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#### EXPERIMENTAL INFECTION OF CATTLE WITH EGGS OF Taenia solium (Infecção experimental de bovinos com ovos de Taenia solium)

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**RESUMO** – O estudo foi realizado em Piraquara, Paraná, Brasil entre 1994 a 1996. Foi avaliada a possibilidade de bovinos terem participação no ciclo de *Taenia solium*. Uma cadeia de seis proglótes grávidas de *T. solium* foi obtida de paciente não medicado. Bovinos da raça *Holstein* foram infectados com 40.000 ovos de *T. solium*. A taxa de infecção foi de 100%. Um total de 44 cisticercos foi recuperado depois de 160 a 570 dias, 18 (40,9%) vivos e 26 (59,1%) calcificados. Dos 44 cisticercos, 19 (43,2%) foram encontrados na musculatura esquelética: 10,5% na cabeça, 36,8% nos membros torácicos, 10,5% no diafragma, 42,1% nos membros pélvicos; e 25 (56,8%) nos órgãos: 80% no fígado, 8% no coração, 8% na língua e 4% nos rins. Cisticercos vivos foram encontrados com acúleos rudimentares, com número variável de 01 a 42 e com 25,4-35,7 µm de comprimento. Os resultados sugerem que bovinos poderiam servir como hospedeiros intermediários da *T. solium*. É necessário continuar os estudos para verificar a infectividade de cistos que se desenvolvem em hospedeiros não habituais.

Palavras chave: Taenia solium, infecção experimental, bovinos, cisticercose.

**ABSTRACT** – A study was performed in Piraquara, Parana, Brazil during 1994-1996 to evaluate the possibility of bovines participation in the epidemic chain of the *Taenia solium*. Six gravid segments of *T. solium* were obtained from a non medicated patient. Holstein bovines were infected with 40,000 eggs of *T. solium*. The infection rate was 100%. A total of 44 cysticerci were recovered: 18 (40.9%) alive and 26 (59.1%) calcified. Of the 44 cysticerci, 19 (43.2%) were found in the skeletal musculature: 10,5% in the head, 36.8% in the anterior members, 10.5% in the diaphragm, 42.1% in posterior members and 25 (56.8%) in the organs: 80% in the liver, 8% in the heart, 8% in the tongue and 4% in the kidneys. Living cysticerci had 1 to 42 short hooks of 25,4 to 35,7 µm. The results suggested that bovines could serve as intermediary host for *T. solium*.

Key words: Taenia solium, cattle, cysticercosis.

#### Introduction

In the State of Paraná, *Taenia saginata* and *Taenia solium* are endemic. The frequency of tapeworm carriers in the municipal districts of Barracão, Salgado Filho, Rio Branco do Sul and Tijucas do Sul, defined by *Taenia* spp. eggs in feces is 1,02, 1,43, 4,6 and 4,3%, respectively (ARRUDA *et al.*, 1990; KOPP *et al.*, 1994). The number of people with neurocysticercosis was high of 51,694 computerised tomographies performed during, 1988-1992, 2,460 (4.76%) had images compatible with living or calcified cysticerci (GUSSO,1997). In Curitiba, there were 9,94%

computerised tomographies showing neurocysticercosis (GRACIA, 1994). During the last three decades, pig breeding received great technological transformations with increase of confinement systems, resulting in the decrease from 3,0 to 0,03% of condemned carcasses for cysticercosis in official inspection (MINOZZO,1997). However, this number may be higher because still today a great number of animals is create in an extensive way, in direct contact with human consumed in the rural areas faeces and without inspection. Researches accomplished in the communities of Postinho (Tijucas do Sul) and Tigre (Rio Branco do Sul) revealed respectively 12,8 and 27,8%, of pigs with cysticercosis (ARRUDA et al., 1990: SCHNEIDER et al., 1994).

Several authors reported that the pigs is the most important intermediary host of the *T. solium*. However, several other domestic

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and wild animals such as cattle could also be intermediary host of T. solium (SOULSBY, 1968; BORCHERT, 1981; LAPAGE, 1981). FAN et al. (1989, 1990a, 1990b, 1992a, 1992b) reported pigs infected artificially and naturally with Т. saginata-like. The consumption of raw or insufficiently cooked cattle meat in Brazil, for cultural or ethnic habits, is more frequent than of pigs one. The objective of this study was to evaluate the infection of cattle with the eggs of T. solium is order to find their role in the epidemic chain of illness.

## **Materials and Methods**

Characterisation of the Animals and Handling. Six Holstein cows between 3 and 18 months of age were separated in two groups. These were obtained from the farms with good sanitary conditions and were not infected with eggs of *Taenia* spp. They were kept order the same environmental conditions of sanitary and alimentary handling. During 60 days the animals were fed with milk and hay and water from public water supply was provided for drinking. In the week immediately after the inoculation, they were confined in individual stalls. In the remaining period, the animals were in available vegetation in CPPI, composed by grass and leguminous.

Obtaining of segments and of eggs. A chain of six gravid segment of *T. solium* was obtained from patient not medicated with anthelmintic, whose faeces were guided for the Laboratory of Parasitology of the Clerkship of Health of the Municipal city of Curitiba, Paraná, Brazil. The segment was identified, broken and eggs obtained were divided in aliquots of 40,000 each. The preparation and dilution was in solution of NaCl 0,09%, in volumes of 40ml. The material obtained was maintained for 10 days in rehearsal tubes, under refrigeration (2-8°C).

*Experimental infection.* For the infection of the animals, the aliquots were aspired with syringes of 20ml and administered orally. For five days, the animals were confined in the stable of CPPI with hod and individual drinking fountain. During the period the supplied feeding was hay and ration.

*Necropsy.* For the discount, the bovine ones were divided in groups of two (an infected and a control), with intervals of pre-patent period of 90, 104 and 111 days. These were taken to butcher shop according to the official norms of inspection. All carcasses and organs were transported and refrigerated then, were inspected, for small slice, the recovery and the identification of the cysticerci. Courts of muscles and organs were made parallel to each other 0.5-1.0 cm.

Study of cysticerci. For evagination of cyst, gastric juice of bovine was used for 45 minutes. Soon after, they were transferred in bovine billiard juice for 15 min, and finally read in microscope. The cysticerci was classified as living (mobile larvae) and calcified. The cysticerci was destined to the making of sheets for optic microscopy, when it was proceeded the measure of the width and length of the protoscolices, the diameter of the rostellum, the length of the hooks, the diameter of the suckers and counting the number of hooks crowns.

## **Results and Discussion**

The control group (three animals) was negative. Of the three bovine infected with eggs of *Taenia solium*, 44 cysticerci were recovered, being 18 (40,9%) alive and 26 (59,1%) calcified. The recovery was of 2-23/40.000 inoculated eggs (TABLE 1).

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Code/animal	Recovered cysticerci nº/40.000 inoculated eggs	Number of Alive cyst	Number of Calcified cyst
01	23	10	13
02	19	07	12
03	02	01	01
TOTAL	44	18	26

TABLE 1 – CYSTICERCI RECOVERED IN EXPERIMENTAL INFECTED BOVINE WITH EGGS OF Taenia solium.

A anatomical distribution of the cysticerci went 19 in to skeletal musculature, being 10,5% in the head, 36,8% in the anterior members, 10,5% in the diaphragm, 42,1% in posterior members and 25 in the organs, being 8% in the language, 8% in the heart,

80% in the liver and 4% in the kidneys. No cyst was found in central nervous system (TABLE 2). About 350 stain (white spots) was found in the liver of the bovine with 581 days of age, what was not observed in the animals with 283 and 294 days of age. The width and medium length of the protoscolices were  $960,4\mu m$  and

975,6µm respectively.

The mean diameter of the *rostellum* length of the hooks were 254 x 27  $\mu$ m respectively. The suckers presented mean diameter of 331,3 $\mu$ m. The number of hooks crowns for cysticerci was of two (TABLE 3).

TABLE 2 – ANATOMICAL DISTRIBUTION OF CYSTICERCI IN EXPERIMENTAL INFECTED BOVINE WITH EGGS OF Taenia solium.

Code	Muscles					Organs			
animal	Head	Anterior Member	Diaphragm	Posterior Member	Heart	Tongue	Brain	Liver	Kidney
01	0	04	01	05	02	0	0	11	0
02	02	03	0	02	0	02	0	09	01
03	0	0	01	01	0	0	0	0	0
Total	02	07	02	08	02	02	0	20	01

The pigs is the habitual intermediary host of the *Taenia solium* (SMYTH,1994). Though, FAN *et al.* (1989, 1990a; 1990b;1992a;1992b) reported that pigs also serves as intermediary host of *T. saginata*-like of Taiwan, Korea, Indonesia, Madagascar and Ethiopia.

TABLE 3 – DIMENSIONS OF PROTOSCOLICES, *ROSTELLUM*, HOOKS, SUCKERS FROM 12 CYSTICERCI OF 44 RECOVERED FROM BOVINE EXPERIMENTAL INFECTED WITH EGGS OF *Taenia solium*. THE RESULTS EXPRESS THE MEAN OF ALL CYSTICECI MEASURED FOR EACH ANIMAL.

Animal	Cysticerci living	Protoscolices		F	Rostellum		
	n °	Width (x) μm	Length (x) μm	Diameter (x) μm	Hooks n⁰	Length (x) μm	Diameter (x) μm
01	04	839,5	854,8	167,6	15	35,6	315,2
02	07	1.095	1.129,5	303,4	79	25,4	340,6
03	01	504,3	381,3	0	0	0	331

In the State of Paraná the neurocysticercosis human cases has been high (GUSSO,1997), and the number of pigs abated in butcher shops with cysticercosis has relatively been low (0,03) (MINOZZO, 1997). Other sources of infection of T. solium were looked for that didn't go the habitual host. Among the victuals consumed by the people, cattle meat is the part of the meal consumed daily by most of them. Thus, cattle could also be intermediary host of T. solium. In this experiment, cattle were infected with 40,000 eggs of T. solium, developed cysticerci in larger number in the organs (56,8%) and in skeletal muscles (43,2%). In cattle infected with eggs of T. saginata (Minozzo, 1997), high cysticerci distribution was observed in the skeletal musculature (589 cysticerci) and smaller amount in the organs (102 cysticerci). In the present study, larger number of calcified cysticerci were observed in bovine infected by T. solium. Also,

contrasting data was observed in relation to the development of T. solium in the habitual host, mainly in what refers to the biological aspects (dimensions, morphology and development stage). The perception that cysticerci of T. solium is developed in the organs of bovine, and come viable it is an argument that could justify the morphologic alterations of the same ones. Another aspect to be taken in consideration is the percentage of degenerate cysticerci (59,1%) in relation to the living ones (40,9%). If compared with data of the experimental infection of bovine, with eggs of T. saginata (MINOZZO et al., 1997), these were significantly different, because 17,8% of cysticerci were degenerate against 82,2% alive. Relationship among the number of inoculated eggs and cysticerci recovered also differed. When compared the bovine ones infected with eggs of T. saginata and T. solium, the recovery were of 2-585/20.000 and 223/40.000 of the inoculated eggs, respectively. The present work showed that eggs of T. solium were developed for mature cyst in bovine. However, the cysticerci presented morphologic alterations as variable number of hooks, from 1 to 42 for cysticerci, and short hooks with medium dimension of 27µm (5,4-49,6µm). That differed from the pattern found by other authors in pigs and bovine. In pigs, different authors (SOULSBY, 1968; BORCHERT, 1981; LAPAGE, 1981; FAN, 1989, 1990a;1990b, 1992a;1992b) found hooks in variable numbers of 22-32 and mean dimensions 94-187µm (50-235µm). However, in pigs the experimental infection with T. saginata like, of several Asian origins, resulted in cysticerci with hooks number and inferior sizes to the found in our experiment, 9 to 18 (1-52) and of 10-12µm (2-28µm). In relation to the number of hooks. Pessoa (1982) described that in the process of degeneration the hooks could be found in the vesicular liquid and also cyst without rostellum and without hooks crown. CANEDO et al. (1982) observed that the cysticerci of T. solium, soon after the evagination don't show the hooks crown, which appeared, later on for 1-2 seconds being followed by retraction. The existence of stumps or sub species have been suggested to explain the differences of the size of the hooks of the cysticerci of the T. solium found in the pigs, cats, dogs and the man (CAÑEDO et al., 1982). In the literature, we found cases of cysticerci without rostellum and without hooks crown demonstrating be Cysticercus cellulosae in degeneration. More work is required to be carried out to study if the cysticerci found in the bovine would develop for adult form in definitive host.

## Conclusion

The infection of cattle with the eggs of *T*. *solium* is possible. A total of 44 cysticerci were recovered from three bovine experimentally infected. 18 cysticerci were (40.9%) alive and 26 (59.1%) calcified. 19 (43.2%) were found in the skeletal musculature and 25 (56.8%) in the organs (80% in the liver, 8% in the heart, 8% in the tongue and 4% in the kidneys). However, more studies are required to be carried out if the cysticerci found in the bovine would develop for adult form in definitive host.

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