparasites from all pigs, which could have added useful information. Specimens were collected from a number of pigs but this was not done systematically.

Additional laboratory experiments conducted after the prevalence survey included artificial digestion to potentially isolate first-stage larvae of *Trichinella* spp. clusters and a mouse bioassay to isolate *T. gondii* bradyzoites for genotyping in ELISA clusters. While the sample for the artificial digestion was large (n=499), no larvae could be isolated and reasons were exhaustively discussed in chapter 5. However, the serological status of the digested samples could not be confirmed due to limited resources. Attempts to isolate *T. gondii* in a mouse bioassay were also not successful, even though only serologically positive samples had been used as recommended (Dubey, 2009). *T. gondii* bioassays in cats have been shown to be more successful in the isolation of viable cysts as larger volumes (500g) or more can be assayed than in mice; however, facilities for experiments involving cats were not available in Uganda at the time of the study.

Future research on pig parasites in Uganda should aim at:

- Quantifying actual losses due to gastrointestinal parasites on farm by means of experimental studies and longitudinal surveys;

- Characterizing pork pathogens, for instance the *Trichinella* species infecting pigs in Uganda or the genotype involved in porcine infection with *T. gondii*;
- Assessing the effect of co-infection on the sensitivity and specificity of serological assays developed and validated in high-income countries;
- Quantifying the risk for pork-borne parasitic diseases to consumers through experimental studies and quantitative risk assessments;
- Assessing host preferences of biting insects such as tsetse flies including pigs;
- Assessing the role of Ugandan pigs as potential reservoir or amplifying host for zoonotic arboviruses such as Ndumu transmitted by *Culicoides* spp. (Masembe et al., 2012); Zika virus transmitted by *Aedes* spp. (Chan et al., 2016), or Japanese encephalitis transmitted by *Culex* spp. (Mellor and Leake, 2000);
- Characterizing African pig breeds and their genetic tolerance or susceptibility to parasitic diseases as has been shown for African cattle (Gifford-Gonzalez and Hanotte, 2011);
- Including wild Suidae in parasitic assessments as they could potentially share vector-borne infections such as with *B. trautmanni* (Penzhorn, 2006), or trypanosomes (Mehlitz, 1987; Mwambu and Woodford, 1972; Van Den Berghe and Zaghi, 1963);
- Investigating the role of potential hosts such as dogs and rodents as mechanical vectors (e.g. *T. gondii*, *A. suum*), or definite hosts (e.g. *Trichinella* spp., *Echinococcus* spp., *Neospora caninum*, *Sarcocystis* spp.) for potentially pig-mediated zoonoses in low-income countries, where they are often stray;
- Testing the feasibility of dogs or chickens as bio-indicators and sentinels for infections with *T. gondii* (Ayinmode and Jones-Akinbobola, 2015; Davoust et al., 2015; Salb et al., 2008; Meireles et al., 2004)

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## Summary

The surging demand for pork in Uganda provides an opportunity to set poor farmers on pathways out of poverty by increasing production and market access. At the same time, markets have become more complex and threaten the continuous presence of smallholder farmers: pig diseases and other factors hinder a constant supply while foodborne diseases may impair the quality and safety of the farmers' products. The intensification level may affect the parasitic burden of pigs and hence the profitability of pig farming as well as the risk to human health associated with pork borne parasites. Research on parasitic hazards in smallholder pig value chains in Uganda has so far been limited, and therefore, the present study aims, as described in chapter 1, to contribute to improving selected smallholder pig value chains in Uganda by increasing the evidence base on prevalent parasitic diseases.

Chapter 2 outlines the history of pigs in Africa, describes the current smallholder pig production systems in sub-Saharan Africa, and reviews the current state of knowledge on parasitic pig diseases compromising farm productivity as well as parasitic diseases that are potentially transmitted to humans through pork consumption or for which pigs are known to be a natural disease reservoir. The review refers to previous research from sub-Saharan Africa, and particularly Uganda.

Chapter 3 describes the context of the study and how it fits into overarching research for development on smallholder pig value chains in Uganda led by the International Livestock Research Institute (ILRI). It further describes how 35 villages were selected to be included into the survey, how value chain systems were defined as a proxy for the level of intensification, and characteristics of the area under study.

Chapter 4 shows that both endo- and ectoparasites are perceived as an important production constraint by smallholder pig farmers in Central and Eastern Uganda, second only to African swine fever. Herd dynamics and husbandry practices in the predominating production settings were established by means of participatory rural appraisals with 340 smallholder pig farmers in the selected sites. Consecutively, a cross-sectional parasitological study in 21 of the 35 selected villages established baseline information on the rate and determinants of infection with gastrointestinal helminths and coccidia in 932 randomly sampled pigs between 3 to 36 months of age. The majority of pigs (61.4%; 95% confidence interval [CI]: 58.2-64.5%) tested positive for one or more gastrointestinal helminths, namely strongyles (57.1%; 95% CI: 53.8-60.3%), *Metastrongylus* spp. (7.6%; 95% CI: 6.1-9.6%), *Ascaris suum* (5.9%; 95% CI: 4.5-7.6%), *Strongyloides ransomi* (4.2%; 95% CI: 3.1-5.7%) and *Trichuris suis* (3.4%; 95% CI: 2.4-4.8%). Coccidia oocysts were found in 40.7% of all pigs sampled (95% CI: 37.5-44.0%). All animals tested negative for *Fasciola* spp. and *Balantidium coli*. There were no statistically significant differences in prevalence across value chain domains, and regression analysis showed that routine management factors had a greater impact on the prevalence of infection than regular preventive medical treatment or the level of confinement.

Chapter 5 provides baseline information on two pork borne zoonoses of global importance, namely infection with *Trichinella* spp. and *Toxoplasma gondii*, in the same cohort of pigs. *Trichinella*-specific immunoglobulin G was found in 6.9% of the animals (95% CI: 5.6-8.6%) using a commercially available enzyme-linked immunosorbent assay (ELISA). Confirmatory testing by Western blot implied an overall seroprevalence of 2.1% (95% CI: 1.4-3.2%), and attempts to isolate muscle larvae using artificial digestion were unsuccessful. Implications for diagnostics

were discussed and the risk to pork consumers for developing trichinellosis was classified as low. Antibodies to *T. gondii* were found in 28.7% of the animals (95% CI: 25.8-31.7%) at significantly higher levels in urban areas of production and consumption, and risk factors that potentially contribute to infection (e.g. biosecurity practices and access to cats) were identified.

Chapter 6 outlines the findings on common preparation and consumption practices as well as sources of pork for consumption in the study area. These show that pork is very popular among pig farmers. In rural areas it is consumed mostly during seasonal festivities and whenever cash is available, in urban areas much more frequently. Pig feed production does not compete with human food production, and some festivals when pork is consumed even coincide with times when other food is scarce. The consumption of raw pork is considered unsafe, and at home meat is cooked for at least one hour.

Chapter 7 discusses the findings, conclusions and limitations of the overall study and generates recommendations for potential interventions and further research. Parasitic infections in smallholder pig value chains in Uganda are common; and while no significant difference was observed in overall infection rates with gastrointestinal helminths, specific parasites were detected at significantly higher levels in urban (e.g. *T. gondii*) or rural value chain types (e.g. *Trichinella* spp.). Further in-depth research is needed to identify the circulating *Trichinella* spp. and *T. gondii* genotype, as well as more experimental and longitudinal studies to quantify economic losses of parasite infection, and to establish predominant transmission routes and thus control points for parasite control.

## Zusammenfassung

### **Untersuchungen zu parasitären Infektionen, inklusive ausgewählter Zoonosen, in Hausschweinepopulationen in kleinbäuerlichen Wertschöpfungsketten in Uganda**

Die steigende Nachfrage nach Schweinefleisch in Uganda bietet ressourcenschwachen Kleinbauern einen Ausweg aus ihrer Armut, sollte es ihnen gelingen, Schweine rentabel zu halten und zu vermarkten. Gleichzeitig werden diese Märkte immer komplexer und erhöhen den Druck auf kleinbäuerliche Schweinehalter: Infektionskrankheiten und andere Faktoren bedrohen eine konstante Versorgung des Marktes, und zoonotische Erreger beeinträchtigen gegebenenfalls die Qualität und Sicherheit des Endproduktes. Der Intensivierungsgrad bedingt möglicherweise das Ausmaß der Infektion mit Parasitosen und somit die Wirtschaftlichkeit der Schweinehaltung und das Risiko für den Verbraucher. Es gibt nur wenige Untersuchungen zu Parasitosen in kleinbäuerlichen Schweinebetrieben in Uganda, und die vorliegende Arbeit hat zum Ziel, wie in Kapitel 1 beschrieben, einige dieser Kenntnislücken schließen zu helfen.

Kapitel 2 skizziert die Geschichte des Hausschweins in Afrika, beschreibt die derzeitigen kleinbäuerlichen Schweinehaltungssysteme im Afrika südlich der Sahara und bespricht den aktuellen Wissensstand zu wirtschaftlich relevanten Schweineparasitosen als auch zoonotischen Erkrankungen, die über den Fleischverzehr übertragen werden oder für die Schweine natürliches Reservoir sind. Die Literaturrecherche bezieht sich besonders auf den Sachstand in Subsahara-Afrika und hier ganz besonders Uganda.

Kapitel 3 beschreibt den Kontext der Studie und ordnet sie in ein umfassendes Programm des International Livestock Research Instituts (ILRI) ein, das durch „Forschung für Entwicklung“ mit der Optimierung kleinbäuerlicher Nutztierwertschöpfungsketten zur Armutsbekämpfung beizutragen versucht. Das Kapitel erläutert ferner, wie die 35 Studienorte ausgewählt, Wertschöpfungsketten stellvertretend für den Intensivierungsgrad definiert wurden und faßt Charakteristika der Studienstandorte zusammen.

Kapitel 4 zeigt, dass Endo- und Ektoparasiten als bedeutendes Produktionshemmnis von kleinbäuerlichen Schweinehaltern in Zentral- und Ost-Uganda wahrgenommen werden; Parasitosen stehen an zweiter Stelle nach der Afrikanischen Schweinepest. Herdendynamik und Haltungsbedingungen wurden mittels partizipativer Methoden mit 340 Schweinehaltern beschrieben und teils quantifiziert. Danach wurde in 21 der 35 Studienstandorte eine Querschnittsstudie durchgeführt, die in einer Stichprobe von 932 Tieren im Alter zwischen 3 und 36 Monaten Daten zu Infektionsrate und korrelierenden Faktoren mit gastrointestinalen Helminthen und Kokzidien erhob. Die Mehrheit der Schweine (61.4%; 95% Konfidenzintervall [KI]: 58.2-64.5%) war mit mindestens einer Helminthenart infiziert, hauptsächlich Magen-Darm-Strongyliden (57.1%; 95% KI: 53.8-60.3%), *Metastrongylus* spp. (7.6%; 95% KI: 6.1-9.6%), *Ascaris suum* (5.9%; 95% KI: 4.5-7.6%), *Strongyloides ransomi* (4.2%; 95% KI: 3.1-5.7%) und *Trichuris suis* (3.4%; 95% KI: 2.4-4.8%). Kokzidienoozysten wurden in 40.7% aller Tiere diagnostiziert (95% KI: 37.5-44.0%) und der Befund für Infektionen mit *Fasciola* spp. und *Balantidium coli* war in allen Tieren negativ. Es gab keine statistisch signifikanten Unterschiede in der Prävalenz zwischen den verschiedenen Typen der Wertschöpfungsketten und eine Regressionsanalyse zeigte, dass routinemäßig ausgeführte Hygienepraktiken einen größeren Einfluss auf die Prävalenz hatten als regelmäßige medikamentöse Prophylaxe oder Aufstallung.

Kapitel 5 präsentiert Basisdaten in der gleichen Kohorte von Tieren zu zwei weltweit vorkommenden Zoonosen, die durch den Verzehr von Schweinefleisch übertragen werden können: *Trichinella* spp. und *Toxoplasma gondii*. *Trichinella*-spezifisches Immunglobulin G wurde mittels kommerziellem ELISA in 6.9% der Tiere detektiert (95% KI: 5.6-8.6%,). Nach Durchführung eines Western Blot zur Bestätigung sank die Seroprävalenz auf 2.1% (95% KI: 1.4-3.2%,). Der Versuch, infektiöse Larven mithilfe der Verdäumethode zu isolieren, blieb erfolglos. Schlussfolgerungen für die Diagnostik wurden diskutiert und das Risiko des Verbrauchers, durch den Verzehr von Schweinefleisch klinische Trichinellose zu entwickeln, wurde als gering eingestuft. Spezifische Antikörper gegen *T. gondii* wurden in 28.7% der Tiere festgestellt (95% KI: 25.8-31.7%,), das Infektionsniveau war signifikant höher in urbanen Produktions- und Verzehrgebieten. Faktoren, die mit einer Infektion korrelierten, waren Hygienepraktiken und der Zugang von Schweinen zu Katzen.

Kapitel 6 stellt die Ergebnisse einer Studie zu den Verzehrsgewohnheiten von Schweinefleisch unter den kleinbäuerlichen Produzenten vor. Schweinefleisch ist sehr beliebt, und während es in ländlichen Gebieten eher unregelmäßig konsumiert wird, z.B. an besonderen Anlässen oder wenn die Familie genug Geld zur Verfügung hat, nimmt der Verbraucher im Städtischen viel häufiger Schweinefleisch zu sich. Die Bereitstellung von Schweinefutter konkurriert nicht mit Lebensmitteln für den menschlichen Verzehr, manche Festlichkeiten, an denen Schweinefleisch gegessen wird, fallen sogar in Zeiten, in denen das Nahrungsmittelangebot knapp ist. Der Verzehr von rohem Fleisch wird als nicht sicher angesehen und Schweinefleisch für den Verzehr zu Hause mindestens eine Stunde erhitzt.

Kapitel 7 diskutiert die Ergebnisse und Schlussfolgerungen der vorangegangenen Kapitel, Grenzen der vorgelegten Arbeit und Möglichkeiten für künftige Interventionen und Untersuchungen. Parasitäre Infektionen in kleinbäuerlich gehaltenen Schweinen in Uganda sind weit verbreitet. Während es zwischen Infektionsraten mit gastrointestinalen Helminthen keine statistisch signifikanten Unterschiede zwischen den verschiedenen Wertschöpfungskettentypen gab, war die Infektionsrate mit *T. gondii* signifikant höher in urbanen Produktionssystemen und das Vorkommen von *Trichinella* spp. Antikörper signifikant höher in ländlichen Gebieten. Tiefergehende Studien zur Bestimmung der zirkulierenden *Trichinella*-Spezies und *T.-gondii*-Genotypen sind nötig, ebenso wie experimentelle und Langzeitstudien, um wirtschaftliche Verluste zu quantifizieren und Übertragungswege und Kritische Kontrollpunkte für Parasitosen zu identifizieren.

**List of own publications**

The following section lists the author's publications during the period of the study.

**1. Peer-reviewed journal publications**

- Kungu, J.M., Dione, M., **Roesel, K.**, Ejobi, F., Ocaido, M. and Grace, D. 2017. Assessment of hygiene practices of pork retail outlets in Kampala district, Uganda. *International Food Research Journal* 24(4): 1368–1373. <http://www.ifrj.upm.edu.my/ifrj-2017-24-issue-4.html>
- Heilmann, M., **Roesel, K.**, Grace, D., Bauer, B. and Clausen, P.-H. 2017. The impact of insecticide treated material to reduce flies among pork outlets in Kampala, Uganda. *Parasitology Research* 116(6):1617-1626. <http://dx.doi.org/10.1007/s00436-017-5450-x>
- Roesel, K.**, Nöckler, K., Baumann, M.P.O., Fries, R., Dione, M.M., Clausen, P.-H. and Grace, D. 2016. First report of the occurrence of *Trichinella*-specific antibodies in domestic pigs in Central and Eastern Uganda. *PLOS ONE* 11(11): e0166258. <http://dx.doi.org/10.1371/journal.pone.0166258>
- Roesel, K.**, Dohoo, I., Baumann, M., Dione, M., Grace, D. and Clausen, P.-H. 2016. Prevalence and risk factors for gastrointestinal parasites in small-scale pig enterprises in Central and Eastern Uganda. *Parasitology Research* <http://dx.doi.org/10.1007/s00436-016-5296-7>
- Alonso, S., Lindahl, J., **Roesel, K.**, Traoré, S.G., Yobouet, B.A., Ndour, A.P.N., Carron, M. and Grace, D. 2016. Where literature is scarce: observations and lessons learnt from four systematic reviews of zoonoses in African countries. *Animal Health Research Reviews* 17(01): 28–38. <http://dx.doi.org/10.1017/S1466252316000104>
- Erume, J., **Roesel, K.**, Dione, M.M., Ejobi, F., Mboowa, G., Kungu, J.M., Akol, J., Pezo, D., El-Adawy, H., Melzer, F., Elschner, M., Neubauer, H. and Grace, D. 2016. Serological and molecular investigation for brucellosis in swine in selected districts of Uganda. *Tropical Animal Health and Production* 48(6): 1147–1155. <http://dx.doi.org/10.1007/s11250-016-1067-9>
- Stentiford, G.D., Becnel, J.J., Weiss, L.M., Keeling, P.J., Didier, E.S., Williams, B.A.P., Bjornson, S., Kent, M.L., Freeman, M.A., Brown, M.J.F., Troemel, E.R., **Roesel, K.**, Sokolova, Y., Snowden, K.F. and Solter, L. 2016. Microsporidia – Emergent pathogens in the global food chain. *Trends in Parasitology* 32(4): 336-348. <http://dx.doi.org/10.1016/j.pt.2015.12.004>
- Atherstone, C., Smith, E., Ochungo, P., **Roesel, K.** and Grace, D. 2017. Assessing the potential role of pigs in the epidemiology of Ebola virus in Uganda. *Transboundary and Emerging Diseases*. <https://dx.doi.org/10.1111/tbed.12394> [Epub Aug 2015]
- Dione, M.M., Akol, J., **Roesel, K.**, Kungu, J., Ouma, E.A., Wieland, B. and Pezo, D. 2017. Risk factors for African swine fever in smallholder pig production systems in Uganda. *Transboundary and Emerging Diseases* 64(3):872-882. <https://dx.doi.org/10.1111/tbed.12452> [Epub Dec 2015]

Dione, M.M., Ouma, E.A., **Roesel, K.**, Kungu, J., Lule, P. and Pezo, D. 2014. Participatory assessment of animal health and husbandry practices in smallholder pig production systems in three high poverty districts in Uganda. *Preventive Veterinary Medicine* 117(3-4):565-576. <https://dx.doi.org/10.1016/j.prevetmed.2014.10.012>; <https://dx.doi.org/10.1016/j.prevetmed.2015.03.010>

**Under review at the time of graduation (shared first authorship):**

Musewa, A., **Roesel, K.**, Grace, D., Dione, M., Erume, J. Detection of *Erysipelothrix rhusiopathiae* in naturally infected pigs in Kamuli district, Uganda.

Ndoboli, D., **Roesel, K.**, Heilmann, M., Alter, T., Clausen, P.-H., Wampande, E., Grace, D., Huehn, S. Serotypes and antimicrobial resistance patterns of *Salmonella enterica* subsp. *enterica* in pork and related fresh vegetable servings among pork outlets in Kampala, Uganda.

## 2. Conference presentations

**Roesel, K.**, Makita, K., Grace, D., 2017. Food safety research for development in sub-Saharan Africa: Tapping the expertise of German partners. Poster presented at the Joint International Symposium “Global Past, Present and Future Challenges in Risk Assessment – Strengthening Consumer Health Protection” at the German Federal Institute for Risk Assessment (BfR), Berlin, Germany, 29 November to 1 December 2017. Nairobi, Kenya: ILRI.

Alonso, S., **Roesel, K.**, Opiyo, S., Stomeo, F. and Grace, D. 2017. Metagenomics in food safety: What's the added value? Case studies from the livestock sector in Tanzania and Uganda. Presented at the FAO Regional Meeting on Agricultural Biotechnologies in Sustainable Food Systems and Nutrition in sub-Saharan Africa, Addis Ababa, Ethiopia, 22–24 November 2017. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/89560>

Grace, D., Alonso, S., Dominguez-Salas, P., Fahrion, A., Häsler, B., Heilmann, M., Hoffmann, V., Kang'ethe, E. and **Roesel, K.** 2017. Food safety metrics relevant to low- and middle-income countries. Poster prepared for the 2nd annual Agriculture, Nutrition and Health (ANH) Academy Week, Kathmandu, Nepal, 9–13 July 2017. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/89032>

**Roesel, K.** 2017. Food safety interventions: economic and health outcomes and impacts. Presentation at a Brussels Development Briefing on "Better targeting food safety investments in low and middle income countries", Brussels, Belgium, 24 May 2017. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/82661>

**Roesel, K.** 2016. Verbreitung des Hausschweins in Uganda: Chancen & Herausforderungen. Presentation at 57. Conference of the food safety chapter of the German Veterinary Association (DVG), 29 September 2016, Garmisch-Partenkirchen, Germany. <https://cgspace.cgiar.org/handle/10568/79996>

- Roesel, K.**, Schares, G., Grace, D., Baumann, M.P.O., Fries, R., Dione, M. and Clausen, P.-H. 2016. The occurrence of porcine *Toxoplasma gondii* infections in smallholder production systems in Uganda. Presentation at the first joint conference of the Association of Institutions for Tropical Veterinary Medicine and the Society of Tropical Veterinary Medicine, Berlin, Germany, 4–8 September 2016. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/77110>
- Ndoboli, D., Heilmann, M., **Roesel, K.**, Clausen, P.-H., Wampande, E., Grace, D., Alter, T. and Huehn, S. 2016. Antimicrobial resistance of *Salmonella enterica* in pork and vegetable servings at pork joints in Kampala, Uganda. Poster presented at the first joint conference of the Association of Institutions for Tropical Veterinary Medicine and the Society of Tropical Veterinary Medicine, Berlin, Germany, 4–8 September 2016. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/77109>
- Heilmann, M., **Roesel, K.**, Clausen, P.-H. and Grace, D. 2016. Knowledge, attitudes and practices among customers at pork butcherries in Kampala, Uganda. Presentation at the first joint conference of the Association of Institutions for Tropical Veterinary Medicine and the Society of Tropical Veterinary Medicine, Berlin, Germany, 4–8 September 2016. Berlin, Germany: Freie Universität Berlin. <https://cgspace.cgiar.org/handle/10568/77090>
- Musewa, A., **Roesel, K.**, Nakanjako, D., Grace, D., Ssenyonga, R., Nangendo, J., Kawooya, I. and Erume, J. 2016. *Erysipelothrix rhusiopathiae* infection in pigs, pork and raw pork handlers in Kamuli District, Eastern Uganda. Presentation at the first joint conference of the Association of Institutions for Tropical Veterinary Medicine and the Society of Tropical Veterinary Medicine, Berlin, Germany, 4–8 September 2016. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/77111>
- Hyera, E., Msalya, G., Karimuribo, E.D., Kurwijila, L.R., Alonso, S., **Roesel, K.** and Grace, D. 2016. Isolation and identification of *Listeria* species along the milk value chain in one region of Tanzania. Poster presented at the first joint conference of the Association of Institutions for Tropical Veterinary Medicine and the Society of Tropical Veterinary Medicine, Berlin, Germany, 4–8 September 2016. Kilimanjaro, Tanzania: Tanzania Livestock Research Institute. <https://cgspace.cgiar.org/handle/10568/77092>
- Alonso, S., Kang'ethe, E., **Roesel, K.**, Dror, I. and Grace, D. 2015. 'You have an SMS': Innovative knowledge transfer for agriculture and health. Poster prepared for the European College of Veterinary Public Health (ECVPH) annual scientific conference, Belgrade, Serbia, 7-10 October 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68716>
- Roesel, K.**, Dione, M., Nöckler, K., Fries, R., Baumann, M.P.O., Clausen, P.-H. and Grace, D. 2015. Exposure of pigs to *Trichinella* spp. in three districts in Central and Eastern Uganda. Abstract of paper presented at the 14th International Conference on Trichinellosis, Berlin, Germany, 14-18 September 2015. <https://cgspace.cgiar.org/handle/10568/68510>
- Heilmann, M., Mtimet, N., **Roesel, K.** and Grace, D. 2015. Assessing Ugandan pork butchers' practices and their perception of customers' preferences: A best-worst approach. Poster prepared for the 9th European Congress on Tropical Medicine and International Health,

- Basel, Switzerland, 6–10 September 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68509>
- Grace, D., Unger, F., **Roesel, K.**, Tinega, G., Ndoboli, D., Sinh Dang-Xuan, Hung Nguyen-Viet and Robinson, T. 2015. Present and future use of antimicrobials in pigs with case studies from Uganda and Vietnam. Presented at the Safe Pork 2015 Conference, Porto, Portugal, 8–10 September 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68304>
- Heilmann, M., Ndoboli, D., **Roesel, K.**, Grace, D., Huehn, S., Bauer, B. and Clausen, P.-H. 2015. Occurrence of *Salmonella* spp. in flies and foodstuff from pork butcheries in Kampala, Uganda. Presented at the Annual Expert Meeting on Parasitology and Parasitic Diseases at the German Veterinary Association, Stralsund, Germany, 29 June–1 July 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68283>
- Roesel, K.**, Dione, M., Fries, R., Baumann, M.P.O., Nöckler, K., Schares, G., Grace, D. and Clausen, P.-H. 2015. Assessment of the parasitic burden in smallholder pig value chains and implications for public health in Uganda. Abstract of paper presented at the annual expert meeting on parasitology and parasitic diseases at the German Veterinary Association, Stralsund, Germany, 29 June – 1 July 2015. <https://cgspace.cgiar.org/handle/10568/68301>
- Dione, M.M., Pezo, D.A., Ouma, E.A., **Roesel, K.**, Brandes-van Dorresteijn, D. and Kawuma, B. 2015. Training on management of endemic diseases for pig value chains in Uganda. Presented at the 4th International Conference on Sustainable Livelihoods and Health in Africa, Kampala, Uganda, 18–19 June 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/67273>
- Alonso, S., Ocaido, M., Carron, M., **Roesel, K.** and Grace, D. 2015. Foodborne hazards in the scientific literature: Results of a systematic literature review in East African countries. Presented at the Regional Conference on Zoonotic Diseases in Eastern Africa, Naivasha, Kenya, 9–12 March 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/59790>
- Ouma, E., **Roesel, K.**, Dione, M., Carter, N., Pezo, D., Ejobi, F. and Grace, D. 2014. Participatory research for development to upgrade smallholder pig value chains in Uganda. Poster presented at the Tropentag 2014 Conference on Bridging the Gap between Increasing Knowledge and Decreasing Resources, Prague, Czech Republic, 17–19 September 2014. Nairobi, Kenya. ILRI. <https://cgspace.cgiar.org/handle/10568/43794>
- Pezo, D., Ouma, E. A., Lule, P., Dione, M., Lukuyu, B., Carter, N. and **Roesel, K.** 2014. The feeding component in rural and peri-urban smallholder pig systems in Uganda. Poster presented at the Tropentag 2014 Conference on Bridging the Gap between Increasing Knowledge and Decreasing Resources, Prague, Czech Republic, 17–19 September 2014. Nairobi, Kenya. ILRI. <https://cgspace.cgiar.org/handle/10568/43793>
- Grace, D., **Roesel, K.**, Bett, B. and Unger, F. 2014. Healthy lives: Tackling food-borne diseases and zoonoses. Presented at the Tropentag 2014 Conference on Bridging the Gap between Increasing Knowledge and Decreasing Resources, Prague, Czech Republic, 17–19 September 2014. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/44918>



- Makita, K., **Roesel, K.**, Hung Nguyen-Viet, Bonfoh, B., Kang'ethe, E., Lapar, L. and Grace, D. 2014. Food safety issues and scientific advances related to animal-source foods. Presented at the Asia-Pacific Association of Agricultural Research Institutions (APAARI)-Japan International Research Center for Agricultural Sciences (JIRCAS) expert consultation on assuring food safety in Asia-Pacific, Tsukuba, Japan, 4-5 August 2014. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/56885>
- Grace, D., Kang'ethe, E., Bonfoh, B., **Roesel, K.** and Makita, K. 2014. Food safety policy in 9 African countries. Presented at the 4th annual Leverhulme Centre for Integrative Research on Agriculture and Health (LCIRAH) conference, London, UK, 3-4 June 2014. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68865>
- Shija F, Nonga H, Kurwijila LR, **Roesel K**, Grace D and Misinzo G. 2013. Determination of risk factors contributing to microbial contamination in milk and identification of presence of selected pathogenic bacteria along dairy value chain in Tanga. Presentation at the 31st Tanzania Veterinary Association scientific conference, Arusha, Tanzania, 3-5 December 2013. <https://cgspace.cgiar.org/handle/10568/34880>
- Atherstone, C., **Roesel, K.** and Grace, D. 2013. Ebola risk in the pig value chain in Uganda. Presented at the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand Bassam, Côte d'Ivoire, 1-5 October 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/34388>
- Grace, D. and **Roesel, K.** 2013. Gender aspects of informal markets. Presented at the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand-Bassam, Côte d'Ivoire, 1-5 October 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/33806>
- Roesel, K.**, Grace, D., Dione, M.M., Ouma, E.A., Pezo, D., Kungu, J., Ejobi, F. and Clausen, P.H. 2013. Assessment of knowledge, attitudes and practices on pork safety among smallholder pig farmers in Uganda. Poster prepared for the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand-Bassam, Côte d'Ivoire, 1-5 October 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/33796> (Award for best poster)
- Msalya, G., Joseph, E., Shija, F., Kurwijila, L.R., Grace, D., **Roesel, K.**, Haesler, B., Ogutu, F., Fetsch, A., Misinzo, G. and Nonga, H. 2013. An integrated approach to assessing and improving milk safety and nutrition in the Tanzanian dairy chain. Presentation at the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand-Bassam, Côte d'Ivoire, 1-5 October 2013. Morogoro, Tanzania: Sokoine University of Agriculture. <https://cgspace.cgiar.org/handle/10568/34867>
- Ocaido, M., **Roesel, K.** and Grace, D. 2013. Food safety and zoonotic hazards in pig value chains in East Africa. Oral presentation at the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand-Bassam, Côte d'Ivoire, 1-5 October 2013. Makerere, Uganda: Makerere University. <https://cgspace.cgiar.org/handle/10568/34869>

- Dewe, T., **Roesel, K.**, Fekete, A., Legese, G. and Grace, D. 2013. An integrated approach to assessing and improving meat and milk safety and nutrition in the Ethiopian sheep and goat value chain. Presented at the First African Regional Conference of the International Association on Ecology and Health (Africa 2013 Ecohealth), Grand-Bassam, Côte d'Ivoire, 1-5 October 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/33805>
- Dione, M.M., Ouma, E.A., **Roesel, K.**, Mayega, L., Nadiope, G., Kiryabwire, D. and Pezo, D. 2013. Participatory assessment of animal health constraints and husbandry practices in the pig production system in three districts of Uganda. Poster prepared for the 14th International Conference of the Association of Institutions for Tropical Veterinary Medicine (AITVM), Johannesburg, South Africa, 25-29 August 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/33980>
- Shija, F., Misinzo, G., Nonga, H., Kurwijila, L.R., **Roesel, K.** and Grace, D. 2013. The use of polymerase chain reaction (PCR) to confirm presence of selected pathogenic bacteria along milk value chain in Tanga region. Paper presented at the 14th international conference of the Association of Institutions for Tropical Veterinary Medicine (AITVM), Johannesburg, South Africa, 25-29 August 2013. <https://cgspace.cgiar.org/handle/10568/33742>
- Roesel, K.**, Holmes, K., Kungu, J., Grace, D., Pezo, D.Q., Ouma, E.A., Baumann, M., Fries, R., Ejobi, F. and Clausen, P.H. 2013. Fit for human consumption? A qualitative survey at a Ugandan pig abattoir. Oral presentation at the 14th international conference of the Association of Institutions for Tropical Veterinary Medicine (AITVM), Johannesburg, South Africa, 25-29 August 2013. <https://cgspace.cgiar.org/handle/10568/33725>
- Dewé, T.C.M., **Roesel, K.**, Legese, G. and Grace, D. 2013. Lambs to the slaughter: Are meat and milk from small ruminants a health risk to Ethiopians? Poster prepared for the 2013 annual conference of the Society for Veterinary Epidemiology and Preventive Medicine held at Madrid, Spain, 20-22 March 2013. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/32848>

### 3. Conference papers

- Roesel, K.** and Grace, D. 2015. Parasites in food chains. Presented at “Microsporidia in the Animal to Human Food Chain: An International Symposium to Address Chronic Epizootic Disease”, Vancouver, Canada, 9-13 August 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/68006>
- Roesel, K.**, Grace, D., Baumann, M., Fries, R. and Clausen, P.-H. 2015. Food safety in low income countries. Presented at the 15th expert conference on meat and poultry hygiene, Berlin, Germany, 3-4 March 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/61834>
- Roesel, K.**, Ouma, E.A., Dione, M.M., Pezo, D., Grace, D. and Alonso, S. 2014. Smallholder pig producers and their pork consumption practices in three districts in Uganda. Presented at the 6th All Africa Conference on Animal Agriculture, Nairobi, Kenya, 27-30 October 2014. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/51344>

#### 4. Presentations at stakeholder workshops

- Ouma, E., Dione, M., **Roesel, K.**, Lule, P., Kawuma, B., Birungi, R., Asiimwe, G., Opio, F. and Lukuyu, B. 2017. Smallholder pig value chains transformation in Uganda: Results, lessons and insights. Presented at the Uganda Livestock Sector Consultative Meeting, Kampala, 14 March 2017. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/81344>
- Roesel, K.** 2017. Deutsche Veterinärmedizin in Sub-Sahara Afrika: Wo kann unser Beitrag liegen? Beispiele aus der Praxis. Invited presentation at the Berlin Veterinary Association (Berliner Tierärztliche Gesellschaft), Berlin, Germany, 12 April 2017. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/80791>
- Roesel, K.** 2016. Nahrungsmittelsicherheit und informelle Märkte in Afrika südlich der Sahara. Invited presentation at the GIZ expert colloquium, 20 July 2016, Bonn, Germany. <https://cgspace.cgiar.org/handle/10568/79987>
- Roesel, K.** 2015. The role of informal food markets: Towards professionalizing, not criminalizing. Presented at the 16th Annual Meeting of the Inter-Agency Donor Group on Pro-Poor Livestock Research and Development, Berlin, Germany, 18-20 November 2015. Nairobi, Kenya: ILRI. <https://cgspace.cgiar.org/handle/10568/69118>
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### **Disclosure of own share in the body of the work**

The share of the authors who were involved in the publications of this work is listed below according to the following criteria:

1. Idea
2. Design of study and/or experiment
3. Test execution
4. Data analysis
5. Manuscript writing
6. Mobilisation of funding and other resources

#### Publication I

Dione, M. M., Ouma, E.A., Roesel, K., Kungu, J., Lule, P., Pezo, D. 2014. **Participatory assessment of animal health and husbandry practices in smallholder pig production systems in three high poverty districts in Uganda.** *Prev Vet Med.* 2014 Dec 1;117(3-4):565-76. doi: 10.1016/j.prevetmed.2014.10.012. Erratum in: *Prev Vet Med.* 2015 May 1;119(3-4):239.

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#### Publication II

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## Publication III

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## Publication IV

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**Statement of authorship**

Except where reference is made in the text, this dissertation contains no material published elsewhere or extracted in whole or in parts from a dissertation presented by myself towards another degree or diploma. Nobody else's work has been used without due acknowledgement in the main text of the dissertation. This dissertation has not been submitted for an award of any other degree or diploma in any tertiary institution.

Berlin, 30<sup>th</sup> November 2017

Kristina Rösel



