Gulf and Caribbean Research Vol. 15, 1–4, 2003 Manuscript received July 22, 2002; accepted November 8, 2002

A NEW GEOGRAPHIC LOCALITY AND THREE NEW HOST RECORDS FOR *NEOBENEDENIA MELLENI* (MACCALLUM) (MONOGENEA: CAPSALIDAE)

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ABSTRACT A new geographic locality record and three new host records for *Neobenedenia melleni* (MacCallum, 1927) (Monogenea: Capsalidae) are provided. Specimens of *N. melleni* were collected from the skin of three Florida pompano, *Trachinotus carolinus* (Linnaeus, 1766) (Carangidae), caught in the northern Gulf of Mexico off Horn Island, Mississippi; from the skin of a bluering angelfish, *Pomacanthus annularis* (Bloch, 1787) (Pomacanthidae), in the Shark Reef Aquarium at Mandalay Bay, Las Vegas, Nevada; from the skin of a rock greenling, *Hexagrammos lagocephalus* (Pallas, 1810) (Hexagrammidae), in the Alaska SeaLife Center, Seward, Alaska; and from the skin of two blue-barred ribbon gobies, *Oxymetopon cyanoctenosum* Klausewitz and Condé, 1981 (Microdesmidae), in a tropical fish clearinghouse in Hayward, California. This is the first published record of the parasite from a microdesmid or wild carangid. Prior to this report, no specimen of *N. melleni* had been reported from a wild-caught fish in the Gulf of Mexico. The presence of *N. melleni* in the Gulf of Mexico is particularly noteworthy because this monogenean is a known pathogen of cultured fishes in netpens and recirculating seawater systems.

Introduction

Neobenedenia melleni (MacCallum, 1927) (Monogenea: Capsalidae) is a serious pathogen of confined teleosts because it exhibits a direct life cycle and a low degree of host specificity as well as an apparent low degree of site specificity. It infects the eyes, fins, gill cavity, nasal cavity, and skin of primarily shallow-water or reef-dwelling teleosts ranging from tropical to temperate marine and brackish waters (Bullard et al. 2000); however, the site preference and microhabitat of *N. melleni* have not been studied in great detail (Whittington and Horton 1996). This parasite is unique among other monogeneans in that it has a broad host range and wide geographic distribution, infecting 26 fishes in 18 genera, 14 families, and three orders in the Caribbean Sea and Eastern Pacific Ocean as well as over 80 teleost species in public aquaria and aquaculture systems (for specific hosts see Whittington and Horton 1996, Bullard et al. 2000). Published wild, free-ranging, host records for *N. melleni* are relatively scarce, probably because specimens are overlooked during routine examinations of fishes: specimens of *N. melleni* are thin, flat, and opaque-white or nearly transparent, making them difficult to see and collect if the host's external surface is not examined critically and with adequate lighting. Furthermore, infected wild hosts are not obvious because they typically harbor few worms and lack a gross lesion. There is no report of a disease and no detailed information regarding a lesion associated with an infection of *N. melleni* in a wild host. Conversely, fishes in netpens, aquaria, and other recirculating seawater systems routinely develop intense infections of *N. melleni* marked by bloody lesions, body discoloration, emaciation, erratic behavior (i.e., rubbing against objects and flashing), and mortality (Jahn and Kuhn 1932, Nigrelli and Breder 1934, Robinson et al. 1992, Ogawa et al. 1995). Mueller et al. (1994) stated that infections of *N. melleni* may constrain the tropical finfish mariculture industry until cost effective and environmentally sound treatments are discovered and implemented.

Materials and Methods

Florida pompano were caught by hook and line, pithed, and placed in a cooler with a small portion of ice prior to necropsy. Infected aquarium-held fishes were pithed and immediately necropsied or fixed whole in 10% neutral buffered formalin (n.b.f.) and necropsied at a later date. All worms were fixed directly in 10% n.b.f. or 70% ethanol (EtOH) without coverslip pressure or heat. Whole mounts of worms were stained in Van Cleave's hematoxylin with several additional drops of Ehrlich's hematoxylin, made basic at 70% EtOH with

lithium carbonate and butyl-amine, dehydrated, cleared in clove oil, and mounted in Canada balsam. Worms were identified using the original description of *N. melleni* (as *Epibdella melleni* MacCallum, 1927) and the redescription of *N. melleni* and key to species of *Neobenedenia* by Whittington and Horton (1996). The primary features that identified our specimens as belonging within *Neobenedenia* Yamaguti, 1963 were 1) accessory gland reservoir inside penis sac, 2) vagina absent, and 3) general orientation and path of ducts associated with the male and female reproductive tracts. Specific features were consistent with MacCallum (1927) and Whittington and Horton (1996). A specimen of *N. melleni* from the Florida pompano, *Trachinotus carolinus* (Linnaeus, 1766) (Carangidae), was deposited in the United States National Parasite Collection (USNPC) at Beltsville, Maryland, (USNPC No. 92528), and a specimen each from the bluering angelfish, *Pomacanthus annularis* (Bloch, 1787) (Pomacanthidae), the rock greenling, *Hexagrammos lagocephalus* (Pallas, 1810) (Hexagrammidae), and the blue-barred ribbon goby, *Oxymetopon cyanoctenosum* (Klausewitz and Condé, 1981) (Microdesmidae), was deposited there (USNPC Nos. 92529, 92530, 92531, respectively) and in the helminth collections of the H. W. Manter Laboratory (HWML) of the University of Nebraska State Museum at Lincoln, Nebraska, (HWML Nos. 16972, 16973, 16974, respectively). Common names of hosts follow those recommended by FishBase (see http://www.fishbase.org).

Results and Discussion

Information on the natural host range and geographic distribution of *N. melleni* could help aquaculture managers or aquarists prevent or control disease associated with infection by *N. melleni*. For example, if the proposed culture facility is sited within the geographic range of *N. melleni* and the fish species selected for culture is highly susceptible to infection by *N. melleni*, selection of another culture site or an alternate, possibly refractory fish species could reduce the risk of an epizootic. Regarding prophylaxis and treatment in aquaria, recognizing limits to the host range of *N. melleni* could allow aquarists to distinguish "low-risk" hosts, those that are refractive to infections, e.g., sharks, rays, and chimaeras (Chondrichthyes), from "high-risk" hosts, those that can serve as foci for captive infections, e.g., the Atlantic spadefish, *Chaetodipterus faber* (Broussonet, 1782) (Ephippidae), or some angelfishes (Pomacanthidae). Based on this information, low-risk hosts may be spared from exposure to potentially stressful anthelminthic chemicals, and high-risk hosts can either be isolated and treated aggressively or excluded altogether from large-volume, species-diverse exhibits. Furthermore, quarantine protocols based on such information may reduce treatment costs, especially if infected fishes reside in voluminous exhibit tanks. Herein, we report specimens of *N. melleni* from a new geographic locality and from three new hosts.

Regarding our new locality record, three adult specimens of *N. melleni* were collected from the skin of three of 31 (10%) adult Florida pompano (each 34–41 cm in total length) caught in July and August 2000 in the northern Gulf of Mexico off Horn Island, Mississippi. This is the first published record of *N. melleni* from a wild carangid. A gross lesion was not evident near the attachment site of *N. melleni*, and infected Florida pompano were grossly indistinguishable from those that were uninfected. *Neobenedenia melleni* was reported previously from Florida pompano both in the New York Aquarium (Jahn and Kuhn 1932, Nigrelli and Breder 1934, Nigrelli 1935, 1937) and in recirculating seawater tanks in Florida (Mueller et al. 1994). However, this is the first published record or confirmed report of *N. melleni* from a wild-caught fish in the Gulf of Mexico. Another report suggests that additional wild hosts reside in the Gulf of Mexico: Bullard et al. (2000) suspected red grouper, *Epinephelus morio* (Valenciennes, 1828) (Serranidae), and red snapper, *Lutjanus campechanus* (Poey, 1860) (Lutjanidae). In addition, Jahn and Kuhn (1932) suspected that *N. melleni* colonized the New York Aquarium by infected fishes that originally were caught off the Florida Keys or off Nassau, Bahamas, indicating that perhaps the parasite has ranged in the Gulf since the 1930s and that it was probably not artificially introduced to that region by the aquarium trade or aquaculture activities. Regarding the wide geographic distribution of *N. melleni*, Bullard et al. (2000) suggested the possibility that infections of *N. melleni* were vectored by infected remoras (Echeneidae), e.g., the whitefin sharksucker, *Echeneis neucratoides* Zouiev, 1786. However, the results of the present study suggest that, more simply, widely distributed hosts, e.g., Florida pompano ranging from the Atlantic Ocean off Massachusetts to off Brazil (Manooch, 1984), may transmit infections of this relatively non-host specific monogenean to endemic fishes throughout its range.

The low prevalence (10%) and intensity (1) of *N. melleni* in wild Florida pompano suggest that host schooling behavior may not facilitate horizontal transfer of *N. melleni*. Intuitively, the distance an

oncomiracidium must swim or crawl to infect a new host is negatively correlated with the chance of it infecting that host. As demonstrated by intense, debilitating infections among fishes crowded in aquaria, schooling behavior would seemingly increase the rate of infection and yield relatively intense infections because susceptible hosts are near each other. Despite this, our extremely limited data from wild Florida pompano did not support this notion. Alternatively, and from an evolutionary perspective, if host schooling behavior facilitated horizontal dispersal of monogenean larvae, the presumably strong selective pressure toward infecting schooling fishes could be evidenced by a species-diverse fauna of monogeneans or perhaps a relatively high intensity of infection on those fishes. However, there are seemingly conflicting reports regarding the matter of species diversity. Poulin and Rohde (1997) found no evidence that schooling fishes were infected by a greater number of gill- and skin-dwelling monogenean species. Santos and Carbonel (2000) considered 49 families of fishes in the midwestern and southwestern Atlantic Ocean and reported that jacks (Carangidae), drums (Sciaenidae), and tunas (Scombridae), all families that include schooling members, hosted the greatest number of monogenean genera: 18, 15, and 12, respectively. Additional collections of specimens of *N. melleni* from free-ranging schooling fishes could shed light on this question.

This report of *N. melleni* from Florida pompano is relevant to aquaculturists in the Gulf of Mexico because, as previously stated, *N. melleni* is a relatively non-host specific pathogen of fishes in aquaculture. Florida pompano is a candidate for culture in netpens or high density recirculating systems primarily because it is presently one of the most valuable table fish in tropical United States waters (Craig, 2000). *Neobenedenia melleni* (as *Neobenedenia girellae* [Hargis, 1955] Yamaguti, 1963) caused mass mortalities of caged amberjacks, *Seriola* spp. (Carangidae), in the Western Pacific Ocean off Japan (Ogawa et al., 1995), and a similar disease could occur in potential Florida pompano culture systems in the northern Gulf of Mexico. To lessen the risk of a captive epizootic, we advocate periodic and critical parasitological examinations of the skin, gills, eyes, and fins of several individual hosts sampled from the captive stock.

Regarding our new host records, specimens of *N. melleni* were also collected from the skin of a rock greenling at the Alaska SeaLife Center, Seward, Alaska. Although a concurrent infection occurred among three kelp greenling (*Hexagrammos decagrammus* [Pallas, 1810]) in a separate tank, a whitespotted greenling (*Hexagrammos stelleri* Tilesius, 1810) and several lingcod (*Ophiodon elongatus* Girard, 1854 [Hexagrammidae]) in the same tank as the infected rock greenling did not show signs of infection. The infected rock greenling was caught in Jakolof Bay (59 28' N; 151 32' W), Alaska, but where this fish and the kelp greenlings initially acquired the infection of *N. melleni* is not known. Whittington and Horton (1996) reported specimens of *N. melleni* from the gills of kelp greenling off San Juan County, Washington, making it plausible that *N. melleni* infects a closely-related, congeneric host north of that locality. The presence of *N. melleni* on a wild greenling off Alaska requires verification; however, if either of these greenlings was infected prior to capture, the northeast Pacific Ocean off Alaska would constitute both a substantial latitudinal range extension and the northern-most geographic locality record for *N. melleni*.

Thirteen specimens of *N. melleni* were collected from the skin of an adult bluering angelfish maintained in a 530,000 L exhibit tank at the Shark Reef Aquarium at Mandalay Bay, Las Vegas, Nevada. The infected bluering angelfish was emaciated and listless and possessed faded body coloration. Angelfishes have been previously identified as hosts for *N. melleni*, and Nigrelli and Breder (1934) stated that angelfishes were foci for epidemics because of their high susceptibility to infection. Nigrelli and Breder (1934) also reported *N. melleni* from the french angelfish, *Pomacanthus paru* (Bloch, 1787) and the gray angelfish, *Pomacanthus arcuatus* (Linnaeus, 1758) in the New York Aquarium.

Forty specimens of *N. melleni* were collected from the skin of two blue-barred ribbon gobies in a fish clearinghouse located in Hayward, California. This is the first report of *N. melleni* from a wormfish (Microdesmidae). Both hosts were flashing, emaciated, exuding excessive amounts of mucous which covered the body, and showing bloody lesions on the lower body surface near the pectoral fins. One of the blue-barred ribbon gobies died, presumably as a result of the infection. Although this fish ranges in the tropical western Pacific Ocean, we do not know the exact geographic origin of these individuals or where they were originally infected by *N. melleni*.

DNA sequence analysis of specimens of *N. melleni* collected from different oceans or from different host species could show that distinct parasite populations or morphologically similar species exist. For this reason and in addition to whole-mounts being deposited in a museum that will loan specimens, we advocate placing

specimens of *N. melleni* in both 95% molecular grade ethanol for eventual molecular analysis and 10% neutral buffered formalin for morphological studies using a light microscope.

ACKNOWLEDGMENTS

We thank Robin M. Overstreet (Gulf Coast Research Laboratory, Department of Coastal Sciences, The University of Southern Mississippi) for generously providing SAB with laboratory supplies and space, Jody Peterson and Jason Steckler (both Gulf Coast Research Laboratory) for catching Florida pompano, and Pam Tumoi, Millie Gray, and Mark Kansteiner (all Alaska SeaLife Center) for collecting specimens of *N. melleni* from the greenlings. Partial funding was provided by a National Oceanic and Atmospheric Administration, National Marine Fisheries Service award NA86FL0476 and a fellowship awarded to SAB by the Mississippi-Alabama Sea Grant Consortium.

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