

Histochemical Localization of Mucopolysaccharides in Skin of Freshwater Fishes: A Comparative Study

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Abstract:

Fish epidermal mucus provides the first line of defense against pathogens. The present work reports on the comparative study of variation in Structure of skin and defense markers among some of the fresh water fishes mrigal (*Cirrhinus mrigala*), grass carp (*Ctenopharyngodon idella*), bighead carp (*Aristichthys nobilis*) snake head (*Channa punctatus*), walking catfish (*Clarius batrachus*) and tilapia (*Oreochromis niloticus*) with respect to their role in innate immune system. The defense markers analyzed include Presence of Different types of mucopolysaccharides in skin. From the histochemical localization of the skin of different species acidic mucopolysaccharides (GAG) and neutral mucopolysaccharides have been localized, which play important role in the defense mechanisms

1. INTRODUCTION

Body surfaces of multicellular organisms are defended by epithelia, which provide a physical barrier between the internal milieu and the external world. Skin is the structure that covers the body and protects it not only from the entry of pathogens or allergens, but also from the leakage of water, solutes, or nutrients. These outside-in and inside-out barrier functions are dependent on the epidermis. While mucus covers the epidermis. Skin is unique and histologically diverse. It is very different from that of mammals, because it secretes mucus which is involved in immune functions. Glycosaminoglycans: (GAGs) are linear acidic polysaccharides found on cell surfaces and in the surrounding extracellular matrix. GAGs participate in and regulate many cellular events in physiological and pathophysiological processes, such as cell proliferation and differentiation, cell-cell and cell-matrix interactions, and viral infection, through their interaction with different proteins. GAGs are divided into four main categories—hyaluronic acid (HA), chondroitin sulfate/dermatan sulfate (CS/DS), heparosan/heparan sulfate/heparin (HN/HS/HP), and keratan sulfate—based on monosaccharide composition and the configuration and position of the glycosidic bonds between their monosaccharides (Zhang et al., 2008). GAG have a large ion- and waterbinding capacity, and play an important role in the maintenance of the viscosity of extracellular ground substances (Muir, 1980). the functional significance of GAG is protection against pathogenic organisms (Marshall, 1978; Hughes, 1980).

2. MATERIAL AND METHODS

Fish collection: *Hypophthalmichthys nobilis*, *cirrhina mrigala*, *ctenopharyngodon idella* and *tilapia* were obtained from juhu-krupa farm, pij village, taluka nadiad (dist. Kheda). From there we are transport the fish in polythene bag with aerated water and immediately collected the mucus.

Histochemical Localization of the Mucopolysaccharides by Alcine blue PAS Technique: Dissect out the fish skin and immediately transfer into neutral buffered formaline for 24 hours. The tissue were dehydrated with alcohol series and blocks were prepared in 60-6C paraffin wax. the section were cut at 10micro meter

and dewax the ribbon and stained with alcine blue, periodic acid and schiff's reagent and Photographs were taken at 40X.

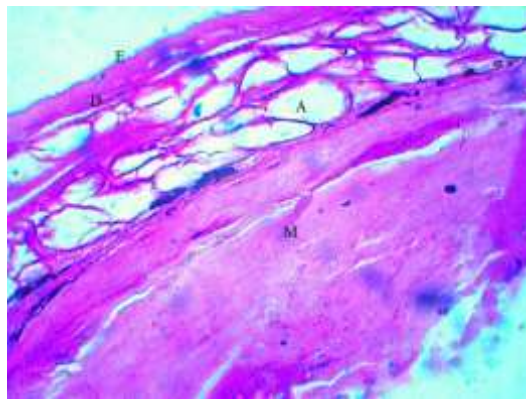
Histology analysis : Dissect out the fish skin and immediately transfer into Bouin's Fixative for 24 hours. The tissue were dehydrated with alcohol series and blocks were prepared in 60-6C paraffin wax.the section were cut at 10micro meter and dewax the ribbon and stained with Hematoxylene-Eosine and Photographs were taken at 40X.

3. RESULTS AND DISCUSSION

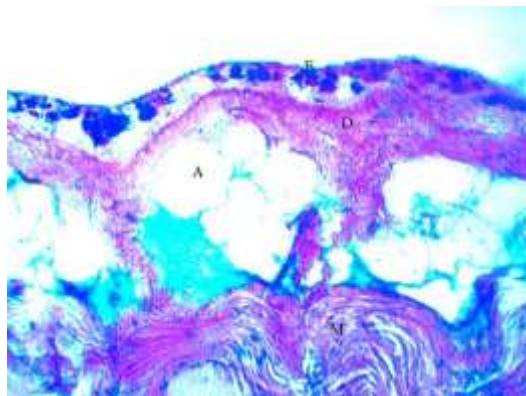
All section shows presence of GAGs are acidic polysaccharides found on cell surfaces and in the surrounding extracellular matrix.GAGs participate in and regulate many cellular events in physiological and pathophysiological processes, such as cell proliferation and differentiation, cell– cell and cell–matrix interactions, and viral infection, through their interaction with different proteins. GAG have a large ion- and waterbinding capacity, and play an important role in the maintenance of the viscosity of extracellular ground substances (Muir, 1980).the functional significance of GAG is protection against pathogenic organisms (Marshall, 1978; Hughes, 1980).

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1. *Channa Punctatus* (40X)



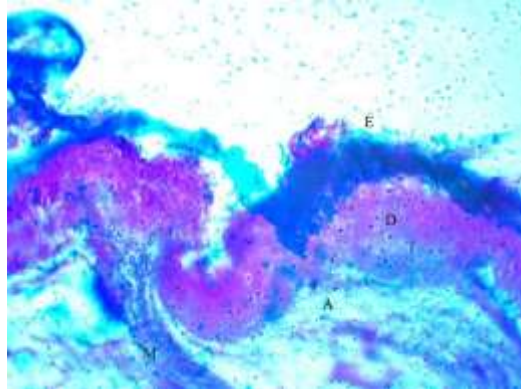
2. *Clarius batrachus* (40X)



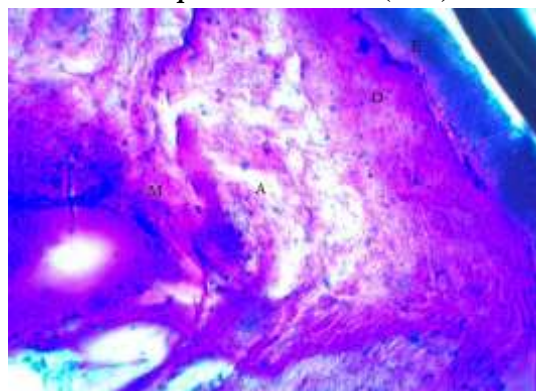
1. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in dermal region especially near adipose tissue. Neutral mucopolysaccharides also clearly seen in the section.
2. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in dermal region especially near adipose tissue and also in some portion of the muscular layer and neutral mucopolysaccharides also clearly seen in the section.
3. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in Epidermal and dermal region especially near adipose tissue and also in some portion of the muscular layer and neutral mucopolysaccharides also clearly seen in the section

4. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in Epidermal and also in some portion of the muscular layer and neutral mucopolysaccharides also clearly seen in the section.

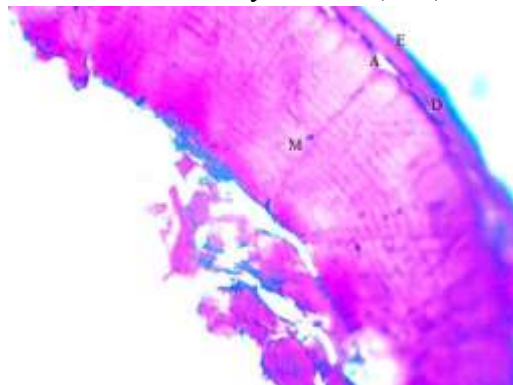
3. *Cirrhina mrigala* (40X)



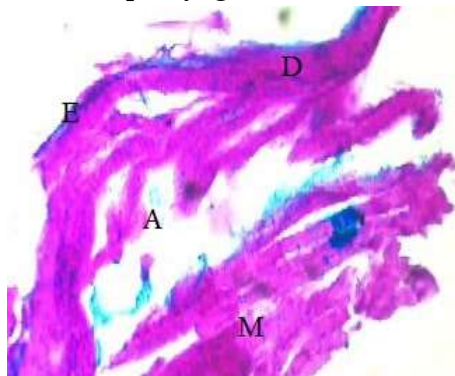
4. *Tilapia mossambica* (40X)



5. *Aristichthys nobilis* (40X)



6. *Ctenopharyngodon idella* (40X)



5. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in Epidermal and dermal region especially near adipose tissue and also in some portion of the muscular layer and neutral mucopolysaccharides also clearly seen in the section.

6. Photographs indicate clear presence of acidic mucopolysaccharides (blue or magenta color) in Epidermal and also in some portion of the muscular layer and neutral mucopolysaccharides also clearly seen in the section.

4. CONCLUSION

From the histochemical localization of the skin of different species acidic mucopolysaccharides (GAG) and neutral mucopolysaccharides have been localized, which play important role in the defense mechanisms. Thus, present work suggest that the primary defense system in terms of chemical and physical properties of mucus is robust in *channa punctatus* quite weaker in *Ctenopharyngodon idella*.

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REFERENCES

1. Esteban, M. Á., & Cerezuela, R. (2015). *Fish mucosal immunity: skin. Mucosal Health in Aquaculture*. Elsevier Inc.
2. Gobinath, C., & Ravichandran, S. (2011). Antimicrobial Peptide from the Epidermal Mucus of Some Estuarine Cat Fishes Faculty of Marine Sciences, CAS in Marine Biology, 12(3), 256–260.
3. Gomez, D., Sunyer, J. O., & Salinas, I. (2013). Fish & Shell fish Immunology The mucosal immune system of fish :The evolution of tolerating commensals while fighting pathogens.
4. Guardiola, F. A., Haro, J. P. De, Díaz-baños, F. G., Cuesta, A., & Esteban, M. Á. (2015). Terminal carbohydrate composition, IgM level and enzymatic and bacteriostatic activity of European sea bass (*Dicentrarchus labrax*) skin epidermis extracts. *Fish and Shellfish Immunology*.
5. Haniffa, M. A., Viswanathan, S., Jancy, D., Poomari, K., & Manikandan, S. (2014). Antibacterial studies of fish mucus from two marketed air-breathing fishes – *Channa striatus* and *Heteropneustes fossilis*, 5(2), 22–27.
6. Nagashima, Y., Tsukamoto, C., Kitani, Y., Ishizaki, S., Nagai, H., & Yanagimoto, T. (2009). Comparative Biochemistry and Physiology , Part B Isolation and cDNA cloning of an antibacterial L -amino acid oxidase from the skin mucus of the great sculpin *Myoxocephalus 53 polyacanthocephalus*. *Comparative Biochemistry and Physiology, Part B*, 154(1), 55–61.
7. Subramanian, S., Mackinnon, S. L., & Ross, N. W. (2007). A comparative study on innate immune parameters in the epidermal mucus of various fish species, 148, 256–263.
8. Valdenegro-vega, V. A., Crosbie, P., Bridle, A., Leef, M., Wilson, R., & Nowak, B. F. (2014). Fish & Shell fish Immunology Differentially expressed proteins in gill and skin mucus of Atlantic salmon (*Salmo salar*) affected by amoebic gill disease. *Fish and Shellfish Immunology*, 40(1), 69–77.
9. Wei, O. N. G. Y., Xavier, R., & Marimuthu, K. (2010). Screening of antibacterial activity of mucus extract of Snakehead fish , *Channa striatus* (Bloch), 675–681.
10. Yoshinaga, T., Nagakura, T., & Ogawa, K. (2002). Attachment-inducing capacities of fish skin epithelial extracts on oncomiracidia of *Benedenia seriola* (Monogenea : Capsalidae), 32, 381–384.
11. Zealand, N. (1976). Mucus Globule Membrane : An Hypothesis Concerning its Role in