

Pathological and Parasitological Investigations in an Adult Bottlenose Dolphin (*Tursiops truncatus*)

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Abstract

This case aimed to perform pathological and parasitological examinations on an adult male bottlenose dolphin (*Tursiops truncatus*), which was found dead on the coast of Aegean Sea (Kusadası/Davutlar/Aydın-Turkey). At necropsy, intestinal volvulus and fibrinopurulent peritonitis were observed. In the histopathologic examination, severe vascular changes including oedema and hemorrhages were observed in the intestines formed by volvulus and peritoneum. In the parasitological examination revealed some ectoparasites (Copepoda: Pennellidae) on the skin. Also, *Stenurus minor* (*S. minor*) was detected in the lungs. In conclusion, severe ectoparasite infestation, intestinal volvulus, *S. minor* in the lungs, and severe renal calcinosis were defined in the adult bottlenose dolphin.

Keywords: Dolphin, intestinal volvulus, Pennellidae, *S. minor*

Şişe Burunlu Bir Yunusta (*Tursiops truncatus*) Patolojik ve Parazitolojik İncelemeler

Özet

Bu raporda, Ege Denizi sahilinde (Kusadası/Davutlar/Aydın-Turkey) ölü olarak bulunan yetişkin şişe burunlu erkek bir yunusta (*Tursiops truncatus*) patolojik ve parazitolojik bulgular değerlendirildi. Nekropside, intestinal volvulus ve fibrinopurulent peritonitis gözlemlendi. Histopatolojik incelemede, volvulus oluşan bağırsak bölümleri ile peritonda ödem ve hemorajileri içeren şiddetli vasküler değişiklikler görüldü. Parazitolojik incelemede, deri üzerinde bazı ektoparazitler (Copepoda: Pennellidae) görüldü. Ayrıca, akciğerlerde *Stenurus minor* (*S. minor*) tespit edildi. Sonuç olarak, yetişkin şişe burunlu bir yunusta şiddetli ektoparazit enfestasyonu, intestinal volvulus, akciğerlerde *S. minor* ve şiddetli böbrek kalsinozu tanımlandı.

Anahtar sözcükler: Yunus, intestinal volvulus, Pennellidae, *S. minor*

INTRODUCTION

Parasitic infections represent a potential threat to endangered populations of marine mammals. However, little information is known about the role parasites in the deaths and strandings of dolphins^[1]. Lungworms (Metastrongyloidea: Pseudalidae) have been implicated as the main factor of natural mortality of dolphins^[2].

The parasites belonging to the Pennellidae family are the common ectoparasites found in large pelagic fishes. A

series of planktonic free swimming larval phases exist in their life cycles after metamorphosis, males are free-living; however, females attach to the body surface of host organisms^[3]. The copepods display a permanent parasitism, and they feed on mucus, epithelial scrapings, and tissue fluids. No literature shows their direct parasitic effect; however, the wounds caused by parasite may serve as points of entrance for pathogenic microorganisms^[4].

In this case, severe ectoparasite infestation, intestinal volvulus, *S. minor* in the lungs, and renal calcinosis were defined in an adult bottlenose dolphin.



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CASE HISTORY

Necropsy was performed on an adult male bottlenose dolphin (*T. truncatus*) found dead on the coast of Aegean Sea (Kusadasi/Davutlar/Aydin-Turkey, 37° 46'N 27° 15'W). Following necropsy, tissue samples (skin, lung, liver, intestines, kidneys, heart and testicles) were fixed in 10% buffer formalin solution, processed routinely, 5 µm sectioned and stained with hematoxylin and eosin (H.E). Furthermore, the sections were stained by melanin removal method II for melanin and periodic acid-Schiff reaction (PAS) for lipofuscin [5]. Skin samples with parasites and tissue samples from the lungs were preserved in 70% alcohol for further parasitological examination.

In the external view, 125 lesions 3.0–5.0 mm in diameter (52 of them had parasites) were counted, especially on the cauda-lateral and abdominal parts of the skin (Fig. 1A). The

parasites were gray-black in color, filament-like structures and 4.5-6.0 cm in length. Some teeth of the dolphin were extracted or rasped.

At necropsy, serosanguinous fluid with fibrin flecks was found in the abdominal cavity. There was an 180° volvulus of 35 cm segment of the jejunum. The involved intestinal segment was dark red in color with fibrinonecrotic exudate on the serosal surfaces (Fig. 1B). Some postmortem changes (autolysis, putrefaction, and pseudo-melanosis) were also observed in all organs.

The histopathological examinations revealed parasitic dermatitis through the parasitic tunnels, surrounded by inflammatory reaction characterized by macrophages, neutrophil leucocytes with areas of hemorrhages (Fig. 2A). These lesions progressed through the muscle tissue, leading to muscle necrosis. There was a thick fibrinopurulent exudate with severe neutrophil leucocytosis, oedema and

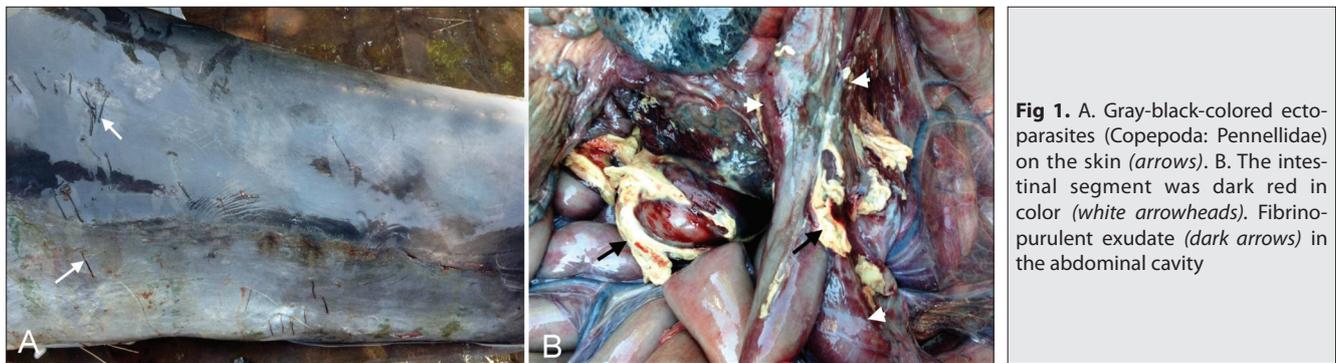


Fig 1. A. Gray-black-colored ectoparasites (Copepoda: Pennellidae) on the skin (arrows). B. The intestinal segment was dark red in color (white arrowheads). Fibrinopurulent exudate (dark arrows) in the abdominal cavity

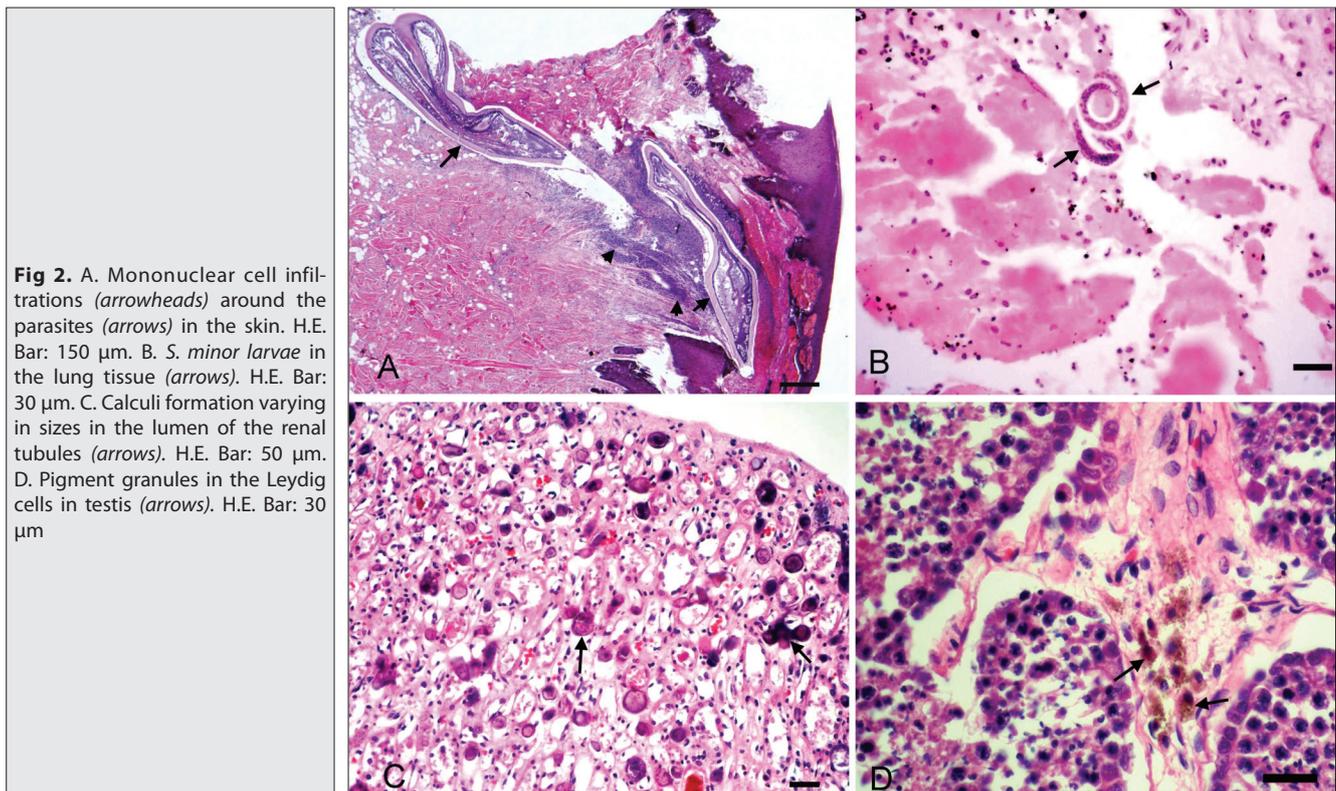


Fig 2. A. Mononuclear cell infiltrations (arrowheads) around the parasites (arrows) in the skin. H.E. Bar: 150 µm. B. *S. minor* larvae in the lung tissue (arrows). H.E. Bar: 30 µm. C. Calculi formation varying in sizes in the lumen of the renal tubules (arrows). H.E. Bar: 50 µm. D. Pigment granules in the Leydig cells in testis (arrows). H.E. Bar: 20 µm

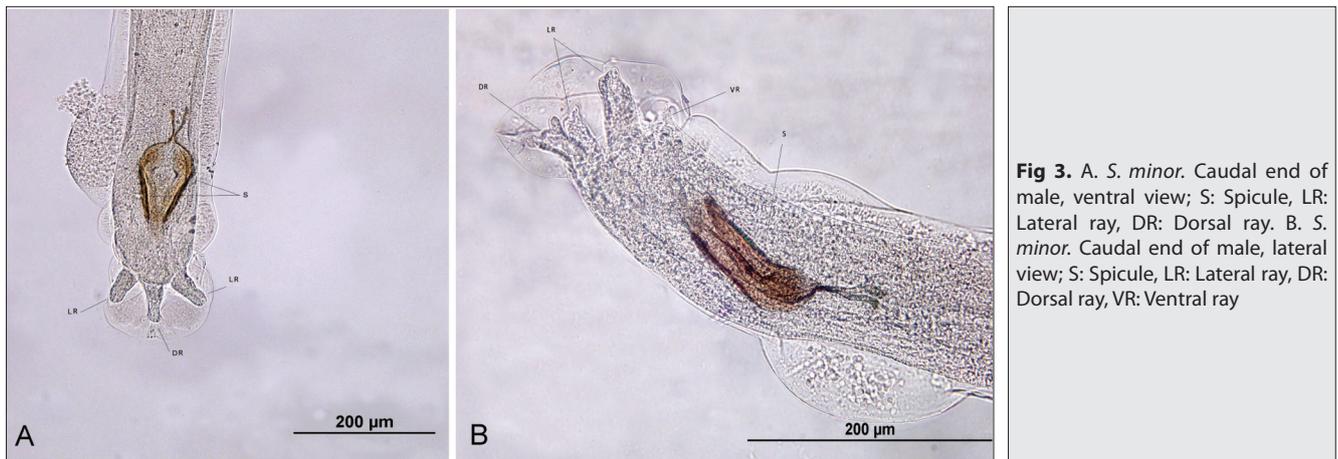


Fig 3. A. *S. minor*. Caudal end of male, ventral view; S: Spicule, LR: Lateral ray, DR: Dorsal ray. B. *S. minor*. Caudal end of male, lateral view; S: Spicule, LR: Lateral ray, DR: Dorsal ray, VR: Ventral ray

hemorrhages in the intestinal serosa. In the lungs, edema, macrophages, and few parasites larvae were seen in the alveolar lumens (Fig. 2B). Diffusely, severe pneumoconiosis (anthracosis) was seen throughout bronchi, bronchioles and alveolar walls. In kidneys, interstitial and glomerular fibrosis were observed. In the medulla, calculi formation (Fig. 2C) in the lumen of the renal tubules were detected. There were diffuse dark brown pigment granules within Leydig cells in testes (Fig. 2D), and these granules were positively stained with melanin removal method II for melanin, but gave a negative reaction for PAS.

The tissues that underwent pepsin-aided digestion were obtained without damaging the outer layer and parasites. However, since the tissues were not extracted from deeper layers (localized just above the head and anterior thorax of the parasite), the parasite could be characterized only at the family level and morphological characteristics and histological appearance of the parasite show similarities with those of Pennellidae family [6,7]. The parasitological examinations of the lungs showed the presence of one male nematode, and the morphological characteristics were found compatible with those of *S. minor* in the light of previous studies [2,8] (Fig. 3A, B).

DISCUSSION

In the previous studies, five species belonging to the Pennellidae family were reported in the seas of Turkey [3,9-12]. *Lernaeolophus sultanus* (in the mouth of Black Sea bream, *Diplodus vulgaris*) [9], *Pennella instructa* (in the anal/pectoral fins and abdominal muscle of swordfish) [10], *P. balaenopterae* (in the skin of fin whale, *Balaenoptera physalus*) [11], *Peniculus fistula* (in the ventral fin of a dolphin species, which is called lambuka, *Coryphaena hippurus*) [12] and *P. filose* (in the fins, body surface and gills of the fishes known as yellowtail, *Seriola dumerili*) [3] were found in the Turkey. *P. balaenopterae* was also found in a harbor dolphin (*Phocoena phocoena*) on the southern Aegean Sea coast of Turkey [6]. Although the parasites in this case could not be characterized at the species level, morphological

characteristics and histological appearance of the parasite show similarities with those of Pennellidae family [6,7]. Besides detecting lesions and parasites, it is very important to highlight that the presence of this parasite has never been reported earlier to this case, and this is the first report of the presence of ectoparasites that belonging to the family Pennellidae from Bottlenose Dolphin (*T. truncatus*) in Turkey.

Intestinal volvulus is one of the most common changes in the intestines of marine mammals. It was reported that abnormal peristaltic movements, inflammation, parasitism, neoplasia, foreign bodies, vigorous exercise or violent rolling are the major risk factors for intestinal volvulus [13]. In the present case, no underlying etiology of the volvulus was determined on gross and histopathologic examination; however, it is suggested that verminous pneumonia and severe skin parasites may be predisposing factors that triggered intestinal volvulus [13,14].

Lungworms (Metastrongyloidea: Pseudaliidae) have been implicated as the main factor of natural mortality of marine mammals [2]. In one study, the prevalence of lungworm infections was determined to be 77% in the stranded bottlenose dolphins [15]. To date, *Stenurus ovatus*, *Halocercus lagenorhynchi*, *Pharurus alatus*, and *Skrijabinalius cryptocephalus* related verminous pneumonia [1,15,16] was reported in the bottlenose dolphins; but there was no case related to *S. minor*. It is reported that the prevalence of *S. minor* infection of harbor dolphins on the coast of Norway, Iceland, Greenland and South America was 66-100% [17,18]. In Turkish Seas, *S. minor* was detected in the lungs of the harbor dolphins on the coast of Black Sea [19], and in the stomach content of the striped dolphins in Eastern Mediterranean coast [20]. Cranial air sinuses, the inner ear, and supracranial airways are the most common locations of *S. minor*, and the parasite has been also found in the lungs, stomach and middle ear [17,21]. The pathological effects of *S. minor* in tissues have not been explained clearly yet. However, it has been reported that parasite-dependent severe inner ear infections may cause dysregulation in hearing and navigation abilities [21]. Although some parasitic larvae were also seen in alveoli in the case

of *S. minor*, not detecting any chronic lesion or adult parasite sections was interpreted as a sign of early infection in this case ^[14-16,19].

In conclusion, the presence of rasped and extracted teeth together with pneumoconiosis was suggested that this dolphin was likely involved in human interaction activities for a while and then left to the open sea.

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