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Iridophoroma in a European grayling (*Thymallus thymallus*) in Switzerland

H. Schmidt-Posthaus *, D. Pugovkin and T. Wahli

Centre for Fish and Wildlife Health, Institute of Animal Pathology, University of Bern, Laenggass-Strasse 122, CH-3012 Bern, Switzerland

Abstract

A European grayling (*Thymallus thymallus*) found dead in a small subalpine Swiss river showed a whitish well demarcated mass on the right body site. Histologically, this tumor consisted of densely packed fusiform neoplastic cells with olive-green crystalline intracytoplasmic structures. Based on these findings an iridophoroma was diagnosed. Until now this is a solitary case suggesting a spontaneous development of this neoplasm.

Integumentary colors in vertebrates are primarily dependent on the presence of specialized pigment cells, chromatophores, in the skin. In fish these cells occur in the dermis, where they can be found in the stratum spongiosum, in the hypodermis, or in both (Fujii, 1993). They derive from the neural crest and are categorized as paraneurons (Fujii, 1993). The classification of chromatophores is based on the color of the pigment they contain. Additionally, they are classified into dendritic and non-dendritic cell types. Dendritic chromatophores include the melanophores, erythrophores, xanthophores, and leucophores. Iridophores belong to the non-dendritic cell type and acquire a round to oval, or polygonal shape, although some iridophores develop dendritic processes (Elliot, 2000). Iridophores are responsible for the silver or blue iridescence of the skin. They contain large crystalline platelets that tend to form stacks in the cytoplasm, with uniform spacing between adjacent platelets within a stack (Fujii, 1993). Each platelet is a monoclinic crystal composed mainly of guanine, but it may also contain purines such as hypoxanthine and uric acid (Fujii, 1993).

Skin chromatophoromas are tumors arising from dermal chromatophores of fish, amphibians and reptiles (Masahito, 1989). The subclassification of chromatophoromas is primarily based on identification of specific pigment organelles (Okihiro, 1988, Baumann & Okihiro, 2000). Melanophoromas, the most common tumor type encountered, are diagnosed via detection of melanosomes by light and electron microscopy. Pigments of xanthophoromas and erythrophoromas are undetectable with routine histological processing and staining. Ultrastructurally, lipid and pterisomes can be identified (Elliot, 2000). Iridophoromas are characterized at the light microscopic level by the presence of olive-green granular pigment, which is birefringent with polarized light.

The river Saane is located in a subalpine region in the northern part of Switzerland.

* Corresponding author's email: heike.schmidt@itpa.unibe.ch

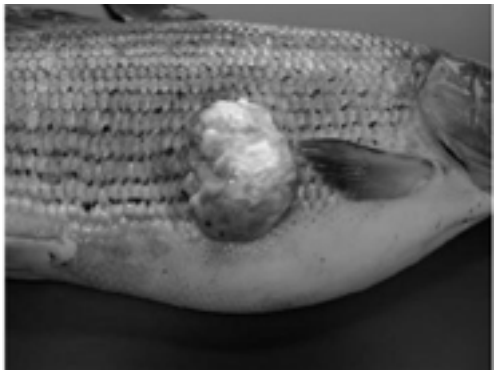

Figure 1. Tumor mass on the right body site.

Besides a stable brown trout population a population of grayling lives in the Saane. The region is mainly dominated by agriculture and small settlements. Further, three sewage treatment plants release their effluents into the river. In April 2001 a grayling found dead by fishermen in the river was sent to the Centre of Fish and Wildlife Health for diagnosis. The grayling was female with a total length of 55 cm and in a good body condition. A macroscopical examination of the exterior and the inner organs was performed. Macroscopically, on the right body site there was a whitish oval mass, 6 x 4 cm in diameter (Figure 1). It was well demarcated and raised above the surface of the skin. The overlying epidermis was slightly ulcerated. No additional pathological lesions or tumor metastases were found. The inner organs were normal in shape and color and the intestine was filled with food. The cause of death remained unclear.

Samples of the tumor were fixed in Bouin's solution for 24 hours, routinely processed for histology and stained with haematoxylin and eosin (HE) and van Gieson (VG).

Histologically, the mass originated from the pigment layer in the dermis (Figure 2a). The mass was partly demarcated and encapsulated (Figure 2b). In few areas the neoplastic cells were infiltrating into the surrounding tissue (Figure 2a, b). The tumor consisted of fusiform cells, which were arranged in bundles (Figure 2d). They had a moderate amount of cytoplasm containing olive to green, crystalline and birefringent pigment (Figure 2e). The nuclei were round to ovoid with one to two nucleoli (Figure 2e). No mitotic figures were visible. VG staining did not specifically stain tumor cells. The epidermis present and normally structured at the sides of the mass was completely eroded on the surface of the tumor. At this location fungal hyphae typical for *Saprolegnia* sp. covered the tumor mass. In the dermis adjacent to the tumor a severe edema was present. From the light microscopic picture an iridophoroma was diagnosed.

In this report we describe a mesenchymal skin neoplasm in the dermis of a wild grayling. These dermal tumors can either arise from connective tissue, peripheral nerve tissue or pigment cells. Chromatophoromas are common fish tumors and large epizootics in marine and freshwater fish have occurred worldwide (Baumann & Okihiro, 2000). However, iridophoromas are rather rare compared with melanomas and erythrophoromas (Masahito et al., 1989). The etiology of chromatophoromas is generally obscure. The pigment cell tumors may occur in combination with hereditary, carcinogenic and/or aging factors (Masahito et al., 1989). Epidemiologic surveys from many studies of chromatophoroma epizootics are at the least

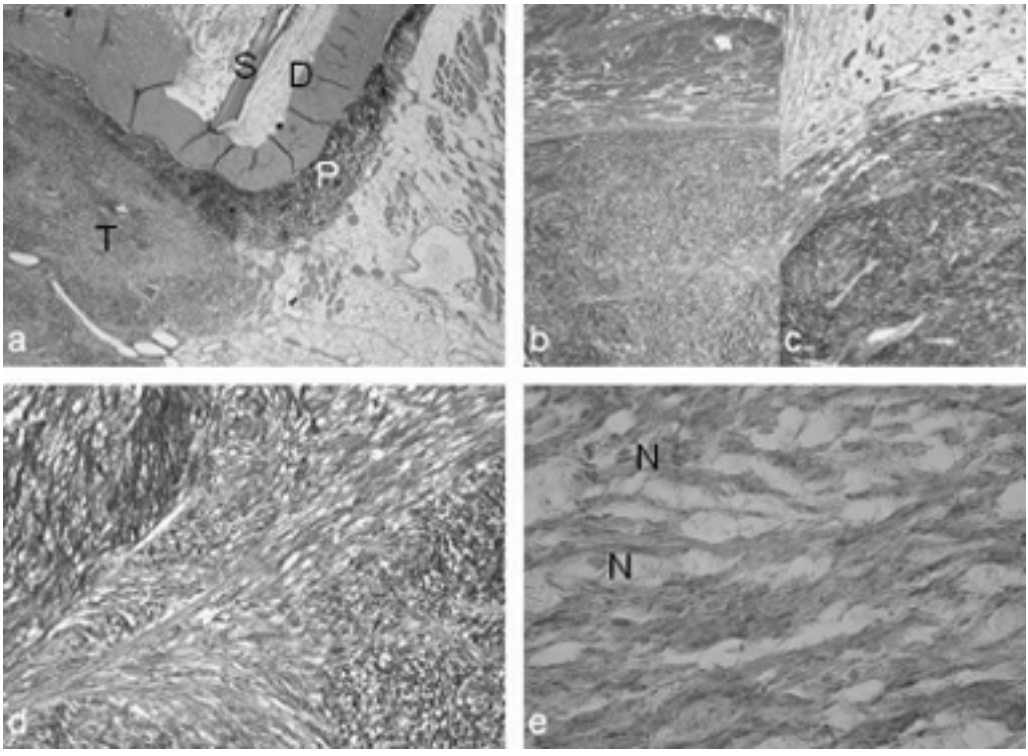

Figure 2. (**a**) Tumor mass (T) arising from the pigment layer (P). Scale (S). Dermis (D) (magnification: x40). (**b**) In some locations tumor mass sharply demarcated against the surrounding tissue and encapsulated by a small fibrous layer or (**c**) infiltrating in the surrounding tissue (magnification: x40). (**d**) Tumor cells arranged in bundles (magnification: x100). (**e**) Cells with moderate amount of cytoplasm containing crystalline pigment. Nuclei round to ovoid (N) with one to two nucleoli (magnification: x 400).

suggestive of possible exposure to anthropogenic carcinogens (Kimura et al., 1989, Okihiro, 1988 Okihiro et al., 1993). In our case, the river Saane is influenced by sewage treatment plant effluents and runoff of agricultural areas. Nevertheless, until now this is an isolated case of a pigment cell tumor in that river which makes an association with environmental pollution factors improbable. Other potential etiologies include genetic predisposition and ultraviolet radiation (Vielkind & Vielkind, 1982) and oncogenic viruses. Viruses from the families herpesvirus, adenovirus, papovavirus and retrovirus have been identified as the cause of neoplasms in fish (Anders & Yoshimizu, 1994). Dermal sarcomas are described in association with walleye dermal sarcoma virus, a retrovirus (Poulet et al., 1995, 1996). Retroviruses are also involved in damselfish neurofibromatosis, characterised by multiple nerve sheath tumors and cutaneous chromatophoromas (Schmale et al., 1986, 1996). Nevertheless, the single occurrence of the neoplasm in our case favours a spontaneous development of the tumor rather than infectious agents in the grayling.

Infection with *Saprolegnia* sp. is often associated with previous trauma (Roberts, 2001). In this case the iridophoroma raised above the surface of the skin and the overlying epidermis was multifocally ulcerated, probably due to trauma. As epidermal erosion is also described with *Saprolegnia* sp. infection it is difficult to evaluate the final cause of erosion at this stage (Roberts, 2001). The dermal edema adjacent to the tumor is probably induced by the *Saprolegnia* sp. infection as dermal necrosis and edema are described with fungal infection of the skin (Roberts, 2001). Additionally, vessel compression by the neoplasm can also have contributed to development of dermal edema.

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