Asian Journal of Aquatic Sciences, April 2025. Vol 8, Issue (1) 1-6
e-issn: 2716-4608
p-issn: 2655-366X

HISTOPATHOLOGICAL OF STRIPED CATFISH (*Pangasianodon hypophthalmus*) REARED IN DIFFERENT SALINITIES

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ABSTRACT

This study aimed to discover the effects of different salinities on histopathological *Pangasianodon hypophthalmus*. The method used is experimental by applying a completely randomized design (CRD) with four treatments: salinity 0 ppt, salinity 5 ppt, salinity 7 ppt, and salinity 9 ppt. The fish specimen is 8-10 cm in length and weighs 5 g. It was raised in a 54 L tank containing 40 L of water at a density of 2 fish 1 L^{-1} and kept for 45 days. Feed with commercial pellet 3 times a day in satiation. The results showed histopathological changes in gills and kidneys occurred at a salinity of 9 ppt. Chloride cell hypertrophy and secondary lamella bend in the gill organs, while the kidneys have acute tubular necrosis. Meanwhile, a salinity of 7 ppt resulted in abnormalities in the kidney organs with hemorrhage. It can be concluded that a salinity of 5 ppt indicates a safe salinity condition for the maintenance of striped catfish.

Keywords: *Pangasianodon hypophthalmus,* Histopathological, Salinity, Gill, Kidney

1. INTRODUCTION

Tiger shrimp producers leave around 6.500 Ha of ponds empty due to their inability to complete cultivation tasks[1]. One is frequent disease outbreaks that fail in the cultivation business, so many farmers leave their land. To reuse cultivation land, a potential commodity alternative can be raised in pond waters with a low salinity of 10 ppt[2-4]. One of the potential commodities is striped catfish (*Pangasianodon hypophthalmus*) because of the nature of conjoined catfish that can grow and develop well against all environmental conditions[5].

Salinity is one factor that affects the physiology of aquatic organisms, involving the work pressure of osmosis. Osmotic working pressure that is too high will cause differences in the body and the environment in the concentration of fluids. Balancing the concentration of the fluid requires large amounts of energy, which will ultimately affect the growth of the fish. Feed energy that should be used for growth is also used to maintain fluctuating osmotic pressure. Fish that cannot control the osmoregulation process that occurs in the body will experience stress, health will be disrupted, and even death will occur[6].

Gill organs, kidneys, and skin are very instrumental in osmoregulation activities. When fish are sick, injured, or stressed, the infiltration process is disrupted; the water is very much into the fish's body, and more

Received: 5 February 2025
Accepted: 29 March 2025

ions will be removed. This causes the kidney organs to pump water out of the body to increase. If this continues, kidney damage and death can occur in fish[7]. The changes in the structure of the osmoregulation organs damaged by differences in salinity can be identified through histopathology. This study was conducted to find out the differences in the histology conditions of gills and kidneys in conjoined striped catfish that are maintained in different salinity so that it is expected to provide information and efforts made if you want to cultivate conjoined catfish in certain salinity conditions.

2. RESEARCH METHOD

Time and Place

This research was conducted in February 2024 at the Marine Microbiology Laboratory, Faculty of Fisheries and Marine, Universitas Riau.

Method

The method used is a Complete Random Design experimental method (CDR) with 4 salinity treatments, including 0 ppt, 5 ppt, 7 ppt, and 9 ppt with three repetitions. The treatment used refers to the research of Fitrani et al.[8], which found the best salinity to be 7 ppt. The fish specimen used is 8-10 cm in length and weighs 5 g. It was raised in a 54 L tank containing 40 L of water at a density of 2 fish 1 L^{-1} and kept for 45 days. Feed with commercial pellet 3 times a day in satiation.

Procedures

The gill and kidney histology sample is taken at 45 days of maintenance. Gill samples are taken by closing the gill (operculum) and cutting the base to release the gills. After that, the kidneys are obtained by opening the abdominal cavity. The 10% formalin of Buffer Phosphate (NBF) is used for organ fixation for 24-48 hours[9]. After fixation, the tissues were dehydrated in a graded series of alcohol and xylol and then embedded in paraffin. All these stages were conducted using a tissue processor. The sections obtained were cut at 3-5 µm thickness, incubated in a water bath at 40°C, and then immediately air-dried for one hour. Afterwards, it was stained using hematoxylin-eosin (HE) and observed under a light microscope at a magnification of 400x.

3. RESULT AND DISCUSSION

Histopathology of Striped Catfish Gills

Gills are the main respiratory organs that work with dissolved oxygen diffusion in water. Gills include gill arches, teeth, and sieve/gill comb[10]. Gills are very vulnerable to decreased water quality because the gills are directly in contact with water. Therefore, whatever changes occur in the aquatic environment, directly and indirectly, affect the structure and function of the gills. The structure of the concentration of the conjoined catfish maintained in different salinity can be seen in Figure 1.

Based on Figure 1. it can be seen that the structure of the conjoined striped catfish in salinity 0 ppt shows the condition of the normal fish gill structure marked by having secondary lamella, which are neatly arranged and regular, mucus cells are still clearly visible, epithelium, lacuna neatly arranged. This follows Dawood et al.[11], a healthy fish gill structure composed of epithelium, mucus cells, lacuna, supporting cells, and chloride cells. The salinity of 5 ppt and 7 ppt showed no change in abnormalities that occur, seemingly secondary lamellas that are arranged neatly and regularly, and mucus cells are still clearly visible, epithelium and lacuna neatly arranged.

In contrast, salinity 9 ppt appears to have changed the presence of hypertrophy of chloride cells. This happens because fish try to remove too many excess ions, so chloride cells require significant power to remove excess ions. Due to the large amount of energy released by chloride cells, chloride cells will experience changes in size to swell or enlarge and increase. Chloride cells function to control the excess ions that enter the body through the gills. The mechanism of chloride cells controlling excess ions is to

carry out active transport to regulate excess Na^+ and Cl^- ions through gills, pump excess ammonia out of the environment, and remove the remaining ions through the kidneys. The primary source of energy for active transport is provided by mitochondria associated with Na^+K^+-ATP, which is located along the basolateral and in the microtubular system chloride cells that extensively and actively do Na+ transport out cells to exchange with K+ into cells[12].

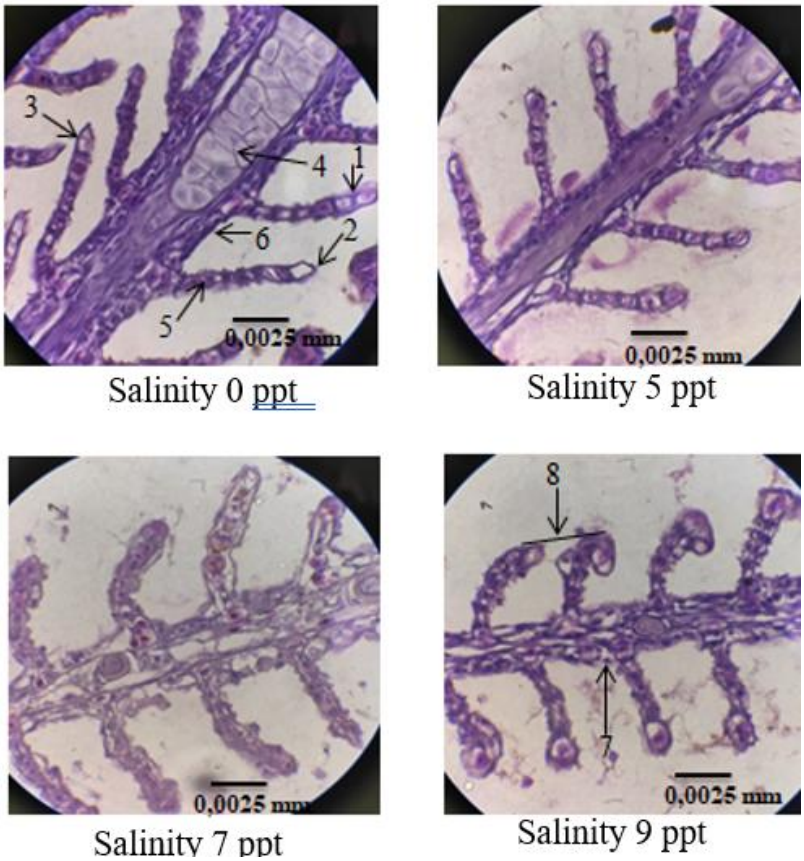

Figure 1. Structure of gill tissue of *P. hypophthalmus* reared in different salinity, 1. Pillar cells, 2. Epithelium, 3. Lacuna, 4. Blood vessel, 5. Mucus cells, 6. Chloride cells, 7. Hypertrophy of chloride cells, 8. Secondary lamella bending. 400x magnification, Staining HE

In addition, abnormalities occur in the structure of the gill tissue in 9 ppt salinity, and secondary lamella bending usually occurs in fish that experience hyperplasia abnormalities. This follows Nirmala et al.[13], which states that gill bending is caused by enlarged cells damaged in hyperplasia, causing an abnormal shape. When compared with the results of Jiang et al.[14], silver carp (*Hypophthalmichthys molitrix*) exposed to 6 ppt salinity experienced changes in the structure of the gill organs, including cracking and breaking of the main part of the gill filaments and bending of the secondary lamella. Further research results in Hossain et al.[3] showed that the gill organs of Striped catfish were abnormal at 12 ppt salinity, changes in chloride cell hypertrophy were seen, telangiectasia at the ends of the secondary lamella, epithelial necrosis, fusion of several secondary lamellae and removal of the squamous epithelium.

Histopathology of Striped Catfish Kidney

Changes that occur in the aquatic environment will, directly and indirectly, impact the kidneys' structure, where the kidneys function as an osmoregulation tool (maintaining salt levels in the fish's body), as a blood filter, and retain substances needed by the body[15]. Striped catfish kidney tissue

e-issn: 2716-4608
p-issn: 2655-366X

structure reared at different salinities can be seen in Figure 2.

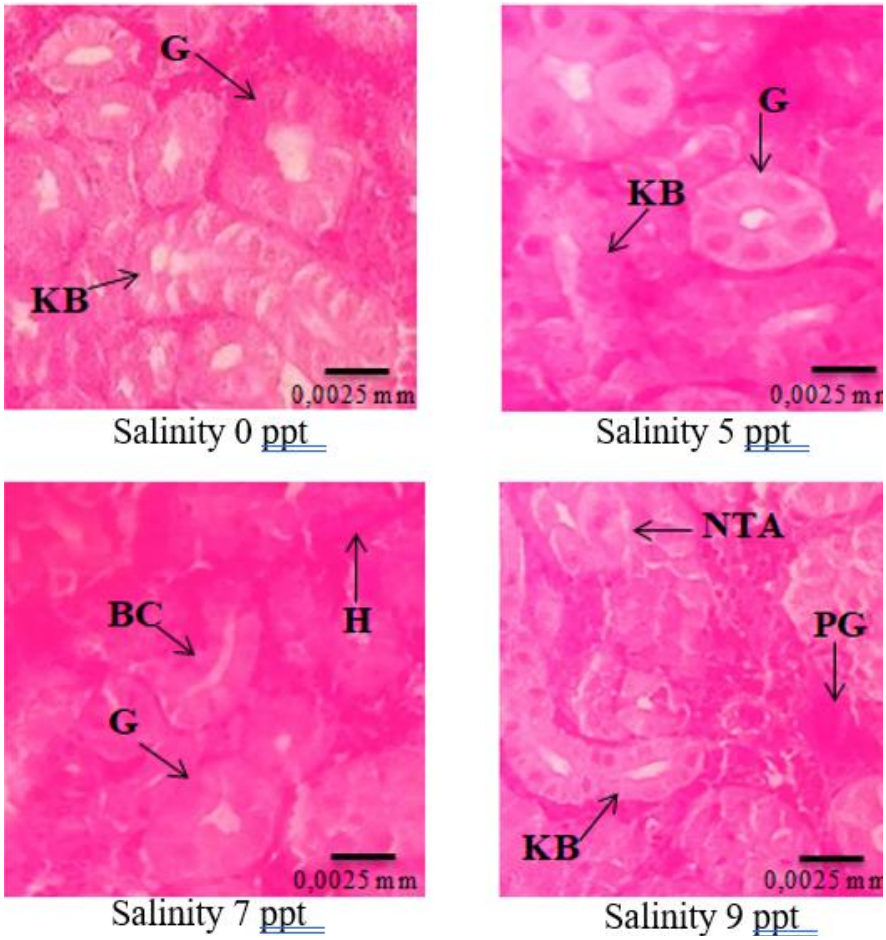

Figure 2. Structure of kidney tissue of *P. hypophthalmus* reared in different salinity, G = Glomerulus, BC = Bowman's Capsule, ATN= Acute Tubular Necrosis, H= Hemorrhage. 400x magnification, Staining HE

Based on Figure 2, no abnormal changes were seen between 0 and 5 ppt salinity. The normal striped catfish kidney structure was shown to be neatly arranged by the glomerulus, which was covered by Bowman's capsule. Bjørgen & Koppang[16], the structure of normal fish kidney tissue is characterized by the presence of cells that make up the glomerulus that is still clearly visible, not completely round but in the shape of a six and bowmen's capsule looks neatly wrapped around the glomerulus.

Abnormal changes in the kidney organs of striped catfish were seen at a salinity of 7 ppt, and there was hemorrhage, and at a salinity of 9 ppt, there was acute tubular necrosis. Hemorrhage is bleeding due to damaged blood vessels. Jamin & Erlangga[17] stated that hemorrhage is bleeding in cells caused by the rupture of blood vessels, causing blood to flow where it shouldn't, both outside the body and into body tissues. Diba & Rahman[18] stated that acute toxic exposure, the presence of bacteria, viruses, and parasites can cause hemorrhagic damage. Necrosis was found in the kidney organs of striped catfish, especially the tubular parts. Necrosis describes a state of decreased tissue activity, characterized by the loss of several cell parts one by one from one tissue so that, in a short time, they will die[19].

Characteristics of necrotic tissue include having a paler colour than normal, loss of viability, and fading cells[20]. This is related to the effects of exposure to salinity. When compared with the results of Hossain et al.[3], abnormalities in the kidney organs of

striped catfish occurred at a salinity of 12 ppt, showing degeneration of the renal tubules, enlarged Bowman's capsule, increased space between the glomerulus and necrotic cells in the hematopoietic tissue.

4. CONCLUSION

The histopathological structure of the gills and kidneys of striped catfish occurs as the salinity increases. The salinity of 9 ppt showed changes in gill structure, which caused chloride cell hypertrophy and secondary lamella bending, while the kidneys experienced acute tubular necrosis. Using a salinity of 5 ppt is recommended to maintain striped catfish.

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