

## Eosinophilic myositis resulted from *Sarcocystis* infection in prime marbled beef of Japanese black cattle

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### Abstract

Partial changes of color (greenish to brownish) were found in prime marbled beef of Japanese black cattle. The disseminated lesions of the skeletal muscles were histopathologically examined in relation to *Sarcocystis* infection. The lesions in the muscles showed granulomas with inflammatory cell infiltration. The sarcocysts had a distinct wall, which was radically striated by palisading villar protrusions. The sarcocyst wall was surrounded by degenerative eosinophils and necrotic muscle fibers. In conclusion, eosinophilic myositis in prime marbled beef of Japanese black cattle resulted from *Sarcocystis* spp. infection. The muscular lesions were characterized by the presence of granulomas and capsulated sarcocysts surrounded by numerous eosinophils.

Key words: eosinophilic myositis, granulomas, Japanese black cattle, sarcocyst, *Sarcocystis* spp.

### Introduction

Previous studies suggested that *Sarcocystis* spp., often found in association with granulomatous myositis, should be responsible for eosinophilic myositis (Jensen *et al.*, 1986; Gajadhar *et al.*, 1986; Gajadhar and Marquardt, 1991; Wouda *et al.*, 2006). Eosinophilic myositis is a relatively rare disease in cattle observed in meat inspection after slaughter. Most investigators describe a high prevalence of *Sarcocystis* infection in heart, esophagus, diaphragm and tongue of slaughtered cattle. Although the skeletal muscles are the most often eaten parts of beef, the prevalence of *Sarcocystis* is relatively low in these muscles. In addition, skeletal muscles are not severely affected by *Sarcocystis*.

The prevalence of *Sarcocystis* spp. cysts was lower in Japanese beef (6.31%) than in beef imported from America (36.78%) or Australia (29.49%) (Ono and Ohisumi, 1999). Marbled beef of Japanese black cattle is highly valued as brand-name beef produced in the specified farms.

I found a partial change of color in prime beef of Japanese black cattle. In this study, the disseminated lesions of the skeletal muscles were histopathologically examined in relation to *Sarcocystis* infection.

### Materials and Methods

Samples of muscles from the hindquarters in

Japanese black cattle were excised in the slaughter house in Matsuzaka-city and submitted to our institutes. The specimens containing visible lesions were fixed in 10% neutral buffered formalin, processed by routine methods and embedded in paraffin wax. 4- $\mu$ m paraffin sections were stained with hematoxylin and eosine (HE) and by van Gieson's and Weigert's staining.

### Results

Macroscopic findings: The marbled beef had well-demarcated, oval or elongated greenish or grayish-green lesions up to 2-10 mm in length and 2-8 mm in diameter. The small disseminated lesions were a typical form of eosinophilic myositis which resulted in carcass condemnation (Figure I). The large-sized lesions were discrete focal stripes or patches. In some portions, there was a rigid bulging of brownish lesions (Figure II).

Histopathological findings: The lesions in the marbled muscles showed granulomas with inflammatory cell infiltration. The granulomatous lesions had a central core consisting of degenerative eosinophilic leucocytes and remnants of necrotic skeletal muscle fibers. Among the marbled muscle fibers, there was accumulation of mixed inflammatory cells, mainly eosinophils with some neutrophils, macrophages and plasma cells, as well as erythrocytes

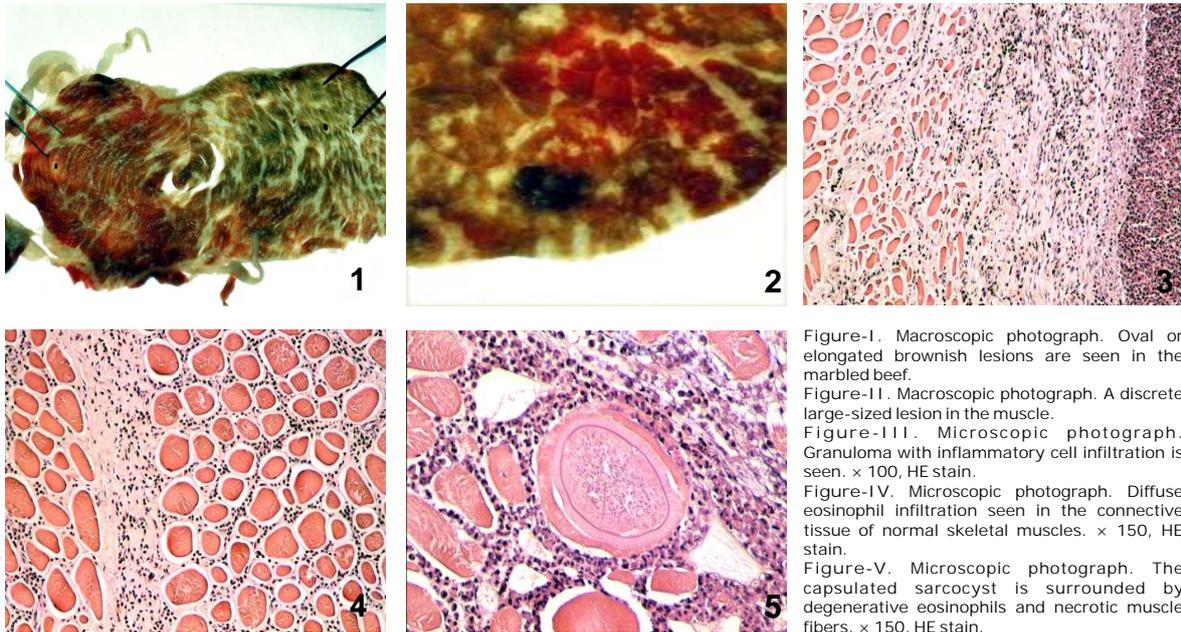


Figure-I. Macroscopic photograph. Oval or elongated brownish lesions are seen in the marbled beef.  
 Figure-II. Macroscopic photograph. A discrete large-sized lesion in the muscle.  
 Figure-III. Microscopic photograph. Granuloma with inflammatory cell infiltration is seen. × 100, HE stain.  
 Figure-IV. Microscopic photograph. Diffuse eosinophil infiltration seen in the connective tissue of normal skeletal muscles. × 150, HE stain.  
 Figure-V. Microscopic photograph. The capsulated sarcocyst is surrounded by degenerative eosinophils and necrotic muscle fibers. × 150, HE stain.

(Figure III). Pale to neutral staining eosinophils were observed around the sarcocysts with thick cyst wall. There was also diffuse eosinophil infiltration in the connective tissue of normal skeletal muscles (Figure IV).

The sarcocysts had a distinct wall, which was radically striated by palisading villar protrusions. The sarcocyst wall was surrounded by degenerative eosinophils and necrotic muscle fibers (Figure IV). Adjacent to the trapped sarcocyst, this cyst were enclosed with a thick outer rim of degenerative hyalinized remnants of muscle fibers.

Histopathological examinations revealed that eosinophilic miositis were closely associated with *Sarcocystis* infection.

Discussion

Muscle sarcocystosis is a parasitic infection acquired by ingestion of sarcocysts of *Sarcocystis* spp. in feces from an infected carnivore. *Sarcocystis* spp. have 2-host life cycle between carnivores and herbivores. In this life cycle, cattle are known to serve as the intermediate host. In the previous study, the cows experimentally challenged with sporocysts showed various clinical signs with myositis (Savini *et al.*, 1996). Histopathologically, skeletal myositis was associated with a mononuclear cell infiltration (primarily lymphocytes). Degenerating sarcocysts in muscles elicited a mononuclear cell (mainly lymphocytic) response as early as 1 to 3 months after infection.

In the present study, histopathological examinations indicated that granuloma-associated sarcocysts were partially destroyed or enclosed by dense, organized accumulations of inflammatory cells. These results showed that the thick cyst walls in the lesions were identical to those of *Sarcocystis hirsute*. The thick walled sarcocyst cannot be distinguished whether it is *Sarcocystis hirsute* or *Sarcocystis hominis*. However, *Sarcocystis hominis* has not yet been reported in Japan.

The pathogenesis of eosinophilic myositis is not clear and this muscular lesion is not always found in cattle suffering from *Sarcocystis* spp. The incidence of eosinophilic myositis is usually low in cattle (Wouda *et al.*, 2006). This breed of cattle was mainly fed with concentrated fed. It is uncertain whether the cattle made some contact with carnivores as the definitive host. The histopathological examinations showed 2 kinds of morphological changes as follows: granulomas with peripheral cell infiltration and capsulated sarcocysts surrounded by degenerative eosinophils. The initial cellular response seemed to be induced by substances (eosinophil chemoattractant) released from the sarcocysts during their normal metabolic process. These findings provided a presumption that cattle with eosinophilic myositis were predisposed to produce IgE in response to *Sarcocystis* bradyzoite antigen, and that eosinophilic myositis represented an abnormal response to sarcocyst degeneration, including a host-dependent,

Sarcocystic, type-I hypersensitivity (Granstrom *et al.*, 1989 and Granstrom *et al.*, 1990). Mononuclear cell infiltration is predominant feature of myositis associated with early Sarcocystis infection in calves without lesions of eosinophilic myositis. It is probable that eosinophilic myositis develops around degenerating sarcocysts in the later stage.

In conclusion, eosinophilic myositis in prime marbled beef of Japanese black cattle resulted from Sarcocystis spp. infection. The muscular lesions contained granulomas and capsulated sarcocysts surrounded by numerous eosinophils.

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