

## Grayish spot on goldfish chest: An explanation

Minoru Kobayashi, Yuichi Sasayama, Ajai K. Srivastav\*, and Nobuo Suzuki

*Noto Marine Laboratory, Faculty of Science, Kanazawa University, Ishikawa 927-0553, Japan, and \*Department of Zoology, University of Gorakhpur, Gorakhpur, 273 009, India.*

**Abstract** We observed that most adult goldfish has a tiny spot on the chest. Histological examination revealed that the area appears as a spot because it lacks the guanophore layer under the scales. In the connective tissue layer under the spot, neurons were recognized, with axons extending to the epidermis. Furthermore, cartilage was present under the spot. This was the coracoid process composing the shoulder girdle. A possible explanation for this spot will be discussed.

### Introduction

Goldfish are popular fish as it is easy to maintain as a pet. Besides, goldfish are used as experimental animals because of their tolerance to various treatments (Ueda *et al.*, 1985; Himick and Peter, 1994; Sasayama *et al.*, 1995). During our experimental studies, we found that in most grown goldfish, there is a tiny grayish spot on the chest. Initially, we assumed that the spot was the result of a simple injury, as that portion of the chest seemed to frequently touch the bottom of the glass aquarium. In this study, we performed anatomical and histological investigations of this spot and provide an explanation for the presence of this spot.

### Materials and Methods

Goldfish (*Carassius auratus*) of various sizes were purchased and maintained on artificial fish food *ad libitum* under natural conditions.

The spot was at first anatomically examined under a binocular dissecting microscope in 62 goldfish (body weight (BW) 5.1–16.8 g; total length (TL) 5.1–8.3 cm) of both sexes. The clarity of the spot was evaluated in four ranks as follows: rank 0 (no spot), rank 1 (faint spot), rank 2 (ordinary spot) and rank 3 (very clear spot). The spot was then histologically examined. The spot area was dissected out and immediately fixed in Bouin's solution for 24 hrs. The tissue was then dehydrated and embedded in paraffin and cut in 8- $\mu$ m-thick sections according to a routine method. The sections were stained with hematoxylin and eosin,

or with Bodian's silver method modified by Otsuka (1962), for detecting neurons.

Student's *t*-test was used to evaluate the numerical data.

### Results and Discussions

#### *Anatomical observations*

The spot was irregular circle, and the edge was rough and only about 1 mm in diameter even in large goldfish (Fig. 1). In small fish, less than 5 cm in TL, the spot was absent or obscure. In 62 goldfish over 5 cm long, 57 had the spot. The clarity of the spot was not related to the sex or body size (Fig. 2). Five fish (2 males and 3 females), which constituted 8% of the goldfish examined, had no spot. These observations suggest that there are individual differences in the clarity of the spot, although it appears that the spot is formed after the goldfish attain a certain size.

Furthermore, the spot was not a result of injury, as normal scales were observed under the spot. Therefore, the spot does not lead to a hole. Cartilage was present just under the spot. This was the coracoid process of the shoulder girdle which supports the pectoral fin (Fig. 3).

#### *Histological observation*

It was noted that average number of mucous cells in the epithelium was significantly decreased ( $p < 0.01$ ) in the spot area compared with the surrounding areas, in 11 goldfish (BW: 7.2–10.4 g; TL: 6.4–7.9 cm) (Figs. 4 and 5). Although the physiological significance of this finding is not clear, it is probable that the epidermis at the spot must not become thin by the shedding of mucous cells.



Fig. 1. Ventral view of goldfish exhibiting the grayish spot (arrowhead) present on the chest. Bar indicates 10 mm.

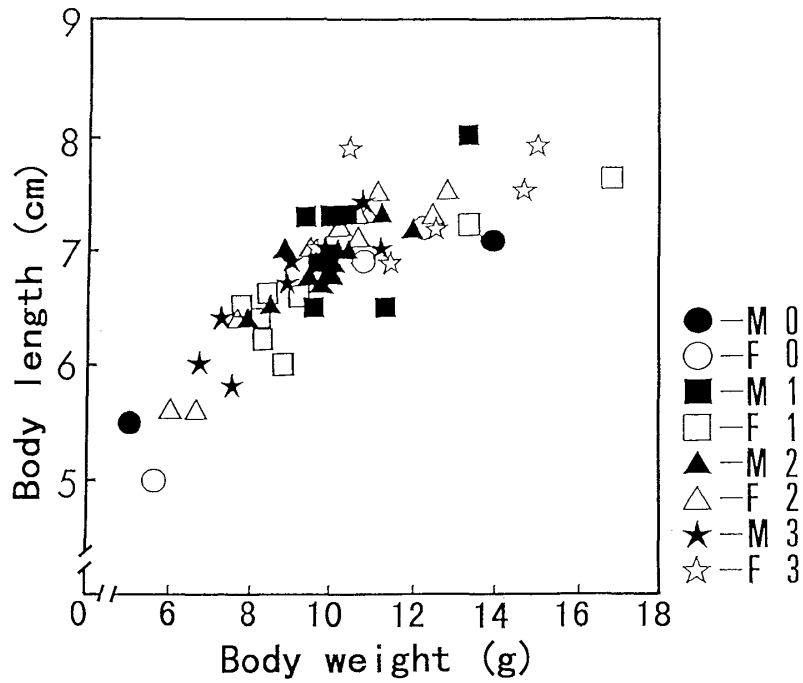


Fig. 2. Clarity of the spot in various sizes of goldfish. Numbers (0-3) refer to the degree of clarity. M: male; F: female.

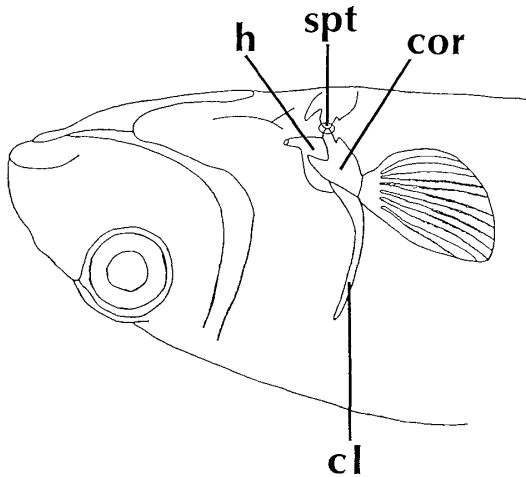


Fig. 3. Anatomy of the spot and the related areas. spt: spot; cor: coracoid; h: heart; cl: clavicle.

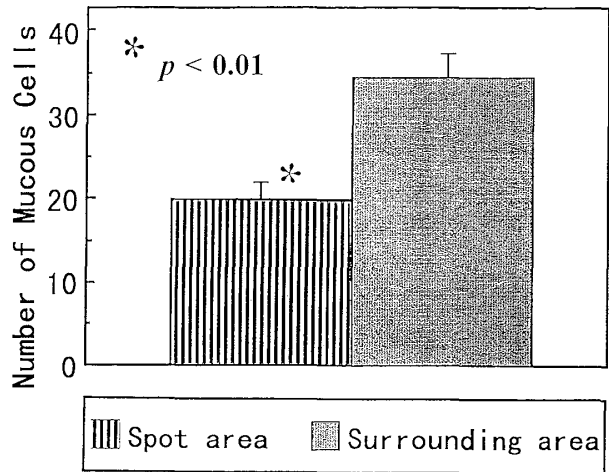


Fig. 4. Comparison of the number of mucous cells in the epithelium, at the spot area and the surrounding areas.

Alternatively, this area may avoid muddiness with mucus on account of the reason below-mentioned. That is, the most striking characteristic of the spot area is that no guanophore layer is present in the dermis (Fig. 5), notwithstanding the fact that guanophores are distributed over the entire abdominal aspect of the goldfish. Therefore, the spot looks grayish, as light is not reflected. Although in the area surrounding the spot, connective tissue is composed of loose fibers (Fig. 6), it is more compact at the spot (Fig. 7). Neurons were identified in the connective tissue layer just above the spot by silver staining (Figs. 8a,b). The cell

bodies of the neurons were located in the deep part of the connective tissue, and neuronal axons extended to the epidermis. These neurons probably function as a kind of photoreceptors for sensing light coming from underneath, because of the reduced number of guanophores in the dermis at this area. It is known that in birds, some neurons in the brain are perceptive to photons (Parry, 1993). An electron microscopic study is needed to confirm this. Alternatively, the neurons probably transmit the physical pressure of the coracoid cartilage to the epidermis; this protects the heart which is present inside a cartilaginous basket. It is

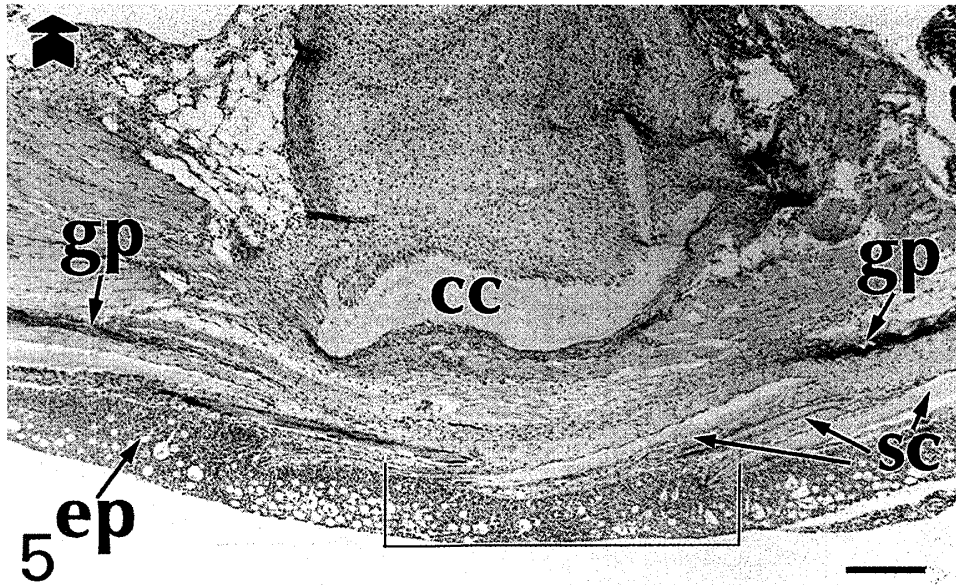


Fig. 5. Histological section of the spot area and the related areas, showing coracoid cartilage (cc), guanophore layer (gp), scales (sc) and epidermis (ep). Note that the guanophore layer is absent under the cartilage. The number of mucous cells in the epidermis in Fig. 4 was counted inside of the bars located in the epidermis. Bar indicates 0.3 mm. The arrow is pointed in the dorsal direction.

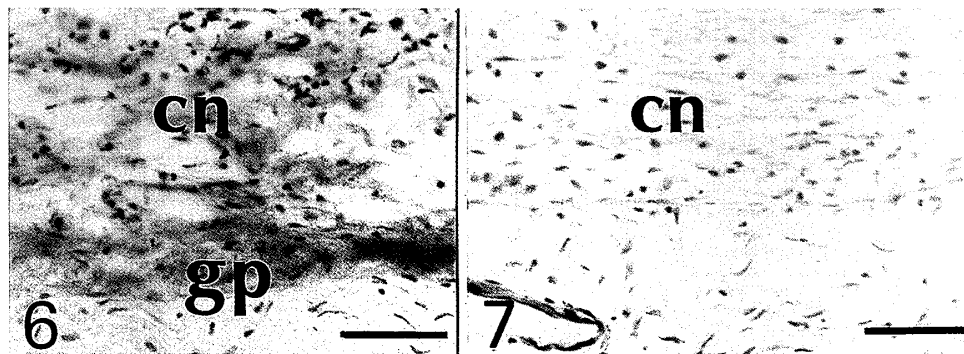


Fig. 6. Histological section of the connective tissue at the surrounding areas. Note the guanophore layer (gp) and that the connective tissue (cn) is composed of rough fibers. Bar indicates 75  $\mu$ m.  
 Fig. 7. Histological section of the connective tissue at the spot. Note the absence of guanophore layer and that the connective tissue (cn) is composed of compact fibers. Bar indicates 75  $\mu$ m.

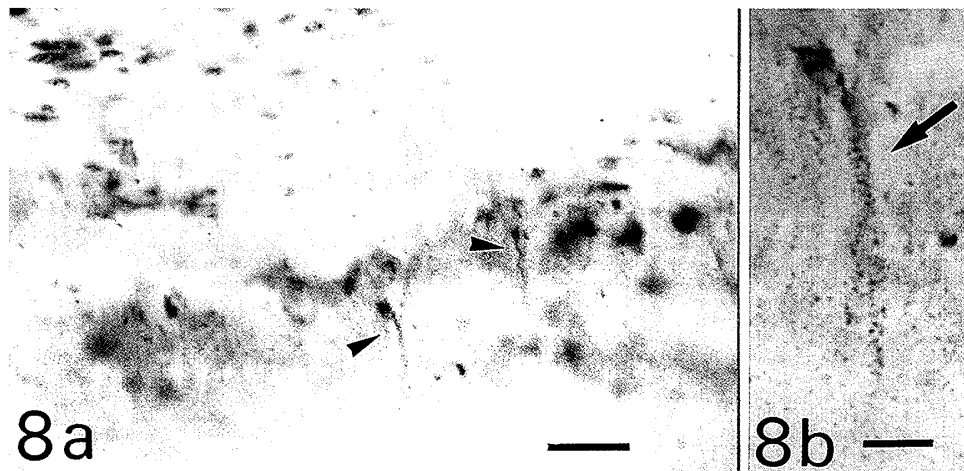


Fig. 8a. Neurons (arrowheads) present in the connective tissue layer above the spot. Bodian's silver staining method modified by Otsuka (1962). Bar indicates 30  $\mu$ m. 8b. A magnified neuron. Bar indicates 10  $\mu$ m.

well known that mechanosensitive neurons are distributed in the skin of mammals (Hulliger, 1979; Kleinbok, 1983). In this context, however, the lack of a guanophore layer at the spot cannot be explained. We have observed similar spots on the chests of other fishes such as horse mackerel. Furthermore, a spot is sometimes recognized in the abdominal fin in goldfish. In this case also, there was a cartilage process under the spot. Recently, it has been reported that cartilage secretes some factors such as inhibitors of vascularization (Homandberg, 1992; Moses, 1993). There is a possibility that in goldfish, the cartilages secrete an unknown factor which inhibits the development of guanophores, although the reason is not clear.

## References

- Kleinbok, Ila (1983) *Fiziol. Zh. SSSR*, **69**: 140–145.
- Himick, B.A. and R.E. Peter (1994) *Am. J. Physiol.*, **267**: R841–R851.
- Homandberg, G.A. (1992) *Connect. Tissue Res.*, **28**: 289–305.
- Hulliger, M. (1979) *J. Physiol.*, **291**: 233–249.
- Moses, M.A. (1993) *Clin. Exp. Rheumatol.*, **11**: S67–S69.
- Otsuka, N. (1962) *Z. Zellforsch.*, **58**: 33–50.
- Parry, D.M. (1993) *J. Neuroendocrinol.*, **5**: 387–395.
- Ueda, H., Kambegawa, A. and Y. Nagahama (1985) *Gen. Comp. Endocrinol.*, **59**: 24–30.