Comparative Surface Architecture Of The Adhesive Apparatus Epidermis Of *Garra Gotyla* (Hamilton) (Hill-Stream Fish) Of Kumaun Himalaya.

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Abstract: The epithelial cells have been demonstrated in the epidermis of adhesive apparatus of *Garra gotyla* (Hamilton), by scanning electron microscope methods, to study its structural and functional organization with special reference to adhesion. The epidermis of mental adhesive apparatus of *G. gotyla* is smooth and uniform in thickness at flat circular part and is papilliated at posterolateal part. The epidermis of flat circular part of *G. gotyla* equipped with epithelial cells and mucous cells. The epidermis of posterolateral part in *G. gotyla* consists of mainly keratinized cells. The secretion of amount of mucus by the epidermis covering the groove regions separating the flat circular part anteriorly from posterior lip and posterolaterally from its posterolateral part is significant. This might help in maintaining the vacuum creating by the muscles under margins and thus preventing the entrance of water and air. *G. gotyla* are fresh water teleosts these fish inhabit water resources in hills which are shallow, well aerated and clear and contain plenty of food in the form of algal slime covering stones and rocks. The strength of water current in these water resources are much higher compared to those of the rivers of plains. These fishes usually cling to the rocky substratum with the help of their modified skin. The epidermis of mental adhesive apparatus is smooth and uniform in thickness at flat circular part and is papilliated at posterolateal part.
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Key Word: Adhesive apparatus, Epidermal cell, *Garra gotyla*, and SEM.

Introduction:

The hill-stream fishes have developed specialized organs for adhesion in several strategic regions of the body. Adhesive apparatus are the modifications of the skin on ventral surface. Epidermis or top most covering of body is the outermost defence organ against the surrounding aquatic environment comes into direct contact with the mechanical hazard. Fish skin is most extensive sense organ of the body for reception of tactile, thermal and painful stimuli between the external and intermal environment of the fish. The most important, role among its numerous functions is to protect the fish from xenobiotics and other environmental hazards. *G. gotyla* (Hamilton) is fresh water teleosts, belonging to the families' cyprinidae. This fishes inhabit water resources in hills which are shallow, well aerated and clear and contain plenty of food in the form of algal slime covering stones and rocks. The strength of water current and the intensity of light in these water resources are much higher compared to those of the rivers of plains.

The present investigation has been designed to study the comparative structural organization and functional significance of the epidermis of adhesive apparatus of *G. gotyla* (Hamilton) fishes in relation to their life in hill-streams. Studies of fish skin indicated that epidermis cells following separate pathways of differentiation in different fishes. In most of the fishes the epidermises is related more to the deposition of slime over its surface and undergo the process of mucogenesis and in some the epidermis cells undergo the process of keratinization forming a layer at the surface (Mittal & Benerjee 1979). Some aspect of ultrastructures of the adhesive apparatus of several fishes have been studied before (Das & Nag, 2005, 2006; Saxena & Chandy, 1966; Hora, 1922; Bhatia, 1950; Saxena, 1963; Lal et al., 1966; Bose et al, 1971 and Singh & Agarwal NK., 1990, 1991).

Materials And Methods:

Live adult specimens of *G. gotyla* (7-9 cm long) were collected from Kosi River at Kakrighat, Distt Nainital, (Uttarakhand, India) respectively water current was very fast having velocity 0.5 to 2.0 m/sec. (Bhatt & Pathak, 1991) and the bed was rocky Specimens were maintained in laboratory at 25 ± 2^0C. The fish were cold anesthetized, following Mittal & Whitear (1978), for SEM preparation skin fragments of about 10×10 mm were cut from dorsal side of just behind the Lip. Tissue were excised and rinsed in 70% ethanol and one change saline solution to remove debris and fixed on 3% Glutaraldehyde in 0.1m phosphate buffer, at pH 7.4 for one night at 4^0c at

Refrigerator. The tissue were washed in 2-3 changes in phosphate buffer and dehydrated in the graded series of ice cold Acetone (30%, 50%, 70%, 90%, and 100% approximate 20-30 min.) and critical point dried, using Critical Point Dryer (BIO-RAD England) with liquid carbon dioxide as the transitional fluid. Tissues were glued to stubs, using Conductive Silver Preparation (Eltecks, Corporation, India) Coated with gold using a sputter Coater (AGAR, B 1340, England) and examined in a Scanning Electron Microscope (Leo, 435, VP, England). The results were recorded using Kodak T-MAX 100 professional film (Kodak Ltd., England).

Observation: The adhesive apparatus is situated ventrally in *G. gotyla* (Fig. 1).

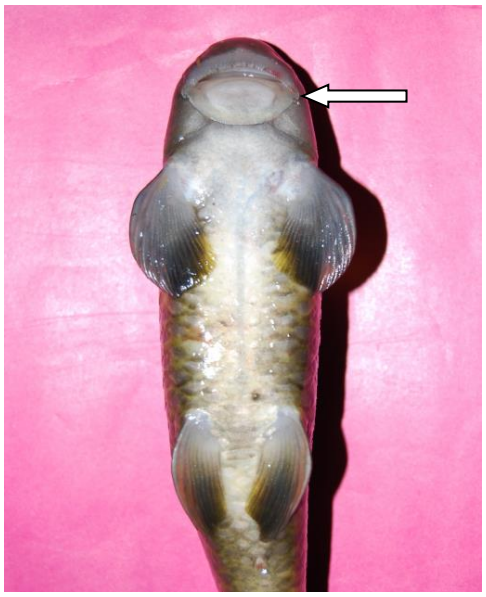

Fig. 1: Ventreal View Of *Garra Gotyla.*

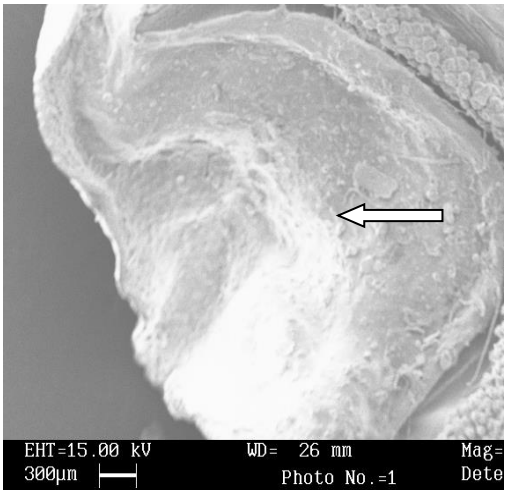

Fig. 2: Surface electron microphotograph (SEMPH) of the adhesive apparatus epidermis of *G. gotyla*

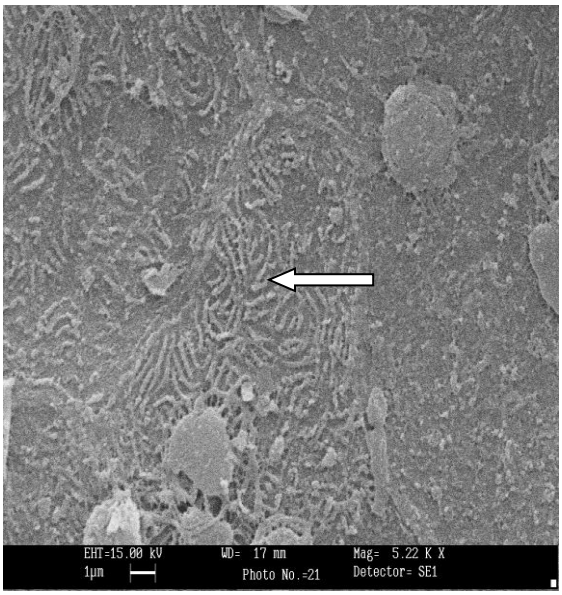

Fig. 3: SEMPH of adhesive apparatus epidermis of *G. gotyla* showing the microridges (marked by arrows) at the surface of epithelia of flat circular major anterior region. (Scale bar- 1 µm).

In *G. gotyla* is commonly known as 'sucker head'. Well developed adhesive apparatus is present just behind the posterior lip and is in the form of a button like disc, known as mental adhesive disc (approximate length 9.2 mm and width 12.1mm) (Shah, K., 1989) (Fig 2). The mental adhesive disc of *G. gotyla* consists of a flat circular major anterior part (approximate $^{2}/_{3}^{rd}$ region of the disc) and a free posterolateral part (approximate $^{1}/_{3}^{rd}$ region of the disc) projecting from the surface, the anterior part of the remains separated from the posterior lip by a deep groove (approximate depth 1.7 mm.). A shallow groove (approximate depth 1 mm.) is also present in between the flat circular part and posterolateral part of the disc (Fig. 2).

The epidermis of adhesive apparatus is mainly composed of epithelial cells, and mucous cells. The cell types and their structure and metabolic state in this region is almost resemble those present in the epidermis of lips of this fish. the microridges is compactly arranged, branched with abrupt ends of irregularly interwoven to form intricate mesh like pattern, and interconnected with microbridges (Fig. 3).

Interspersed between the epithelial cells, mucous cells are distinguished. The mucous cell opens with well developed mucous cell opening (Fig. 4).

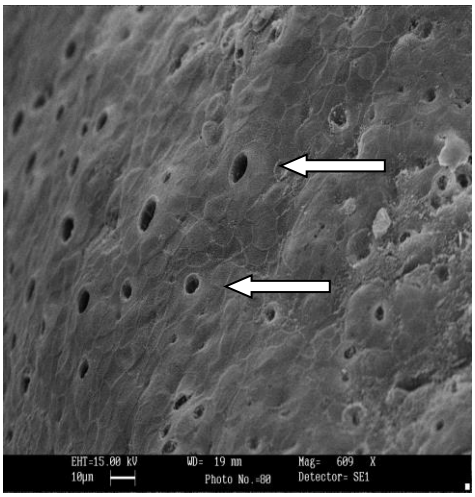

Fig 4: SEMPH of flat circular part of *G. gotyla* showing the openings of mucous cells (marked by arrows (Scale bar – 10 µm).

Some developed taste buds are described in the adhesive apparatus (flat circular part) of *G. gotyla* (Fig. 5).

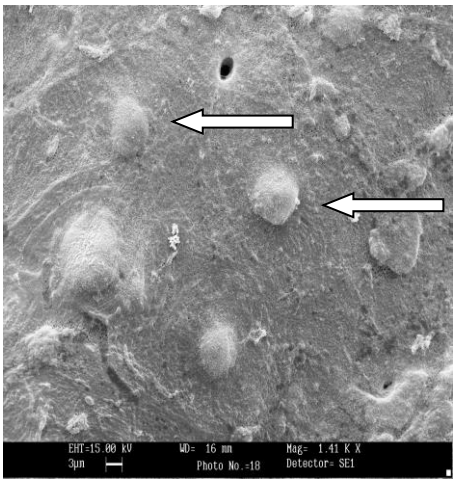

Fig 5: SEMPH of adhesive apparatus epidermis of *G. gotyla* showing well developed taste buds (marked by arrow) (Scale bar – 3µm).

Discussion: The epidermis of the adhesive apparatus, and the structures associated with them show considerable structural modifications. These may be considered as adaptations in relation to its peculiar adhesive procuring device, feeding habit, feeding zone and the nature of its rivers. Practically all the three fishes are bottom dweller inhabiting torrential hill-stream with an uneven bed composed of stones, rocks, boulders, gravel and sand, The presence of well developed mucus cells in the epidermis of flat circular part forming a slime layer on the surface of disc in *G. gotyla* it significant as chest region of the fish is liable to friction especially when it swims upstream. This may provide sufficient lubrication to reduce friction between body surface and water currents, thus protecting the epidermis from wear and tear during locomotion.

The epidermis of groove, separating the flat circular part from the posterior lip anteriorly and posterolateral part postrolateally in *G. gotyla*, is subjected to more frictional stress than that of the outer surface due to the movement of flat circular part during the operation of suctorial function. The presence of comparatively numerous mucous cells in the epidermis of these regions, secreting profuse amount of mucus is thus significant in *G. gotyla*. This may provide sufficient lubrication to reduce friction at hinge thus protecting the epidermis from wear and tear during adhesion. Secretary activities of epithelial cells and mucous cells in the nonkeratinized epidermal furrows may provide protection to the fish against various harmful factors in the environment, (Mittal et. al., 1994 a). In addition, mucus secretion has been associated with diverse activities such as recognition of cells, reception of chemical informations and the maintenance of a suitable environment (Mittal et. al., 1995). Hora (1922), Saxena (1959), Ojha & Singh (1922) and Singh et al., (1994) think that the central portion of the adhesive pad constitute an effective sucker when the fish rest on the substratum however, the mucogenic epithelia covering the rostral cap and the central region of the adhesive pad are likely to be subjected to wear and tear during frequently adhesion and friction of the fish with the substratum as they are on the ventral side of the body. Thus heavy secretions elaborated by the epithelial cells and the mucous cells could be regarded as an adaptation to lubricate and protect the epithelial cell from abrasion.

In *G. gotyla*, the sudden spread of mucus of flat circular part (FCP) is facilitated by numerous canaliculi formed by epidermal microridges (MR). Hence cumulative action of tubercles and mucus enables the fish to make firm hold on the substratum. Das et. al., (2006, 2008) suggested the bases of the available morphological evidence, it is possible to suggest the mechanism of attachment in *G. gotyla*. It is likely that mechanical interlocking and suction and perhaps viscous forces are involved in this mechanism already mectioned, the epidermal tubercles are equipped with spines, whereas the grooves between them and the central part are devoid of spines, when the spines are pressed against the substratum a vacuum is likely created by the contraction of the dermal muscles attached to the groove regions of the epidermis. The spines assist in effective anchorage as well as interlocking with the organic growth of the submerged rocks. The mucus secretion from the mucous cell protects the spines from abrasion during attachment and interlocking. The spines get dislodged

when the callus part of the adhesive apparatus is pressed to words the substratum. The keratinization of the epidermis spines and the outer row cells is perhaps involves in protecting the adhesive apparatus surface against mechanical damage attachment. Ojha & Singh (1922) suggested a possible involvement of the adhesive apparatus in food scraping in a related cyprinid species *G. lamta lamta.*

Hill-stream fish is inhabitant of the fast flowing cold water streams. *G. gotyla*, do bottom dwellers inhabit torrential hill-streams with anuneven bed composed of stones, rocks, boulders, gravel or sand. The spine border on the tubercles are very well marked in the upper liable folds of respective lips like, the epithelial cells at the base of the tubercles are very clear. The disc comes in contact with the substratum, first which not only anchor to the substratum but also act as mechano-sensory organs. This process is following by the secreting of mucus of flat circular part of *G. gotyla* is significant as chest region of the fish is liable friction especially when it swims upstream.This may provide reduce friction between body surface and water currents that protecting the epidermis from wear and tear during locomotion. In *G. telchelta* (Bhatia, 1950; Lal et. al, 1966; Singh et. al., 1990; Singh et. al., 1991; Das et. al., 2004 and 2006) adhesive apparatus comprise the longitudinal to long body axes. In other species *Schizotharax plogiostomus*, *crossocheilus latius latius* (Sing & Agarwal, 1990) adhesive apparatus consists of transverse band behind scrapping plate and provided with numerous tuberculated projections. While in *Bhavania annandalai* (Hora, 1922), *Garra annadalei* (Hora, 1923), *Garra mullya* (Saxena, 1962) and *Garra gotyla* (Singh & Agarwal, 1990) it is complex with marked anterior labial fold, posterior labial fold , callous portion of disc and posterior free margin of disc. The present study also reported the occurrence secretary activities of epithelial cells and mucous cells in the non-keratinized epidermal may provide protection to the fish from various harmful factors in the environment (Mittlal et. al., 1994). In addition, mucus secretion has been associated with diverse activities such as recognition of cells, reception of chemical information and the maintenance of a suitable environment (Mittal et. al., 1995). The interspaces between the tubercles provide continuous flow of water for aeration. The microridges (MR) provide structural integrity to epithelium of flat circular part and increase the surface area and also prevent mechanical abrasions (Aslon, 1995).

The copious amount of mucus secretion by the epidermis covering the grooves in all the *G. gotyla* is significant. This might help in maintaining the vaccum created by the muscles forming a film of mucus along its margins and thus preventing the enterance of water and air.

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