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Mechanism of adhesion in a hillstream fish, *Glyptothorax garhwali* Tilak, as revealed by scanning electron microscopy of adhesive apparatus

The Western Himalayan hillstreams pose harsh living conditions to the fish communities because of varied topography, torrential water currents coupled with a variety of substratum. The most important characteristics in response to these conditions are the integumentary modifications in the form of an adhesive disc, which has become a life-saving kit for most of the hillstream fishes. Though investigations have been carried out using light microscope[1,2], the exact mechanism of adhesion and detailed structure of the adhesive apparatus are still not well-known. Hence, an attempt has been made to study the details of the adhesive mechanism of a typical hillstream fish, *Glyptothorax garhwali* Tilak using scanning electron microscopy (SEM).

G. garhwali is an extremely specialized fish inhabiting the fast-flowing upper reaches of Western Himalayas. It has a well-developed adhesive apparatus, unlike *Schizothorax richardsonii* in which the lower lip is modified to form a suctorial disc. In the present case, the lateral folds of the skin just above the adhesive disc and a portion of the adhesive disc were subjected to SEM investigation. The adhesive apparatus was removed with the help of a sharp blade and fixed in 2.5% glutaraldehyde in 0.1 M sodium cacodylate buffer at pH 7.2–7.4 for 24 h. After several washings in the rinsing buffer, 0.1 M sodium cacodylate buffer containing 7% sucrose was added and further dehydration was carried out in various grades of acetone. The specimens after acetone treatment were transferred into emylacetate solution, dried in a Polaram Critical Point Dryer (CPP), mounted on metal stubs and then coated with 100 Å thick layer of gold in JEOL sputter ion coater. The specimens were examined with JEOL TSM 6100 SEM at 20 kV and the images were observed on the screen. Negatives were prepared for photography.

The studies indicated the presence of numerous mucous pores (MP) over the lateral folds and the adhesive disc. The primary function of these is the secretion of mucus (Figure 1 *a*). Tiny hook-shaped projections arising out of the epithelial cells (Figure 1 *a*) are present alongside. The adhesive disc has numerous long, hook-shaped, spine-like structures (LHSs) (Figure 1 *c* and *d*), which are the epidermal growths (EG) (Figure 1 *b*). These epidermal growths are present all over the central pit, which aids in the process of adhesion. For the purpose of attachment, the LHSs get entangled with the rough surface of the substratum, forming a sort of interlocking mechanism. Along with the mucus openings, these hook-like, spiny structures present the most advanced case of morphological adaptation amongst the hillstream fishes. What seems to be the case here is that mucus is secreted on receiving the necessary stimuli from the surrounding environment, providing a sort of platform in the form of a feeble adhesion for the secondary adhesion of spines with the rough surface of the substratum. There is

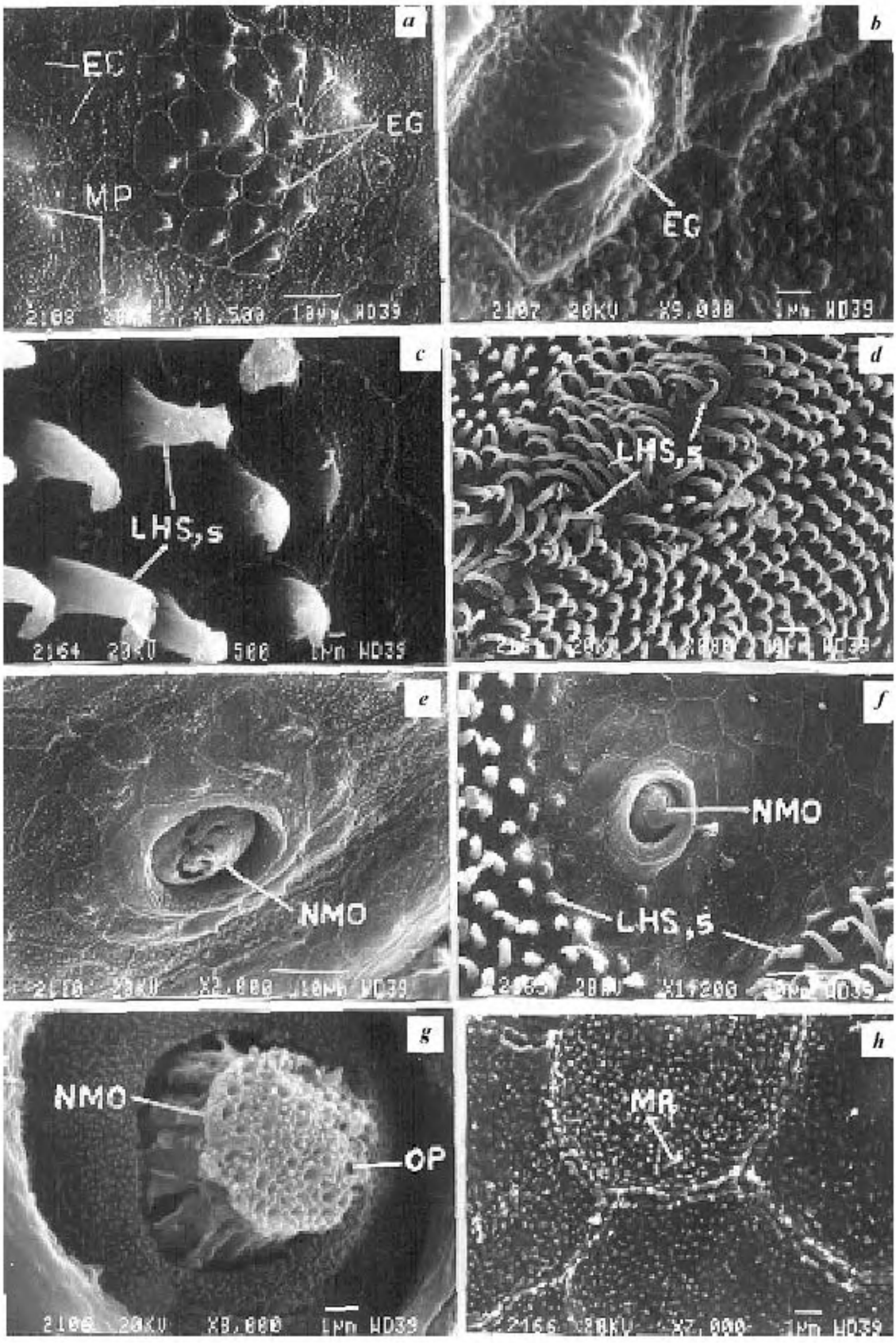

Figure 1. Scanning electron micrographs of the adhesive apparatus of *Glyptothorax garhwali* Tilak. EC, Epithelial cells; EG, Epidermal growths; MP, Mucous pores; LHS, Long hooked spines; NMO, Neuromuscular organs; MR, Microridges.

a possibility that the mucus and spines may be complementing each other simultaneously. It is opined that certain neuromuscular organs (NMO) arising out of large openings (Figure 1 *e–g*) aid in sensing the stimuli. When observed at higher magnification the epithelial cells showed numerous microridges (MR) (Figure 1 *h*) which increase the surface area, besides providing protection and increasing the mobility of the mucus. Similar structures were observed on the lateral folds also.

The earlier light microscopic studies have indicated that the adhesive apparatus of *G. telchitta*, *Pseudechenesis sulcatus* and *G. pectinopterus* works mainly by vacuum created by the adhesive apparatus, which is responsible for the adhesion to the substratum[1,3,4]. The SEM investigations carried out on a few hillstream fishes like *Garra gotyla gotyla*[4,5] and *Glyptothorax dakpathari*[6] have changed this view altogether and the changed concept is as follows.

The mucus secreted by the numerous mucous pores present on the adhesive disc provides protection from mechanical abrasions, besides its immunological functions[7,8]. It also aids in the process of adhesion. The LHSs (Figure 1 *d*) form a sort of 'hand-in-glove' mechanism with the rough surface of the substratum following primary adhesion. It is thus hypothesized that if the surface of stones lying in the stream is free from depositions other than food materials and have holes corresponding to the sizes of the denticles, tubercles and protrusion, it will provide an excellent surface for clinging to the fishes. If the holes of the stones are filled or covered with sediments, the fishes will not be able to cling to the stones; hence they move to some other areas in search of a suitable substratum. So the degradation of substrate is responsible for the non-availability of substratum and food to the hillstream fishes, effecting growth and reproduction.

Other investigators observed similar structures in case of *G. g. gotyla* where numerous stub-shaped tubercles (ST) are present, which in turn have numerous spines[4]. These not only help the fish to adhere to the substratum, but also act as mechano-cum-sensory receptors[9]. In the case of *G. g. gotyla* it was found that the cumulative action of the spines and mucus helps the fish to adhere to the substratum[5]. Friel observed these spiny structures on the ventral surface of catfish *Acanthobunocephalus nicoi*[10]. In the present case, the microridges present on *G. garhwali* increase the area of absorption, provide protection and increase the mobility of the mucus[8].

Hence *G. garhwali* has well-developed, specialized apparatus with specialized structures such as the long, hook-shaped spiny structures which form an interlocking mechanism with the rough surface of the substratum, to represent the most advanced case of morphological adaptation in the hillstream fishes. However, we do not preclude the role played by the mucus in strengthening the adhesion to the substratum.

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