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A survey of *Alaria alata* mesocercariae in slaughter pigs (*Sus scrofa domestica*, Linnaeus, 1758) in the Mekong delta area, Vietnam

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PEER REVIEW

Peer reviewer

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Comments

This is a significant study that shows mesocercariae of *Alaria* spp. are not present in investigated area. The results are based on a well-designed study with relevant information regarding consumer protection.
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ABSTRACT

Objective: To analyze the prevalence of *Alaria* sp. mesocercariae in slaughter pigs in the Mekong Delta in Vietnam.

Methods: From December 2012 to March 2013, 621 carcasses of slaughter pigs originating from nine Vietnamese provinces (69 carcasses per province) bordering the Mekong Delta were tested for the presence of *Alaria alata* mesocercariae. From each carcass, 30 g tissue (a mix of peritoneal fat and cheek tissue) were examined using the *Alaria*-migration-technique. Animals originated from traditional farming with free access of pigs to water/wetlands. Fattening pigs (5.5–7.5 months of age, 84% males) constituted 97.1% of the sample set, whereas sows >26 months accounted for 2.9%.

Results: All samples tested were negative for motile mesocercariae.

Conclusions: Results indicate that the actual prevalence of this parasite will not exceed 5% ($\alpha=0.05$) in pigs from the study area. Further studies could elucidate if this is due to (unapproved) antimollusc and antiparasitic treatment in the study area or if the parasite is absent in the wildlife at all.

KEYWORDS

Foodborne parasite, *Alaria alata*, Domestic pigs, Vietnam, Risk factors

1. Introduction

Alaria sp. are trematodes with a life cycle that involves two intermediate water bound hosts[1]. The final hosts are carnivores, which are infested by oral uptake of intermediate

hosts carrying mesocercariae. In other vertebrates, ingested mesocercariae will not complete their life cycle, but migrate through the body, with a tendency to accumulate in the anterior body. Such an accumulation of parasitic stages in motile and not-waterborne hosts might ultimately facilitate the spread of

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the parasite. Consumption of raw or undercooked meat carrying mesocercariae, namely, *Alaria americana*, has been associated with human pulmonary and ocular disease, and a zoonotic potential is assumed for other *Alaria* species also[1]. Number of reported human cases is admittedly low, compared to other waterborne, snail-mediated zoonotic helminthoses.

Alaria are widely distributed on the Eurasian continent. There is an increasing number of research done on the prevalence of *Alaria alata* (*A. alata*) in (wild) carnivores, and also on wild boar as an abundant paratenic host, which is used for meat production. Although mesocercarial stages of *A. alata* have first been described in domestic pig rather than wild boar, modern farming practices will minimize contact of domestic pigs to intermediate hosts. A different situation could be experienced under conditions of extensive or combined farming in suitable biotopes. Such a combination would be expected under the combined small-scale farming, fish-raising and pork-fattening (Vuon-Ao-Chung) farming system as it is common in southern regions of Vietnam[2]. Fishborne zoonotic trematodes have been studied in the Mekong Delta area, but there is no report on the snail-mediated *Alaria* sp[3,4]. The similarity in the biology of trematodes and, more specifically, the presence of intermediate hosts from Planorbidae family, *i.e.* at least *Indoplanorbis* sp. indicates that *Alaria* could be present in the study area. Although it can be argued that a systematic study on this parasite might focus on the presence of parasitic stages in first-intermediate or on adult stages in definite hosts, we choose a food-safety oriented approach by testing potential paratenic hosts, *i.e.* pigs, which contribute about 3/4 of the meat component in the human diet in Vietnam[5].

2. Material and methods

2.1. Sampling plan

In the period from December 2012 to March 2013, samples were taken from carcasses of slaughter pigs during the course of meat inspection. Animals originated from nine provinces bordering the Mekong River, *i.e.* Long An, Tien Giang, Ben Tre, Dong Thap, Can Tho-Vinh Long, Tra Vinh, An Giang, Soc Trang and Kien Giang. Sample size per province was $n=69$, calculated from $n = [1 - (1-p)^{1/d}] * (N-d/2) + 1$ [6], where p is the probability of finding at least one positive case, N the population size (in this case the number of slaughtered pigs/year, ranging from 160516 to 645569 heads, according to province, 2012 data), and d the number of affected animals in the population. A 5% prevalence was assumed with 5% accepted error, 95% confidence interval, and sample size was adjusted for 100% specificity and 85% sensitivity[7]. Animals originated from small farms (<20 heads) with low biosecurity. Such farms deliver 97% of slaughter pigs in these provinces[8]. Sampling was done at the largest slaughterhouses (1–3 plants with a capacity of 50–500 heads per night) in each province, on one day per slaughterhouse. The areas from which the pigs originated were visited and information of the use of molluscicides in rice fields as well as of anthelmintics in pig fattening was gathered in informal talks with farmers.

From each carcass, about 50 g mixed tissue from the cheek (comprising roughly equal parts of glandular, adipose and muscle tissue), and 50 g peritoneal fat were taken to represent a sample unit and kept at 0–4 °C until analysis, which was done within 7 d after slaughter. Samples were accompanied by

information on age, gender and origin of pigs.

2.2. Examination of samples

From each sample, equal parts of cheek and peritoneal tissues were combined to give 30 g, then chopped to cubes of about 5 mm and tested for motile mesocercariae of *A. alata* by means of a migration technique[7]. Presumptive mesocercariae were identified by their motility and morphology[1].

3. Results

From none of the 621 samples, motile objects were recovered by the *Alaria* migration technique. Fattening pigs (5.5–7.5 months of age, 84% males) constituted 97.1% of the sample set, whereas sows >26 months accounted for 2.9%. The sampling plan was adjusted to represent small farms with low biosecurity, where the presence of this parasite would be more likely. The result indicates that in the study period, the “true” prevalence at province level would not have exceeded 5%, with 95% confidence.

4. Discussion

4.1. Detection of parasitic stages and interpretation of the result with regard to food safety

Results indicate that the prevalence of the parasitic stages in fattening pigs in the study area and in the study period would not exceed 5%. To the knowledge of the authors, there are no comparable studies in pig populations in Asian countries, and also no recent surveys in domestic pigs in Europe or in other continents. For free-ranging wild boars in Europe, a number of studies report average frequencies around or below 5%, but there is both spatial and temporal clustering[9–13]. The exposure of consumers to viable mesocercariae is not only determined by the presence of such parasites in pork, but also on the effect of culinary preparation. It should be noted that not only cooked, but also raw pork is commonly consumed in Vietnam[5].

4.2. Factors which might have influenced the results

Whereas the sampling plan was designed to represent a certain type of pork production, the habitat characteristics were suitable for snail-mediated trematodes and the currently most sensitive available method of detection was applied on a mix of typical predilection sites[7]. Some other factors need to be considered when interpreting the results. Seasonal variations in detection of mesocercariae in pigs and wild boars have been reported, insofar as detection rates in wild boar should increase when the abundance of frogs increases[11]. Whereas the average temperature in the Mekong Delta shows only minor seasonal variation, there is a distinct “rainy season” from May to October[14]. In our study, sampling was done after the “rainy season” and it is conceivable that in the rainy season, pigs would have more contact to frogs.

Informal talks with farmers gave evidence of the unapproved use of anthelmintics in pig fattening as well as the application of niclosamide and metaldehyde on rice fields. Such measures could interrupt the life cycle of *Alaria*, if established in the study region at all.

4.3. Conclusion

In the present study, *Alaria* sp. mesocercariae could not be recovered from any of the 621 slaughter pigs originating from nine provinces bordering the Mekong Delta in South Vietnam. This was not expected, as a variety of mostly fishborne trematodes are enzootic in this area and habitat characteristics, type of pig farming and presumed presence of specific intermediate hosts would favour the occurrence of this parasite. This finding is satisfactory from the viewpoint of consumer protection, but there might be a bias due to (uncontrolled) use of antiparasitics or pesticides. Further research might be conducted on abundance and infestation of intermediate and definite hosts.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgements

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Comments

Background

A. alata is a trematode widely distributed on the Eurasian continent. Its zoonotic potential as a foodborne parasite is contradictorily discussed. Actually there is a lot of research regarding this parasite and its paratenic stage. No information exists about the occurrence of *A. alata* mesocercariae in Vietnam.

Research frontiers

The aim of the study was to confirm the presence of the parasite in a defined geographic area and to investigate its prevalence in a paratenic host which is important for the human food chain.

Related reports

As the two intermediate hosts are present in the area, it is a surprise that mesocercariae of *Alaria* spp. could not be confirmed in the investigated paratenic host. Presumptive mesocercariae were identified by their motility and morphology.

Innovations & breakthroughs

The sampling plan was adjusted to represent small farms with low biosecurity, where the presence of this parasite would be more likely. The result indicates that in the study period, the “true” prevalence at province level would not have exceeded 5%, with 95% confidence.

Applications

Although it can be argued that a systematic study on this parasite might focus on the presence of parasitic stages in

first–intermediate or on adult stages in definite hosts, we chose a food–safety oriented approach by testing potential paratenic hosts, *i.e.* pigs, which contribute about ¾ of the meat component in the human diet in Vietnam.

Peer review

This is a significant study that shows mesocercariae of *Alaria* spp. are not present in investigated area. The results are based on a well–designed study with relevant information regarding consumer protection.

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