Vol. 29: 237–240, 1997 | DISEASES OF AQUATIC ORGANISMS Dis Aquat Org | Published June 19

NOTE

Guanophoroma in the Red Sea silverside *Atherinomorus lacunosus* (Perciformes, Atherinidae)

A. Colorni*

Israel Oceanographic and Limnological Research, National Center for Mariculture, PO Box 1212, Eilat 88112, Israel

ABSTRACT: The occurrence of a spontaneous tumor affecting the guanophore iridocytes of the Red Sea silverside *Atherinomorus lacunosus* is described. The tumoral mass consisted mainly of yellowish, light-reflecting guanine platelets. The tumor infiltrated the fibers of the underlying muscle tissue, growing in whorls around them. A delicate network of capillaries provided the tumor with a blood supply. No viral particles were observed. This rare form of tumor is the first of this kind reported in a feral fish from the Red Sea.

KEY WORDS: Guanophoroma · Neoplasia · Iridocyte · Guanine · Atherinidae · Red Sea

This report describes a single occurrence of a spontaneous tumor affecting the guanophore iridocytes of the silverside *Atherinomorus lacunosus* Bloch & Schneider, 1801 (Perciformes, Atherinidae). The fish, a gregarious species that forms large schools along the shallow littoral of the Red Sea, has little or no commercial value but is an important forage fish for larger predators in the coral reef ecosystem (Ben-Tuvia 1968).

The specimen in question, 9 cm in length, displayed a large circular growth 8 mm in diameter, conspicuously raised from the body surface and located on its left side at approximately two-thirds of the body length (Fig. 1). The fish was caught by beach seine and was the only one displaying such a condition in over 1000 individuals of the same species examined over a period of 1 yr. The tumor and surrounding area were excised, fixed in buffered neutral formalin, embedded in paraffin and stained with hematoxylin-eosin (H&E) according to standard procedures (Sheehan & Hrapchak 1980). For electron microscopy, the tissue was reclaimed from the paraffin block, deparaffinized in xylene, re-hydrated in a descending alcohol series, stained en block with 2% osmium tetroxide and 0.5% uranyl acetate in veronal-acetate buffer, then dehydrated in an ascending ethanol series and propylene oxide, and finally embedded in Agar 100 resin according to standard procedures (Glauert 1975). Ultrathin sections were cut with a diamond knife and stained with lead citrate. Possible involvement of viral particles was investigated using a Jeol JEM 100-CX II Transmission Electron Microscope.

The tumoral mass consisted mainly of stubby, 8 ± 4 µm long, yellowish, light-reflecting platelets, readily soluble in acids and alkalis. These characteristics clearly identified the platelets as guanine. The emerging growth had broken through the skin, none of which covered the lesion. At its basal portion, the tumor appeared to invade the underlying muscle tissue, separating fiber bundles from one another and growing in whorls around them (Figs. 2 & 3). Because of the brilliance of the platelets in histological sections, cytological details could be observed only after the platelets had been dissolved by dipping the slides in a solution of either 1 N HCl or 10% KOH. The neoplastic cells within the tumor mass had a pleomorphic appearance and displayed large, elongate, granular nuclei surrounded by scarce cytoplasm. The anarchic formation of the guanine platelets apparently caused the rapid destruction of most of the iridocytes that had produced them. Sparse aggregates of melanin-containing cells and free melanin pigment were also present. A delicate network of capillaries provided the tumor with a blood supply (Fig. 4). No virus-like particles were observed in the tumor. No metastasis sites were found.

This type of neoplasia was called 'guanophoroma' by Takahashi (1929), who first described it in the Japanese greenling *Hexagrammus otakii*, a marine species. Stolk (1959) reported a similar tumor in 6 specimens, all from the same brood, of the characid *Ctenobrycon*

*E-mail: angelo@ocean.org.il

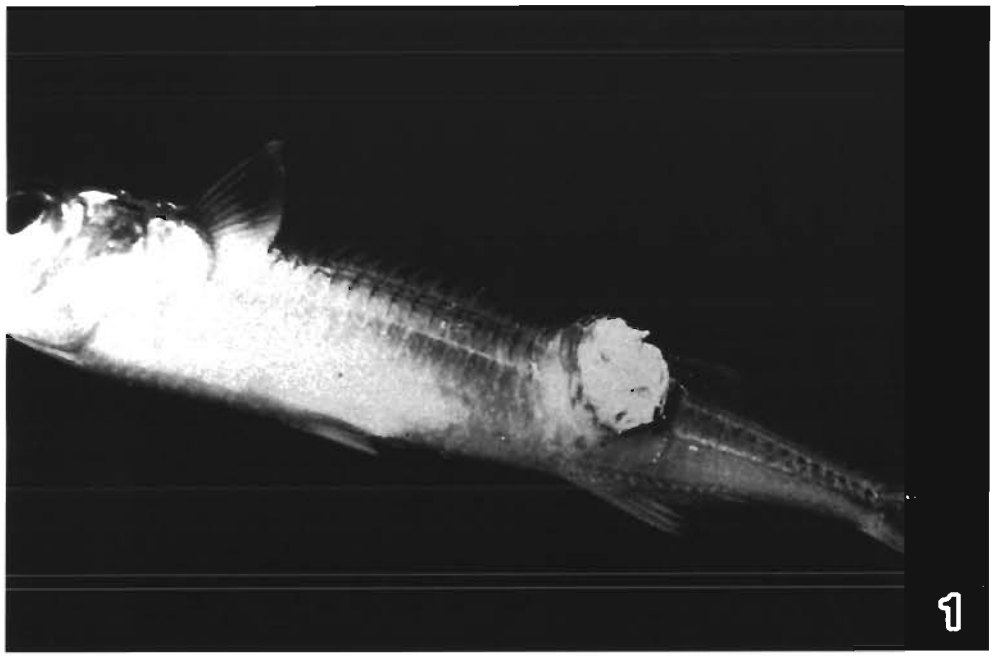

Fig. 1. *Atherinomorus lacunosus* with large guanophoroma between the second dorsal and the anal fins

spilurus, a freshwater species. The term 'iridophoromas' was preferred by Matsumoto et al. (1981) for tumors with iridescent and/or reflecting substances. Iridophoromas, sometimes mixed with melanophoromas to form single neoplastic lesions (irido-melanophoromas), were described by Kimura et al. (1984) in the nibe croaker *Nibea mitsukurii* inhabiting the Pacific coastal waters of Japan. Using biopsy material from these cancerous croakers, Matsumoto et al. (1981) established permanent cell lines and studied the mode of differentiation of the neoplastic pigment cells *in vitro*.

Studies of piscine tumors are of importance. Wild fish can serve as sentinels for environmental carcinogenic agents potentially harmful to humans or other terrestrial vertebrates; captive fish can serve as experimental models for chemically or virally induced neoplasms, and the comparative analysis of the biology of neoplastic diseases in fish and higher vertebrates can help in elucidating the causes and mechanisms of carcinogenesis (Kimura et al. 1984, Hayes & Ferguson 1989, Kent & Fournie 1993).

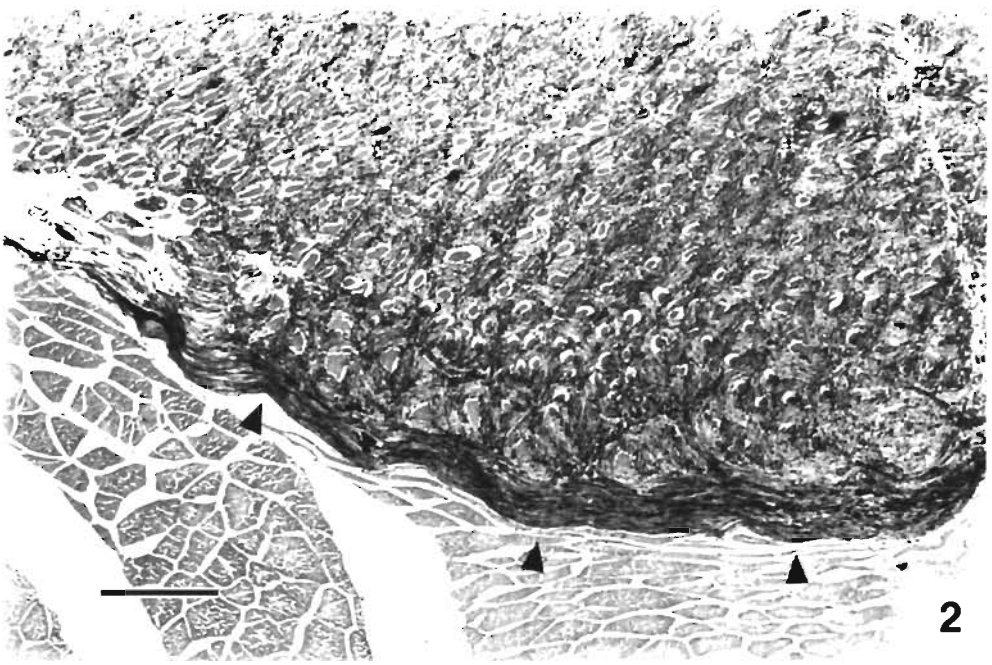

Fig. 2. *Atherinomorus lacunosus*. Interface of the tumor (arrowheads) with the underlying muscle tissue. H&E stained. Scale bar = 200 µm

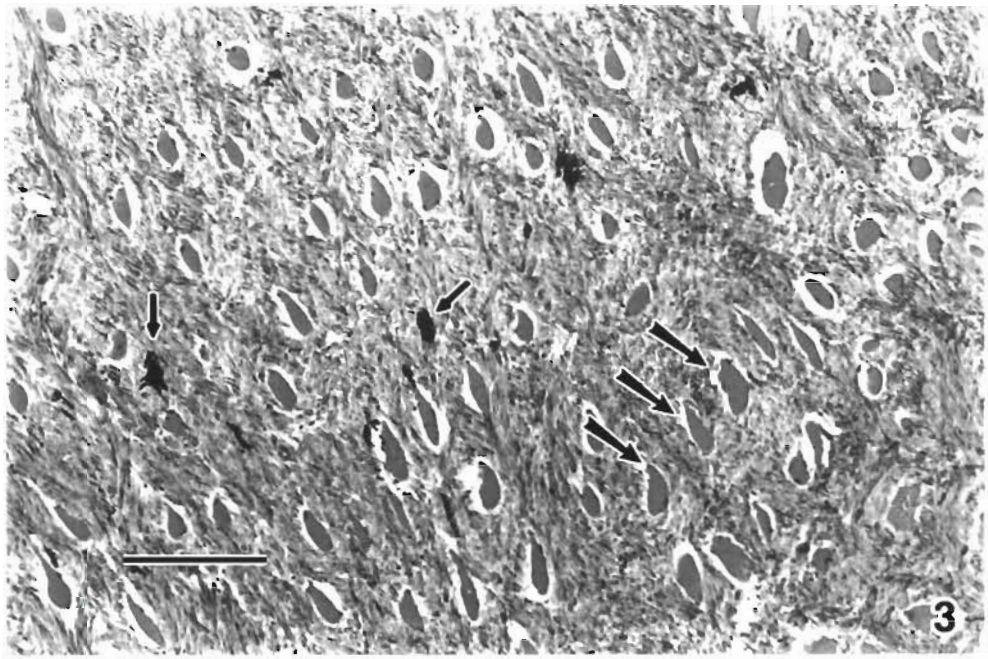

Fig. 3. *Atherinomorus lacunosus*. Muscle fibers (parallel arrows) isolated by whorls of guanine platelets. The short arrows point to aggregates of melanin-containing cells. H&E stained. Scale bar = 100 µm

Tumors involving guanine may be of particular importance in cancer research. Guanine is the preferred base of attack in the DNA by known carcinogens such as aromatic amines and related molecules; aberrant guanine can cause mutations in the genetic material eventually leading to cancer (Taussig 1984). This nucleotide, in an altered form, was found by Malins & Haimanot (1990) in the DNA from neoplastic livers of the English sole *Parophrys vetulus* that had been exposed to aromatic hydrocarbons. Their findings support the hypothesis that when attacked by a carcinogenic agent modified purine nucleotides play a crucial role in tumor formation. Purines and reflecting platelets are well-defined and readily identifiable phenotypic markers of fish iridocytes (Matsumoto et al. 1981), which normally contain purines, primarily guanine, as a breakdown product of nucleic acids (Lagler et al. 1977). In our case guanine was the most conspicuous end product of an uncontrolled proliferation of guanophore iridocytes. As guanophoromas have rarely been observed, little is known about this type of tumor (Peters 1984) and any conjecture about the pathway of their formation remains highly speculative.

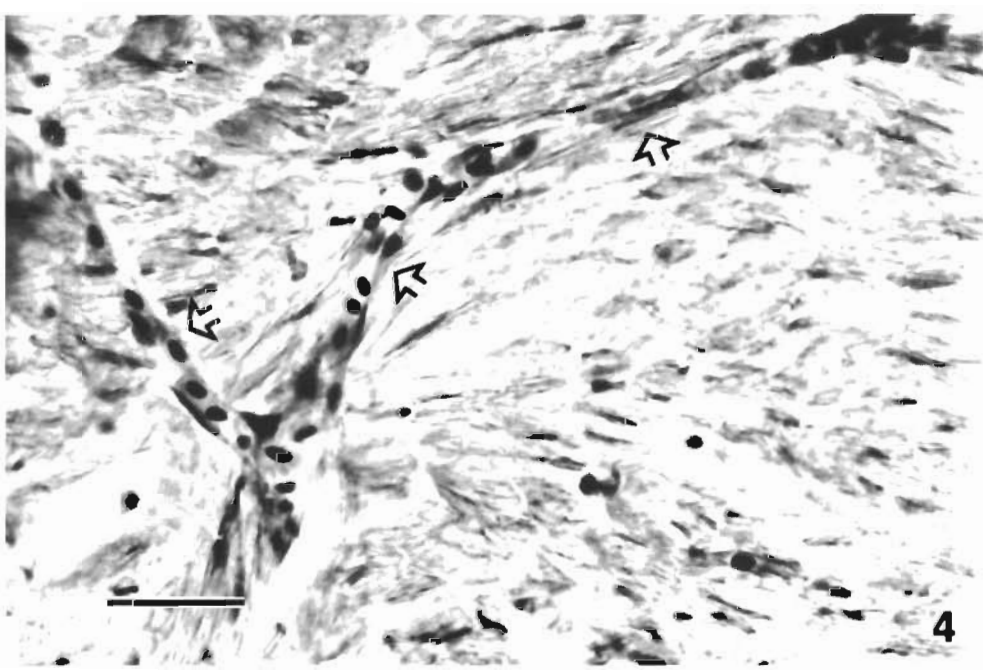

Fig. 4. *Atherinomorus lacunosus*. Blood capillaries (arrowed) in the tumoral mass. H&E stain. Scale bar = 20 µm

The only other record of neoplasia from a fish indigenous to the Red Sea relates to a mandibular lesion in the 2-bar sea bream *Acanthopagrus bifasciatus*, classified as an ameloblastoma by the Registry of Tumors in Lower Animals of the Smithsonian Institution, Washington, DC (RTLA #3912) (Diamant & Colorni, I.O.L.R. Internal Annual Report, 1987). The fish had been held captive for several years in a large display tank at the Eilat Coral World, Israel. Further reports of piscine tumors from the Red Sea refer to the gilthead sea bream *Sparus aurata* (Paperna et al. 1977, Nash & Porter 1985) and European sea bass *Dicentrarchus labrax* (Colorni 1990). Both species of fish, however, were originally imported for mariculture purposes from the Mediterranean Sea.

Acknowledgements. I am endebted to Dr A. Diamant for the electron microscopy analysis and critical comments. B. Colorni's technical expertise in the histological work was invaluable and is greatly appreciated. Prof. N. Peters of the Zoologisches Institut der Universität Hamburg, Germany, provided me with relevant literature and his assistance is gratefully acknowledged. This study was funded by the Israeli Ministry of National Infrastructures.

LITERATURE CITED

Ben-Tuvia A (1968) Report on the fisheries investigations of the Israel South Red Sea Expedition, 1962. Bull Sea Fish Res Stn Haifa 33:21–55

Colorni A (1990) Pathology of marine warmwater finfish in Israel: problems and research. Advances in Tropical Mariculture, Tahiti, February 20–March 4, 1989, AQUACOP, IFREMER, Plouzané, Actes de Colloque 9, p 133–142

Glauert AM (1975) Fixation, dehydration and embedding of biological specimens. Elsevier North-Holland Biomedical Press, Amsterdam

Haynes MA, Ferguson HW (1989) Neoplasia in fish. In: Ferguson HW (ed) Systemic pathology of fish. Iowa State University Press, Ames, p 230–247

Kent ML, Fournie JW (1993) Importance of marine fish diseases—an overview. In: Couch JA, Fournie JW (eds) Pathobiology of marine and estuarine organisms. Advances in fisheries science. CRC Press, Boca Raton, FL, p 1–24

Kimura I, Taniguchi N, Kumai H, Tomita I, Kinae N, Yoshizaki K, Ito M, Ishikawa T (1984) Correlation of epizootiological observations with experimental data: chemical induction of chromatophoromas in the croaker, *Nibea mitsukurii*. Use of small fish in carcinogenicity testing. National Cancer Institute Monograph No. 65, US Dept. Health and Human Services, Bethesda, MD, p 139–154

Lagler KF, Bardach JE, Miller RR, May Passino DR (1977) Skin. In: Lagler KF, Bardach JE, Miller RR, May Passino DR (eds) Ichthyology, 2nd edn. J Wiley & Sons, New York, p 104–128

Malins DC, Haimanot R (1990) 4,6-Diamino-5-formamidopyrimidine, 8-hydroxyguanine and 8-hydroxyadenine in DNA from neoplastic liver of English sole exposed to carcinogens. Biochem Biophys Res Commun 173(2): 614–619

Matsumoto J, Ishikawa T, Masahito P, Oikawa A, Takayama S (1981) Multiplicity in phenotypic expression of fish erythrophoroma and irido-melanophoroma cells *in vitro*. In: Dawe CJ et al. (eds) Phyletic approaches to cancer. Japan Sci Soc Press, Tokyo, p 253–266

Nash G, Porter C (1985) Branchial osteochondroma in a gilthead sea bream *Sparus aurata* L., cultured in the Gulf of Aqaba. J Fish Dis 8:333–336

Paperna I, Colorni A, Gordin H, Kissil GWm (1977) Diseases of *Sparus aurata* in marine culture at Elat. Aquaculture 10:195–213

Peters N (1984) Diseases caused by neoplasia. In: Kinne O (ed) Diseases of marine animals, Vol. IV. Biologische Anstalt Helgoland, Hamburg, p 400–423

Sheehan DC, Hrapchak BB (1980) Theory and practice of histotechnology, 2nd edn. CV Mosby Co, St. Louis, MO

Stolk A (1959) Tumours of fishes. XXVII. Guanophoroma in the characid *Ctenobrycon spilurus* (Valenciennes). Proc K Ned Akad Wet (Ser C Biol Med Sci) 62C:155–162

Takahashi K (1929) Studie über die Fischgeschwülste. Z Krebsforsch 29:1–73

Taussig MJ (1984) Neoplasia. In: Taussig MJ (ed) Processes in pathology and microbiology, 2nd edn. Blackwell, Oxford, p 689–809

Responsible Subject Editor: N. Peters, Hamburg, Germany

Manuscript first received: December 20, 1996
Revised version accepted: April 15, 1997