

A Localized Melanoma in a Hybrid Fish *Lebistes* × *Mollienesia*

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The most commonly reported pigment-cell tumor in fishes living under natural conditions is the melanoma; it has been described from species representing eleven orders of elasmobranchs and teleosts (5, 15). Melanomas have also been produced experimentally by three independent investigators and innumerable aquarists in the hybrid offspring of the black spotted platyfish (*Xiphophorus maculatus*) and the swordtail (*X. helleri*). One of the special features of this experimental melanoma is that it is initiated genetically—primarily by a dominant gene for macromelanophores (3). In the absence of the hereditary macromelanophores, melanomas do not develop in platyfish-swordtail hybrids (5). Platyfish, swordtails, and their hybrids also have a smaller type of pigment cell known as the micromelanophore, the presence of which, alone, does not favor the development of melanomas in their hybrids (3, 5, 6). Melanophores of fishes are pigment-effector cells; they are not pigment-carrying macrophages or melanophages (6, 7).

In contrast to the melanomas of platyfish-swordtail hybrids which are invariably associated with macromelanophores, the melanophores of other species have not been studied relative to their possible relationship to the development of melanomas. It was therefore fortunate that we obtained a melanomatous hybrid fish of two other distinct species, the black mollie (*Mollienesia sphenops*) and the guppy (*Lebistes reticulatus*). We have also obtained several color varieties of guppies and mollies and have found that they possess only micromelanophores. The completely black mollies achieve their intensely dark coloring by virtue of an excessive number of micromelanophores. The pigmentation became so intense in one tumorous *Mollienesia velifera*, described by Ermin

(1), that it resembled a state of melanosis produced by macromelanophores (5). The genetic difference between the black and the gray mollie is complex (4), but there is nothing in its inheritance comparable to the macromelanophore alleles of the platyfish.

HISTORY AND GROSS ANATOMY

The specimen was a male, a first-generation hybrid produced by mating a guppy (*Lebistes reticulatus*) female and a black mollie (*Mollienesia sphenops*) male. At 18 months the hybrid measured 5.6 cm. in total length. It was colored a matte black, with a few faintly iridescent greenish spots. It had a lobulated black tumor, approximately 6 × 6 × 3 mm., which arose by a broad pedicle from the right dorsolateral aspect of the cranium (Fig. 1). The epidermis was smooth and intact except for a small ulcerated area on the upper surface of the tumor.

The guppy-mollie hybrid was a member of a brood of eleven and was one of three obtained from Mr. G. W. Rose, an aquarist. When the aquarist attempted to mate the guppy-mollie hybrids back to either a guppy female or a mollie female, the matings failed. When one of the black hybrids was 15 months old, it developed a slow-growing melanotic tumor on the side of the head. Three months later the fish became sluggish and ate only occasionally. It was fixed in 10 per cent formalin.

The authenticity of the alleged guppy-mollie hybrid was verified by Mr. Donn Eric Rosen, who compared its gonopodium with those of the pure guppy and of the pure mollie. The hybrid's gonopodium was definitely intermediate. It also closely resembled the *Lebistes* × *Mollienesia* F₁ hybrid obtained experimentally by Samokhvalova (14) by means of artificial insemination and one illustrated by Innes (12) and verified by Dr. Carl L. Hubbs.

MICROSCOPIC APPEARANCE

The tumor extended from the epidermis down through the dermis to the outer surface of the

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cranium (Fig. 2). The epidermis covering the tumor was somewhat thickened at places, and infiltrated by small and large melanin-laden macrophages (Fig. 3). Medially, the tumor just overlay the midline, while laterally it extended to the base of the right gill cover. Local infiltration of the tissues surrounding the scales was evident, and these were lifted from their normal position (Figs. 2, 4). An abundance of melanin pigment present, both in the cells and in the stroma of the tumor, almost completely masked the histological and cytological details (Figs. 4, 5). No secondary growths were found in the internal organs.

The cytological elements show considerable pleomorphism. Some of the pigmented cells (melanophores) in the superficial parts of the growth are very large and show one or more faintly stained vesicular nuclei, with one or two prominent nucleoli and a faint basic chromatin network. Large irregular masses of cytoplasm surround these nuclei. In the deeper parts of the tumor small, round cells have dark, almost pyknotic nuclei and abundant melanin in the cytoplasm; these cells alternate with whorls of spindle-shaped cells also containing pigment (Figs. 6, 7); the latter, seen in a bleached section, we presume, are melanocytes (Fig. 8). The growth is highly vascular and studded with numerous fine-walled capillaries. In many cases it is impossible to discern the walls of these vessels because of the heavy deposition of melanin granules (Fig. 5).

DISCUSSION AND SUMMARY

While the melanomas of platyfish-swordtail hybrids involve the macromelanophores (3), the melanomas of other species, including that of the mollie-guppy hybrid, apparently arise without the presence of homologous large black chromatophores. The pigment cell that is basic to the actual formation of melanoma, however, is not a melanophore, either large or small, because these are mature cells, but it is the melanocyte—that is, the

postembryonic pigment cell (9). Indeed, the melanocyte is the basic pigment-cell type in the melanomas of all vertebrates (5); melanocytes have a common embryological site of origin, namely, from the neural crest (6) or neural tube (10). According to the results of experiments and observations in genetics (3, 7), in the cytology and cytochemistry of tissue-cultured cells (9, 10), in transplantation (13), in regeneration (2), and in experimental embryology (11), it has been determined that some of the melanocytes in platyfish-swordtail hybrids transform into macromelanophores, others into micromelanophores. Thus far, only those melanocytes that develop into macromelanophores have produced melanomas in xiphophorin hybrids. But it is now clear that melanocytes that transform into micromelanophores in mollies, in mollie-guppy hybrids, and in other fishes may also exhibit atypical growth and produce melanomas.

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FIG. 1.—Molliesia × Lebistes hybrid (5.6 cm.) with a melanoma on the cranium.

FIG. 2.—Transverse section of the melanoma of the mollie-guppy hybrid showing the intensity of the natural melanin pigmentation. In this section the melanoma is divided by a scale in the head region. ×11.

FIG. 3.—The epidermis (upper lighter layer) is thickened in some areas and infiltrated by melanin-laden macrophages. The dermal layer below contains melanophores, melanocytes, and melanin-laden macrophages. ×500.

FIG. 4.—Section of a scale and its dermal pocket within the

tumor mass. Above the scale, the pigment cells are interpreted to be mostly melanocytes, although a few macrophages are close to the thin epidermal layer. Below the level of the scale, the pigment cells are more darkly pigmented, and they have the appearance of mostly melanophores in mass formation. ×130.

Note: In the descriptions the use of the following terms: melanocytes, melanophores, and macrophages (melanophages) are in accordance with the recommendations made at the Third Conference on the Biology of Normal and Atypical Pigment Cell Growth (7).

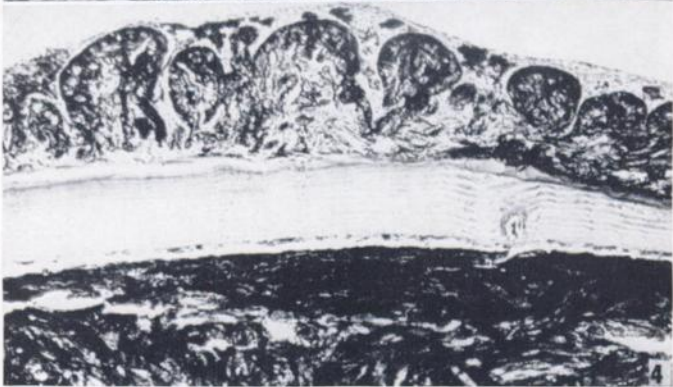
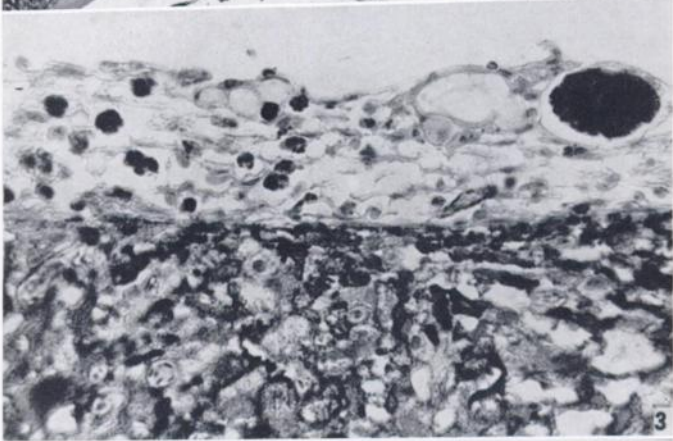


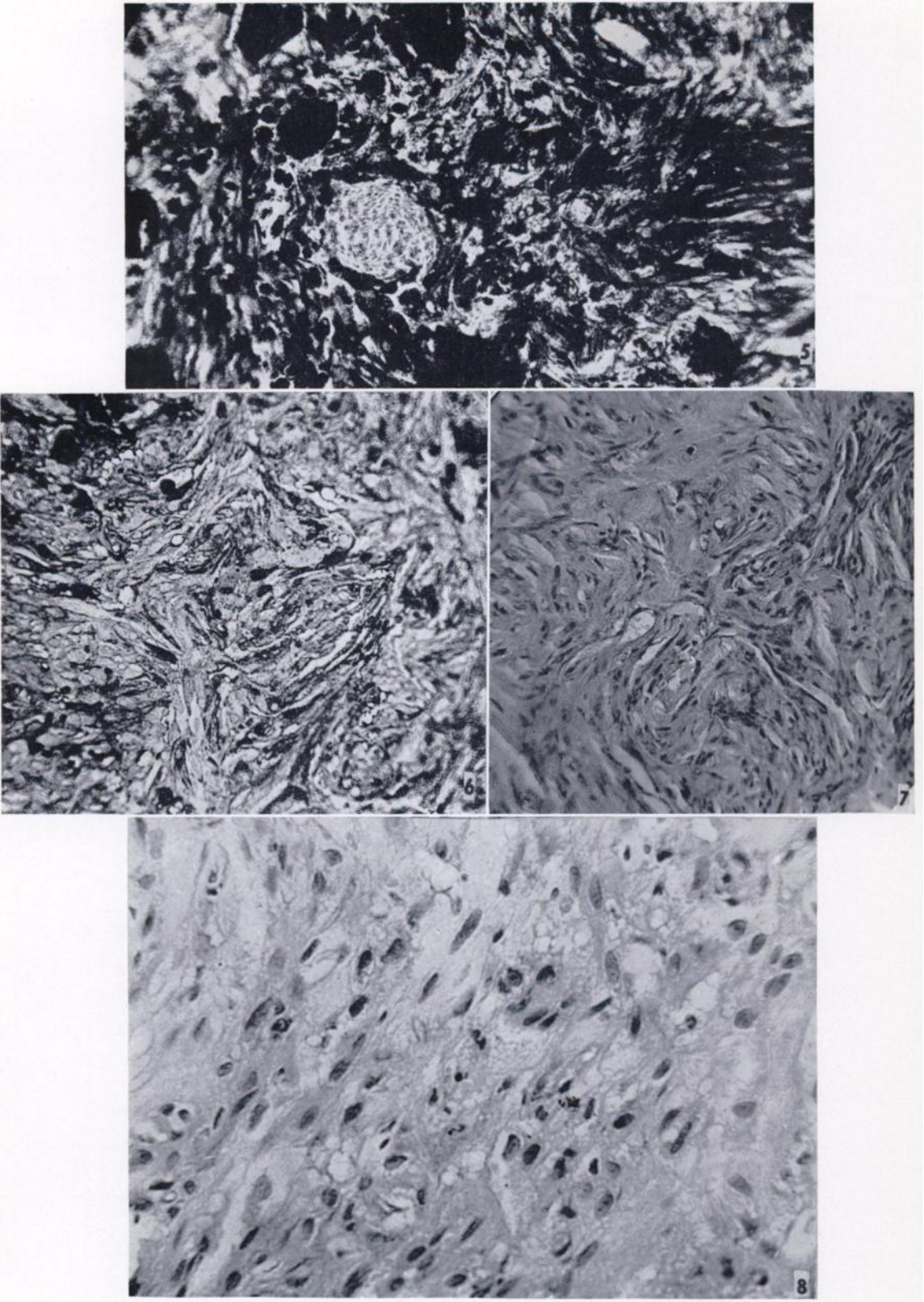
FIG. 5.—The melanoma is heavily pigmented; the cellular details are obscured for the most part, but note the blood vessel which is surrounded by melanin-bearing cells, some of which are melanocytes. The large, round, solid black bodies probably represent melanin-laden macrophages (melanophages). $\times 300$.

FIG. 6.—Deeper parts of the tumor showing whorls of spindle-shaped cells which are interpreted to be melanocytes. $\times 300$.

FIG. 7.—Generally the same as Figure 6 but from a bleached section of the whorled area and showing more cellular details. $\times 300$.

FIG. 8.—Spindle-shaped cells presumed to be melanocytes in a bleached section of the melanoma. $\times 560$.

Photomicrographs, Figs. 5-8, by Pamela Alexander, New York Zoological Society.



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