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Dermocystidium sp. (Mesomycetozoea: Dermocystidiaceae) primary ocular infection in a koi carp (Cyprinus carpio var. koi)

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Keywords:	Mesomycetozoea, Dermocystidium sp., koi carp, eye



1	SHORT COMMUNICATION
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3	Dermocystidium sp. (Mesomycetozoea: Dermocystidiaceae) primary ocular infection in a koi
4	carp (<i>Cyprinus carpio</i> var. <i>koi</i>)
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14	Running head: Dermocystidium sp. ocular infection in a koi
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Journal of Fish Diseases

Dermocystidium spp. (Mesomycetozoea: Dermocystidiaceae) comprises several species able to infect a wide range of marine and freshwater fish (Gozlan et al., 2014; Rowley et al., 2013). The taxonomic position of *Dermocystidium* is still under debate, even though Dyková and Lom (1992) observed hyphae associated to *Dermocystidium koi* infection, suggesting its fungal nature. Currently, the genus is classified in the superclass Mesomycetozoea which includes protists "fungus-like" in the supergroup Opisthokonta (Gozlan et al., 2014).

The lesions caused by *Dermocystidium* spp. in fish are chronic and characterized by visible cysts or nodules on skin and fins (Feist et al., 2004; Wildgoose, 1995; Zhang & Wang, 2005), but severe gills and systemic infections causing mortality have been described in several species (Höglund et al., 1997; Olson & Holt, 1995; Athanassopoulou, 1998; Landsberg & Paperna, 1992; Kirkbright et al., 2016; Fujimoto et al., 2018).

The present study described a primary ocular infection by *Dermocystidium* sp. in a koi carp 37 (Cyprinus carpio var. koi). The koi carp was found dead by the owner of a private pond and sent 38 refrigerated to the laboratory. The fish presented an evident subconjunctival whitish exophytic 39 tissue, surrounded by reddish edges, that protruded over both ocular bulbs (Fig. 1a). The fish was 40 necropsied and tissues fixed in 10% buffered formalin. The ocular globes were then dissected under 41 stereomicroscope and smears of the exophytic tissue placed on slides, clarified with lactophenol or 42 43 stained with China ink, and examined under light microscope. For histopathology, tissues were routinely processed, sections were cut at 4 µm and stained with Haematoxylin and Eosin (H&E), 44 Periodic Acid Shiff (PAS) and Gomori. 45

At the stereomicroscope, the peribulbar tissue and anterior chamber presented numerous filiform 3mm-length white hyphal-like cysts (Fig. 1b). At the light microscope, the cysts were filled with round variably sized (5.53-12.90 μ m in diameter; mean=9.71; sd=1.99; n=80) mature spores referable to *Dermocystidium* sp. admixed with developmental stages (>5 μ m). The spores showed a large refractile body or central vacuole and a peripheral cytoplasm with a nucleus and three or more vacuoles (Fig. 1c).

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At histology, the peribulbar soft tissues until muscle and the choroid space of the posterior chamber, 52 53 adjacent to the retina, were infiltrated by numerous elongated cysts (Fig. 2f-h). The cysts had a thick eosinophilic wall and contained numerous spores (Fig. 2h,i). Around the cysts, a 54 granulomatous inflammatory reaction was visible (Fig. 2h). The iris and the ciliary body were also 55 replete by numerous free mature spores. An intense mixed inflammatory infiltrate with 56 multinucleated giant cells, vascular congestion and abundant granulation tissue were observed (Fig. 57 2g). Haemorrhagic and necrotic areas were observed. Some basophilic granular aggregates referable 58 to bacteria were also found. Neither developmental stages of *Dermocystidium* sp. nor inflammatory 59 reaction were seen in internal organs. PAS and Gomori stains helped to visualize the developmental 60 stages (Fig. 1d,e) and exclude the presence of spores in other organs. 61

In literature, only three descriptions of ocular infection by Dermocystidium sp. are reported in fish 62 (Elkan, 1962; Molnar et al., 2008; Fujimoto et al., 2018). The present case is the first report of a 63 64 primary ocular infection in a koi carp. The spore size and morphology are consistent with literature reports of *Dermocystidium koi* (Gjurcevic et al., 2008; Hoshina & Sahara, 1950). The life cycle of 65 Dermocystidium spp. is partly unknown. The parasite produces motile zoospores which encyst in 66 the host tissues, form sporonts by division of the plasmodia until subsequent maturation of spores 67 (Lotman et al., 2000; Mahboub & Shaheen, 2020). In experimental trials, it has been demonstrated 68 69 that fish can acquire the infection through intra-gastric and immersion routes. The pathogen cycle begins from the ingestion of spores through feeding on infected fish or contaminated feed or via 70 penetration of spores into the gills (Mahboub & Shaheen, 2020). 71

Considering the economic value of koi carps and the possible direct transmission of the infection in
 optimal temperatures condition, *Dermocystidium* spp. infection should be considered as an
 important disease also in ornamental species.

75

76 Conflicts of interests

77 The authors declare to have no conflict of interests.

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79	Data availability
80	The data that support the findings of this study are available from the corresponding author upon
81	request.
82	
83	References
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137 Figure legends

Figure 1. (a) Koi carp showing an ocular exophytic whitish tissue. (b) Numerous filiform hyphallike cysts visible at the stereomicroscope (bar=100 μ m). (c) The cysts are filled with round mature spores morphologically referable to *Dermocystidium* sp. (bar=10 μ m). The spores show a large refractile body (RB) and a peripheral cytoplasm with a nucleus and vacuoles (inset, arrow). (d) Gomori stain shows cytological details of the spores (bar=10 μ m). (e) PAS stain evidences the developmental stages of the parasite. Four zoospores are visible inside a spore (arrowhead) (bar=10 μ m).

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Figure 2. (f) The choroid space near the retina is infiltrated by numerous cysts (asterisk) (H&E, bar=500 μ m). (g) The iris and the ciliary body are replete by numerous free mature spores (arrowhead). A severe inflammatory reaction and granulation tissue are evident. Note the multinucleated giant cells (inset) (H&E, bar=50 μ m). (h) Around the cysts, a granulomatous inflammatory reaction characterized by a wall of epithelioid cells is visible (inset) (H&E, bar=100 μ m). (i) The cysts show a thick eosinophilic wall and contain spores at different developmental stages (H&E, bar=50 μ m).

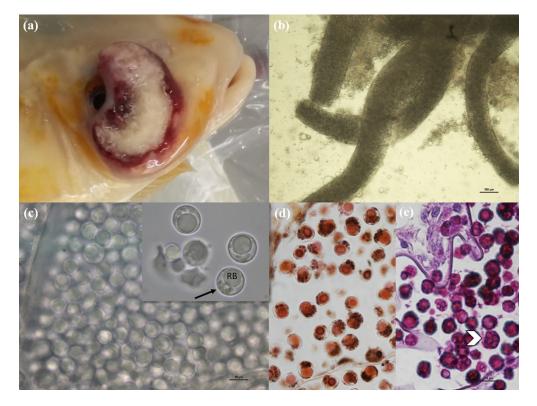


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144x108mm (300 x 300 DPI)

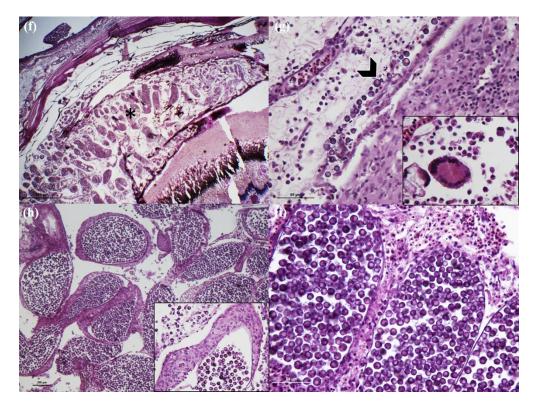


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