Barbosa et al., J Vet Sci Med Diagn 2017, 6:4
DOI: 10.4172/2325-9590.1000237

Journal of Veterinary Science & Medical Diagnosis

Research Article

A SCITECHNOL JOURNAL

Identification of *Macracanthorhynchus Hirudinaceus, Stephanurus Dentatus* and *Trichuris Suis* in Native Pigs on Marajó Island

José Diomedes Barbosa[1]*, Jenevaldo Barbosa da Silva[2], Alessandra dos Santos Belo Reis[1], Henrique dos Anjos Bomjardim[1], David Driemeier[3], Felipe Masiero Salvarani[1], Cairo Henrique Sousa de Oliveira[4], Carlos Magno Chaves Oliveira[1], Marilene de Farias Brito[5]

Abstract

Technification of pig farming in recent decades has led to a significant reduction in the occurrence of helminth parasitism. However, the production of pigs in free-range systems is still a common reality in rural areas of Brazil, such as Marajó Island, in the State of Pará, a Brazilian Amazonian biome. Therefore, the present work describes the main parasitological and anatomopathological findings of six pigs in a sample of 23, of different age groups, raised under free-range conditions on native pastures on Marajó Island. During slaughter, the presence of *Stephanurus dentatus* in the ureters and renal pelvis, *Macracanthorhynchus hirudinaceus* in the small intestine and *Trichuris suis* in the cecum and colon was detected in these six pigs. The histopathology results showed different types of lesions, including mild granulomatous infiltrates in the lymph nodes, granulomatous inflammation and brown pigment in the tonsils, hyperkeratosis in the esophagus and non-glandular stomach, lung with dark pigment in the bronchioles and intra-alveolar macrophages, as well as squamous metaplasia of the peribronchial glands, hemosiderosis in the spleen, liver with marked extramedullary hematopoiesis, mild hepatitis and abscess caused by parasitic migration with eosinophils, intestinal edema and neuronal lipofuscinosis in the spinal cord. From these findings, it can be inferred that free-range pigs on Marajó Island are susceptible to helminths, reflecting the high degree of environmental contamination in which these animals are reared, and that such ecosystems can function as reservoirs of these agents. Helminth infections lead to losses in pig farming in the Amazon biome, showing the importance of the strategic control of these parasites.

Keywords:

Helminths; Pig farming; Amazon; Parasitism; Pathology

Introduction

High occurrence and intensity of gastrointestinal helminth infection has been a strong trend in pig herds kept under a traditional management system. In this context, it is common to find between 5 and 10 species of helminths and high levels of infection in free-range reared pigs [1-3]. However, in "technified" and industrial pig farming systems, this scenario is no longer observed due to the adoption of a series of preventive hygienic-health measures [4,5].

Brazil is the fourth largest producer and exporter of pork in the world [6]; however, the production of pigs in free-range systems is still a common reality in rural and riverine areas of the country, especially in specific ecosystems such as the Mato Grosso and Mato Grosso do Sul Pantanal regions, the Maranhão lowlands and Marajó Island. Pigs raised in free-range systems in the country are generally managed without the use of anthelmintics, food supplementation, vaccination or routine veterinary medical follow-up, in a purely extractive system. Thus, although the epidemiology of the major diseases in these pigs is not known, high levels of helminth infection are expected at all stages of production.

The objective of this work was to describe the necropsy findings and to identify the main species of helminths and the microscopic changes caused by helminths in pigs farmed in an extensive production system on Marajó Island, State of Pará, Brazil.

Materials and Methods

A clinical examination of 23 pigs of both sexes, ranging from five months to two years of age, was conducted extensively on native pastures, belonging to small properties located in the municipality of Cachoeira do Arari, Marajó Island, State of Pará, Brazil. Data on the nutritional status, origin and clinical history of all pigs were organized in individual records.

Of the 23 pigs in the sample, six were slaughtered by the property owners, in the places of origin, for household consumption. For the study, the animals were identified as one to six, being five females (four two-year-old pigs and one six-month-old pig) and a seven-month-old male pig. From the six pigs, fragments of injured organs were collected and fixed in 10% formalin and then sent for histopathological analysis in the Veterinary Pathology Sector of the Federal University of Rio Grande do Sul. Endoparasites were collected and stored in individual labeled jars with 70% ethanol fixative solution for later classification at the Veterinary Medicine Institute of the Federal University of Pará.

The parasite load was classified as absent, mild, moderate or marked, according to the number of parasites found. The same criterion was adopted to classify the intensity of the macroscopic findings, according to the severity of the lesions, as absent, mild, moderate or marked.

Results

The pigs evaluated according to the history provided by the farmers were raised in a free-range system (Figures 1 and 2), together with other animal species (cattle, horses, buffalo, sheep and chickens), without adequate nutritional and health management, and had never received treatment against ecto and endoparasites or mineral supplementation. The animals fed on carcasses, insects and native grass. Clinical examination showed retarded growth and poor nutritional status. The two young pigs that were slaughtered were found mired next to puddles of still water, with obvious weakness and malnutrition.

***Corresponding author:** José Diomedes Barbosa Neto, Instituto de Medicina Veterinária, Campus de Castanhal, Universidade Federal do Pará, Rodovia BR-316 km 61, 68741-740, Castanhal, PA, Brazil, Telephone: +9137211686 ; Fax: +9137211686; E-mail: diomedes@ufpa.br

Received: August 23, 2017 **Accepted:** September 08, 2017 **Published:** September 12, 2017

Figure 1: Native pigs, reared on a free-range system on Marajó Island, Pará, with parasitic infection and poor nutritional status.

Figure 2: Native pigs with parasitic infection and poor nutritional status, reared on a free-range system on Marajó Island, Pará, found mired.

Tables 1 and 2 describe the parasite load and macroscopic findings of the six pigs from a sample of 23 animals from Marajó Island.

At slaughtering, it was observed that 50% (3/6) of the animals (Pigs 1, 4 and 5) had small nodules in the serosa of the small intestine (Figure 3) and that the mesenteric lymph nodes were enlarged. In the intestinal lumens of the same pigs, 30 helminths on average, with sizes ranging from 2 to 30 cm in length, were found attached to the mucosa (Figures 3-5), and the nodules found in the serosa coincided with the point of proboscis attachment of the parasitic worms. In some segments of the small intestine, dilatation and partial obstruction of the intestine (Figure 6) were observed due to infestation of worms in the lumen; this parasite was identified as *Macracanthorhynchus hirudinaceus* (Figures 3 and 6).

Additionally, during the necropsy analysis, a high parasite load of *Stephanurus dentatus* nematodes on the ureters and renal pelvis (Figures 7 and 8) was observed in 66.6% (4/6) of the animals (Pigs 1, 2, 3 and 5). Abscesses were observed on the ureter wall (Figure 9), renal pelvis (Figure 10), renal capsule, bladder, liver and lung. One of the animals (Pig 5) presented lesions caused by parasitic migration.

Macroscopic alterations were observed, such as small areas of calcification in the lung, nodules adjacent to the ureters, bladder and kidneys, which presented a whitish caseous mass when sectioned

Table 1: Parasite load in slaughtered pigs on Marajó Island, Pará.

Identification	*S. dentatus*	*M. hirudinaceus*	*T. suis*
Pig 1	+++	+++	++
Pig 2	++	-	++
Pig 3	++	-	-
Pig 4	-	++	-
Pig 5	+++	++	-
Pig 6	-	-	++

Parasite load: absent = -, mild = +, moderate = ++, marked = +++

Table 2: Macroscopic findings in slaughtered pigs on Marajó Island, Pará.

Identification	*S. dentatus*	*M. hirudinaceus*	*T. suis*
Pig 1	+++	++	-
Pig 2	++	-	-
Pig 3	++	-	-
Pig 4	-	++	-
Pig 5	+++	+	-
Pig 6	-	-	-

Lesion: absent = -, mild = +, moderate = ++, marked = +++

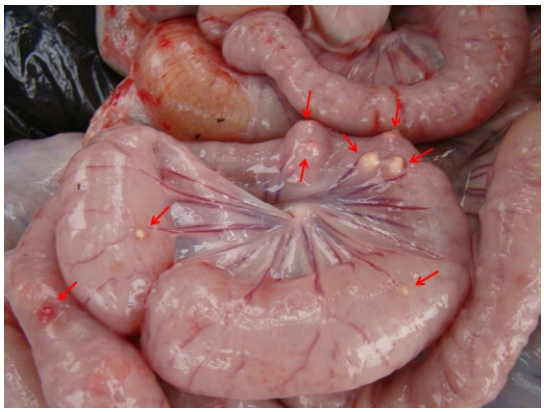

Figure 3: *Macracanthorhynchus hirudinaceus* infection nodules (arrows) in the intestinal serosa, also called “blackberry spots”.

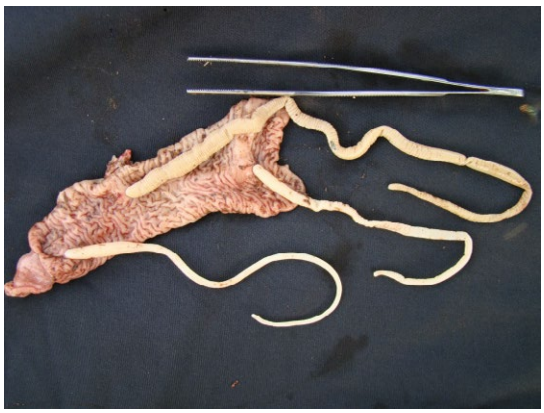

Figure 4: *Macracanthorhynchus hirudinaceus* with proboscis attached to the mucosa of the small intestine, with a thick, long and transversely wrinkled body.

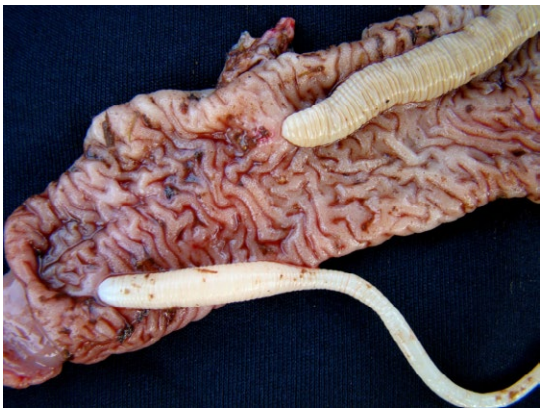

Figure 5: Details of Figure 4 and evidence of the fixation of the proboscis of the parasites to the mucosa of the small intestine.

Figure 6: Several worms attached to the mucosa, causing dilatation and partial obstruction of the small intestine

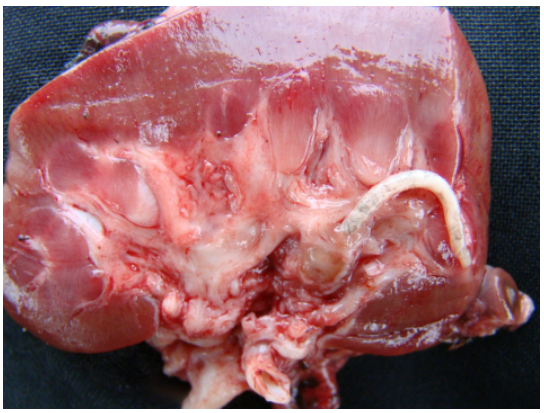

Figure 7: Presence of nematodes in the renal pelvis of the pig.

(Figures 9 and 10), liver slightly yellow with small abscesses, edema in the mucous membranes of the stomach and intestine, enlarged mesenteric lymph nodes, abscesses in the renal capsule, and a whitish mass near the kidney and renal pelvis.

Trichuris suis was found in the cecum and colon of 50% (3/6) of the animals (Pigs 1, 2 and 6). There were no significant macroscopic lesions related to this parasite, and the histopathological findings of the pigs with *Trichuris suis* were restricted to mild edema in the large intestine, with mixed infiltrate in the mucosa, macrophages and hemosiderin.

Table 3 shows the histopathological lesions observed in each of the six animals sampled on Marajó Island, with lesions detected in various organs and tissues.

The main histopathological findings of pigs with *M. hirudinaceus* were edema and mixed infiltrate in the small intestine mucosa and lymph nodes with mild macrophage infiltrate. In the pigs with *Stephanurus dentatus,* ureter with nematodes, abscess in the wall of the ureter, cystic area in the adrenal gland and abscesses in the kidneys were observed. In the liver, mild, moderate and marked hepatocellular swelling, abscess caused by parasitic migration with eosinophils, marked extramedullary hematopoiesis and mild hepatitis were observed. In the tonsils, lymphoid hyperplasia, the

Figure 8: Specimens of Stephanurus dentatus.

Figure 9: Abscess in the lumen of the pig ureter.

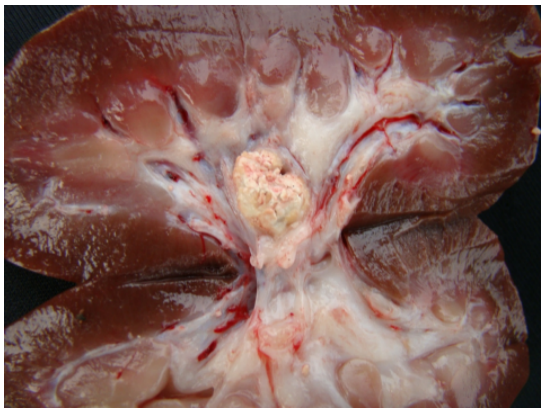

Figure 10: Abscess in the renal pelvis of the pig.

Table 3: Histological findings in pigs slaughtered on Marajó Island, Pará.

Identification	Description of the main lesions observed under microscopy
Pig 1	Liver with mild hepatocellular swelling, tonsils with lymphoid hyperplasia and macrophages with dark pigment, ureter with parasites and formation of abscess; mild edema in the submucosa of the large intestine. Small intestine, glandular stomach, heart, brain, cerebellum and lung without histopathological changes.
Pig 2	Submucosal edema and mixed infiltrate in the large intestine, mild macrophage infiltration in the lymph nodes, liver with moderate hepatocellular swelling. Duodenum, heart, kidney, lung, brain, cerebellum, spinal cord and spleen without histopathological changes.
Pig 3	Mild granulomatous infiltrate in the lymph nodes, hyperkeratosis in the non-glandular stomach, large intestine with mixed infiltrate in the mucosa, with mild edema, macrophages and hemosiderin; ureter with nematodes, cystic area in the adrenal gland, granulomatous inflammation area and brown pigment in the tonsils, neuronal lipofuscinosis in the spinal cord, hemosiderosis in the spleen, lung with dark pigment in the bronchioles and intra-alveolar macrophages. Glandular stomach, thyroid, bladder, small intestine, cerebral cortex, esophagus, kidney, heart, liver and pancreas without histopathological changes.
Pig 4	Lung with hypertrophy of muscles of bronchioles, nematode parasites in bronchial lumen, squamous metaplasia of peribronchial glands; liver with marked extramedullary hematopoiesis, mild hepatitis and abscess caused by parasitic migration with eosinophils; lymph nodes with mild macrophage infiltration in follicular centers, with microabscesses and granulomatous infiltrate. Cerebellum, brain, heart, tonsil, adrenal, glandular stomach and kidneys without histopathological changes.
Pig 5	Abscess on the wall of the ureter; macrophages with dark pigment and microabscesses in the tonsils; abscess with large numbers of eosinophils and marked hepatocellular swelling in the liver; abscess in the kidney; spleen with hemosiderosis. Adrenal, pancreas, lung, brain, cerebellum, salivary gland, duodenum, small intestine and bladder without histopathological changes.
Pig 6	Edema in the large intestine, mild hyperkeratosis in the esophagus, macrophage infiltrate in the small intestine mucosa, hydropic degeneration with inclusion body and neutrophil infiltrate in the non-glandular stomach, hemosiderosis in the spleen, microabscess in the tonsil. Heart, kidney, brain and spinal cord without histopathological changes.

presence of macrophages with dark pigment, microabscesses, areas of granulomatous inflammation and brown pigment were observed. In the lymph nodes, mild macrophage infiltrate with microabscesses and mild granulomatous infiltrate occurred. In the lungs, dark pigment in the bronchioles and intra-alveolar macrophages, bronchial muscle hypertrophy, parasites in the bronchial lumen, squamous metaplasia of the peribronchial glands, hemosiderosis in the spleen and neuronal lipofuscinosis in the spinal cord were observed.

Discussion

The results of the present work show the challenges that helminths pose to small producers who raise free-range pigs. Although pig helminths are rare in industrial farms today, this disease still causes severe losses in family farms in various tropical regions [7]. The macroscopic and histopathological findings of this study described pigs with a high helminth parasite load, a fact that is not commonly observed in other regions of the world, even those in which pigs are reared in an intensive free-range system (SISCAL). In the present study, the helminths found were *Stephanurus dentatus*, *Macracanthorhynchus hirudinaceus* and *Trichuris suis*.

The industrialization of Brazilian pig production in the last decades has limited the occurrence of most species of helminths; however, pig production in free-range systems, which are more rustic, without any hygienic-health and food control, is still a common reality in rural areas of Brazil. This is especially true in biomes such as Marajó Island, the Maranhão lowlands and the Mato Grosso and Mato Grosso do Sul Pantanal regions. According to Costa et al. [8], this parasitic disease is not common in "technified" production systems and occurs only in animals raised in SISCAL or extensive production systems, as on Marajó Island.

Macracanthorhynchus hirudinaceus is a round acanthocephalan parasite with a body and a proboscis formed by retractile spiny rings by which it attaches to the wall of the small intestine of its host; its evolutionary cycle is indirect. The worm has a thick, long and transversely wrinkled body and no digestive tract; nutrients are absorbed through the integument. Males measure approximately 10 cm, and females are up to 35 cm in length; females release numerous highly resistant, long-lasting eggs that are infective to pigs only after development for two to three months inside beetles. The coprophagous beetles of the *Scarabaeidae* family are carriers of infective larvae and act as intermediate hosts. Swine are infected when they ingest an infected larva or beetle. The attachment of the larva to the wall of the small intestine produces nodules that are also called "blackberry spots". Severe infestations by *M. hirudinaceus* decelerate growth and cause weight loss [9,10], as observed in the animals in this study, which had poor nutritional status. Infections can also result in perforation of the intestinal wall and peritonitis [9-11]. On Marajó Island, adverse environmental conditions, such as floods where land is flooded and muddy, and food shortages are other factors that may have contributed to animal weight loss and may also have increased proliferation and transmission of the parasite by ingestion of the beetle that transmits that helminth. High parasite loads of these helminths can be observed in periods of high temperature, especially in tropical and subtropical regions, a condition compatible with that of the Marajó Island biome. According to Radostits et al. [9] and Souza et al. [10], nodules in the serosa of the small intestine, known as "blackberry spots", are common findings in necropsies of pigs infested with *M. hirudinaceus*; these nodules occur due to the fixation of the spiny rings of the helminth proboscis in the intestinal mucosa. Histological examination revealed edema and mixed infiltrate in the mucosa of the small intestine, findings similar to those of Souza et al. [10]. *Stephanurus dentatus*, of the Stephanuridae family, is a stout parasite that is approximately 2 mm by 40 mm long, found in the hepatic, renal and perirenal tissues, axial musculature and spinal canal of pigs and, sometimes, cattle. The buccal cavity is cup-shaped and is directed straightforward, with 6 to 10 triangular teeth at its base. The gut is convoluted, the spicules are equal and short, and the bursa is reduced [12]. This parasite is a problem in countries with a hot climate [13]. The evolutionary cycle may be direct or may involve worms as facultative intermediate hosts. Infection occurs by ingestion or skin penetration of third-stage larvae or by oral ingestion of infected worms that act as paratenic hosts [12]. *Stephanurus dentatus* is a less prevalent helminth because it has a long life cycle and its cycle has been disrupted by the industrialization of pig production in most of the world. This helminth has a prepatent period ranging from nine to 16 months. A large number of eggs are eliminated along with the urine. In the environment, third-stage infective larvae may develop within a few days. Marajó Island has the peculiarity of presenting high temperatures and large marsh areas for a long period of the year, which may represent an ecological benefit for maintenance of the parasite in the environment because the paratenic host, earthworms

have affinity for moist soils. The macroscopic findings observed in the pigs of the present study are in agreement with those of Bowman et al. [12], who reported the formation of abscesses in the liver due to the erratic, destructive migration of the larvae of *Stephanurus dentatus* to this organ, which the larvae migrate through for four to nine months. According to McGavin et al. [13], adult worms usually encyst in the perirenal fat; however, some parasites may reside in the kidneys, as observed in the pigs of this study. The histopathological findings in the six pigs were similar to those reported by Bowman et al. [12], i.e., some larvae of *Stephanurus dentatus* are trapped by an encapsulating tissue reaction and the rest migrate to the retroperitoneal tissues surrounding the kidneys and ureters. The hepatitis observed in the liver of these pigs, similar to the findings of McGavin et al. [13], occurs due to the migration of parasite larvae.

Trichuris is characterized by a whip-shaped body with a thin anterior portion that is usually completely buried into the mucosa of the large intestine, while the posterior part is stout and is typically free in the intestinal lumen [14]. *T. suis* infection presents some biological characteristics in common with *Ascaris suum*. Both have a direct life cycle with a prepatent period of six to eight weeks. Larvae develop inside highly resistant eggs and remain infective to pigs for up to 11 years. Infection induces strong immunity. However, the egg embryonation progresses slowly. Although *T. suis* eggs possess a high survival potential, egg inviability may be elevated under certain conditions, possibly due to biotic and abiotic factors in the soil environment [15]. In most infestations, a low number of adult *T. suis* are present in the cecum and colon, but on some occasions, the parasite load of the worm can be high and cause weakness and death of the animals. In the pigs studied, this parasite had lower severity compared to the parasite load and the lesions found in *Macracanthorhynchus hirudinaceus* and *Stephanurus dentatus* infections; however, Jubb et al. [16] note that if there are enough *T. suis* worms, mucohemorrhagic typhlocolitis may occur in clinical association with anorexia, diarrhea, dysentery, dehydration, malnutrition and, in some cases, death. Such a severe clinical condition, attributed only to *T. suis*, was not observed in the pigs studied here. Those authors also reported that the mucosa becomes thickened, edematous, reddish, eroded, and with increased secretion of mucus, which was not observed in the pigs of our study.

Conclusions

Pigs raised in a free-range system in the country, such as on Marajó Island, are generally managed without the use of anthelmintics or food supplementation and without vaccination or routine veterinary monitoring. This low management approach facilitates the occurrence of helminths that are practically extinct in modern pig farming. As a consequence of not using the various anthelmintics available on the market, these environments function as a genetic reservoir of sensitive worms. Thus, in the near future, with the spread of anti-helminth resistance throughout the world, these areas will become refuges of sensitive worms for studies and tests of anthelmintics that work in populations of animals still non-resistant. Thus, it is necessary to know the occurrence of the main helminths in these regions of Brazil to formulate adequate control strategies. In this type of parasitism, control strategies for free-range pigs should be based on the adoption of management techniques that aim to interrupt the helminth cycle, which are aided by the strategic-tactical use of anthelmintics.

Conflict of interest statement

None of the authors of this work has a financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of the paper.

Acknowledgements

We thank the Coordination for the Improvement of Higher Level of Education Personnel (CAPES) foundation and the National Council of Technological and Scientific Development (CNPq) for their financial support.

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Author Affiliation

Top

[1]*Instituto de Medicina Veterinária, Campus de Castanhal, Universidade Federal do Pará, Rodovia BR-316 km 61, 68741-740, Castanhal, PA, Brazil*

[2]*Instituto de Ciências Agrárias, Universidade Federal dos Vales do Jequitinhonha e Mucuri, Avenida Vereador João Narciso 1380, 38610-000, Unaí, MG, Brazil*

[3]*Escola de Medicina Veterinária da Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves 9090, Porto Alegre, RS, Brazil*

[4]*Setor de Medicina Veterinária Preventiva, Escola de Veterinária e Zootecnia, Universidade Federal de Goiás, Campus Samambaia 74690900, Goiânia, GO, Brazil*

[5]*Instituto de Medicina Veterinária, Universidade Federal Rural de Rio de Janeiro, Br 465, Km 7, 23890-000, Seropédica, RJ, Brazil*