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Mucor amphibiorum Infection in Platypus (*Ornithorhynchus anatinus*) from Tasmania

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ABSTRACT: *Mucor amphibiorum*, a fungus previously isolated from frogs and toads, is reported from free-living platypus, *Ornithorhynchus anatinus*, from rivers in northern Tasmania. This fungus is responsible for the severe ulcerative skin condition originally described by Munday and Peel (1983). *Mucor amphibiorum* was isolated from dermal lesions on four separate occasions. The gross and histopathological appearance of the fungal lesions were similar to the earlier description. In vivo this fungus develops as spherical forms containing a number of daughter spherules; no mycelia are seen in tissue sections. By contrast, the in vitro growth consists of aerial aseptate mycelia and sporangia, features typical of the genus *Mucor*. This is the first report of this organism causing a fatal disease in a mammal. Susceptibility to infection may be due to the platypus having a body temperature of 32 C while the maximum temperature for growth of *M. amphibiorum* is 36 C.

Key words: *Mucor amphibiorum*, platypus, *Ornithorhynchus anatinus*, skin infection.

Munday and Peel (1983) reported on an ulcerative skin condition in platypus (*Ornithorhynchus anatinus*) from Tasmania. Numerous spherical thick-walled organisms measuring 13 to 23 μm ($\bar{x}$ = 18 μm) were seen in the pyogranulomatous dermal lesions. Attempts were made to isolate the organism using selective media for fungi and algae; however, the only organism consistently recovered was a fungus of the genus *Mucor*. The authors observed that no mycelia or hyphal development occurred with the spherical organisms in vivo, in contrast to the growth of *Mucor* in vitro. They concluded that the organism observed by light and electron microscopy was unlikely to be the same as the cultured *Mucor* sp., but they remained puzzled by the inability to culture any other organism, including algae, which resembled those seen in the skin lesions.

Nine additional platypus with similar ulcerative dermatitis were examined. All animals were found moribund or dead at the water's edge or on the banks of natural waterways in northern Tasmania: five came from the South Esk river (Perth, 147°35′E, 41°35′S), three came from the Meander river (Deloraine, 146°39′E, 41°32′S; Westbury, 146°50′E, 41°31′S), and one came from Brumbys creek (Cressy, 147°47′E, 41°45′S). Routine post mortem examination of these platypuses revealed gross and histopathological lesions similar to those described by Munday and Peel (1983). Ulcerated lesions were found on the skin of ventral tail, the haired areas of the upper limbs, neck, head and dorsal body, as well as the unhaired areas of the distal limbs. One platypus had several open skin wounds along the dorsum adjacent to large numbers of engorging ticks, *Ixodes ornithorhynchi* (predominantly nymphs and adult females). Another platypus, which was found moribund, had several ulcers on the head and neck (3 to 5 cm in diameter) in which calliphorid blowflies had oviposited. Numerous maggots were feeding under the skin at the periphery of the ulcers. The platypus from Brumbys creek was observed and filmed by a trout fisherman.

In four cases for which specimens were available, organisms identical to those described by Munday and Peel (1983) were observed in the pyogranulomatous inflammatory tissues underlying the ulcerated skin lesions. The outer wall and internal septa of the organisms stained specifically with periodic acid-Schiff (PAS) stain (Culling, 1963) and Gomori methanamine silver nitrate (Armed Forces Institute of Pathology, 1960); no vegetative mycelia or

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hyphae were seen. Organisms, also were found in suppurative dermal lesions of the feet and upper legs in the absence of any obvious skin wounds. Several small pulmonary granulomas (≤1 mm in diameter) distributed thoughout the lungs also contained the organisms.

Fresh samples of skin lesions from three cases were cultured on Sabouraud's medium (Oxoid Unipath Ltd., Basingstoke, Hants, England). A *Mucor* sp. was recovered from skin lesions in all three cases. The growth of the fungus on Sabouraud's medium was similar to that originally isolated by Munday and Peel (1983). Isolates sent to the National Reference Laboratory for Medical Mycology, Royal North Shore Hospital, St. Leonards, New South Wales were identified as *M. amphibiorum* (Schipper, 1978).

Mucor amphibiorum was first isolated in 1972 from an Australian green tree frog (*Hyla* syn. *Litoria caerulea*) held in a collection in West Germany (Frank et al., 1974). Subsequently, other amphibians kept in the same or neighbouring exhibits also became infected with *M. amphibiorum*. Frank et al. (1974) suggested that the tree frog may have been infected when imported from Australia. At the time of its original isolation, Frank et al. (1974) demonstrated that *M. amphibiorum* formed spherules (tissue forms of sporangia/sporangiophores) containing daughter spherules (sporangiospores/chlamydospores) within tissue lesions. In cultures, these authors observed aerial mycelia and sporangiophores more typical of other members of the genus *Mucor*.

Frank et al. (1974) experimentally infected European frogs (*Rana temporaria, R. esculenta*) and a toad (*Bufo bufo spinosa*) by skin abrasion and intra-peritoneal inoculation. The infection was fatal causing numerous white lesions containing the organism in many internal organs, most commonly in the spleen and the liver (Frank, 1976). Experimentally infected reptiles remained healthy and at necropsy, only small lesions containing a few organisms were observed. The organism failed to establish or cause lesions in laboratory mice, rats and guinea pigs (Frank et al., 1974).

The infectivity of *M. amphibiorum* to amphibia may be due to its temperature tolerance. The maximium temperature at which *M. amphibiorum* grows in vitro is 36 C (Scholer et al., 1983). In the platypus, a mammal with a body temperature of 32 C (Grant and Dawson, 1978), lesions occur predominantly in dermal tissues of the body and extremities. This distribution may indicate an even lower temperature tolerance in vivo. Severe disseminated granulomas such as those seen in the poikilothermic amphibians have not been seen in platypus.

Our findings establish that *M. amphibiorum* can infect the aquatic monotreme, *O. anatinus*. Apart from the original studies of Frank et al. (1974), the organism also has been recovered from cane toads (*Bufo marinus*) in northeastern Australia (R. Speare, pers. comm.).

The route of infection for this organism in the platypus is not known. Frank (1976) suggested that anurans may be infected by ingestion or via skin abrasions. Skin wounds, abrasions or injuries may be a route of entry for platypus and might explain the ulcerative dermatitis of platypus. Such skin injuries could result from intraspecific fighting with spur penetration, bites from other aquatic animals (water rats, eels, etc.) or ectoparasites like ticks and mites.

Insufficient cases have been examined to assess the significance of this disease to wild platypus populations in Tasmania. To date the infection has not been reported from platypuses on the Australian mainland. At least two affected animals also were severely emaciated, based on the absence of body fat, especially the subcutaneous fat layer in the tail (Grant and Temple-Smith, 1983). It is not known whether the infection is the cause or the effect of the observed debilitation/emaciation. Further work is required to monitor environmental sources of *M. amphibiorum*.

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