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# Gastrointestinal motor disturbance in rabbits experimentally infected with *Strongyloides papillosus*

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**Abstract**

*Strongyloides papillosus* is a common nematode in ruminants, and the rabbit is the only susceptible experimental animal that has been identified to date. It is known that heavy infection with *S. papillosus* causes death in a number of animals. However, even though a number of fatal cases have been reported, the mechanism by which *S. papillosus* infection leads to death remains unknown. In this study, the pathogenic effect of *S. papillosus* infection on gastrointestinal motility in infected rabbits was investigated by radiographic means. Gastrointestinal motility in rabbits experimentally infected with *S. papillosus* was assessed by contrast radiography after oral administration of barium sulfate on days 11 (group A) and 13 (group B) of infection. Body weight, food intake, fecal weight and egg count per gram of feces (EPG) were examined in order to investigate the effect of infection on gastrointestinal motility. Seven rabbits from each *S. papillosus*-infected and uninfected group were examined. Significant declines in body weight, daily food intake, and fecal weight, as well as gastrointestinal motor disturbances, were observed in association with elevated EPG counts in infected rabbits. This was only observed during the intestinal phase of *S. papillosus* infection. These results suggest that gastrointestinal motor disturbances underlie the anorexia, weight loss and subsequent death observed in rabbits infected with adult stage *S. papillosus*.


*Keywords:* Anorexia; Weight loss; Gastrointestinal motor disturbance; Rabbit; *Strongyloides papillosus*

## 1. Introduction

*Strongyloides papillosus*, a universally common intestinal nematode infection of ruminants, results in serious economic loss for livestock producers. In addition to any subclinical damage that occurs in infected animals, as with other gastrointestinal nematodes, warm and humid circumstance increases an incidence of disease and subsequent mortality in juvenile ruminants (Taira and Ura, 1991). A number of pathological studies have been done to reveal mechanisms to explain the damage resulting from *S. papillosus* infection. These observed damages are diverse due to variations in the number of infected larvae to which the animals are exposed, as well as environmental factors related to the care of the domestic animals. Heavy infection with *S. papillosus* can be fatal in lambs (Woodhouse, 1948; Nakamura et al., 1994a) and calves (Vegors, 1954; Turner, 1959; Taira and Ura, 1991). *S. papillosus* infection has been shown to cause sudden death in calves following experimental hyper-infection with *S. papillosus* larvae, characterized by sudden cardiac arrest without clinical signs (Taira and

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Ura, 1991; Taira et al., 1992). The incidence of sudden death in the field might be influenced by substandard management systems due to economic constraints, resulting in a serious loss of productivity (Kváč and Vítovec, 2007).

*S. papillosus* infection can be established experimentally in rabbits. A considerable number of studies have examined rabbits infected with this parasite in order to clarify the host–parasite relationship and pathogenicity of the infection. Heavily infected rabbits often die due to significant wasting during the intestinal phase of *S. papillosus* infection. This is characterized by anemia, severe anorexia and weight loss (Stankiewicz, 1964). Normal cardiac rhythms have been demonstrated in these rabbits until the time of death, even once they have become emaciated (Nakamura et al., 1994b). The cause of death in these rabbits has yet to be identified and structural lesions have not been identified at autopsy. Although reduced fecal output has also been observed in rabbits heavily infected with *S. papillosus*, no studies have identified specific gastrointestinal motor disturbances to date. Therefore, the purpose of this study was to identify gastrointestinal motor disturbances following infection with *S. papillosus* in rabbits, and to further investigate the pathogenicity of this parasite.

## 2. Materials and methods

### 2.1. Animals

Fourteen male Japanese-White rabbits were purchased from Biotec. Ltd., Saga, Japan, for use in this study. They were parasite-free and six weeks of age, weighing 1.0–1.3 kg, at the start of experimentation. They were kept separately in stainless steel cages containing feces trays covered with plastic mesh in order to collect fecal samples. The body weight of each animal was measured and they were given 200 g of standard pellet food everyday at 6:00 p.m. Water was provided *ad libitum*. Their daily food intake was determined by subtracting the weight of remaining food from the amount provided (200 g) at the time of next feeding. Wet fecal weight was measured everyday for each rabbit at the same time. Ten grams of feces, or the entire sample if the total weight was less than 10 g, were dried by a freeze dryer. After drying, dried fecal weights were measured and divided by original weight (g) in order to calculate the dry weight per gram of wet feces, then multiplied by total grams of wet fecal weight. All protocols were approved by the Institutional Review Board for animal experiments of the University of Miyazaki.

### 2.2. Parasitological techniques

A strain of *S. papillosus* was originally isolated from a naturally infected calf in Miyazaki, Japan, and then maintained in our laboratory by serial passage in rabbits. Third-stage infective larvae (L3) were obtained from fecal culture using a filter paper culture method. In brief, feces were smeared in the middle of filter paper (12 cm × 60 cm) and, after banded by elastic band, the edge of rolled filter paper were soaked in tapped water in a plastic container for incubation at 25 °C until time of use. L3 that emerged into the water were collected following centrifugation, counted under a stereomicroscope, then used for infection. The rabbits were infected with L3 of *S. papillosus* by subcutaneous inoculation into the hypogastric region. Freshly prepared L3 was used for infection within 7 days of culture. Examination of fresh wet feces was carried out for each rabbit daily from the time of infection until the administration of barium sulfate. The egg count (EPG) was determined using a modified McMaster technique (Taira et al., 1991).

### 2.3. Experimental design and radiographical analysis

This experiment was performed to investigate the effect of *S. papillosus* infection on gastrointestinal motility. To begin, a total of fourteen male rabbits were randomly allocated into two groups of seven rabbits each. A group of seven rabbits were infected subcutaneously with 100,000 of L3 *S. papillosus* on day 0 for heavy infection. The same number of control rabbits received only subcutaneous saline. Following this, three out of seven rabbits in both the infected and control groups were merged into group A ($n = 6$), which received barium sulfate on day 11 of infection. The remaining four rabbits in each group were then allocated into group B ($n = 8$), which received barium on day 13 of infection. Barium-based contrast radiography was employed to evaluate the gastrointestinal transit time in each rabbit. A 3-ml aliquot of barium (100%-w/v) was administered by catheter directly into the stomach on day 11 of infection in group A and on day 13 of infection in group B. Radiographs were taken 1, 3, 6, 12 and 24 h after barium instillation in group A, and 1, 3, 6, 12, 24, 48 and 72 h after barium instillation in group B. Gastrointestinal motility was assessed based on the amount of barium sulfate remaining in the stomach as follows: 0: empty; 1: almost empty; 2: some remaining; 3: a large amount remaining.