Veterinary Integrative Sciences 2020; 18(1): 33-41

Vet Integr Sci

Veterinary Integrative Sciences

ISSN; 2629-9968 (online)

Website; www.vet.cmu.ac.th/cmvj

Research article

Histological observation of digestive system of malayan halfbeak, *Dermogenys pusilla* (Kuhl & van Hasselt, 1823) during juvenile stage from Thailand

Sinlapachai Senarat[1,*], Pisit Poolprasert[2], Jes Kettratad[1,3], Piyakorn Boonyoung[4], Wannee Jiraungkoorskul[5], Shuaiqin Huang[6], Theerakamol Pengsakul[7], Pahol Kosiyachinda[8] and Chanyut Sudtongkong[9]

[1]Department of Marine Science, Faculty of Science, Chulalongkorn University, Bangkok 10330, Thailand
[2]Program of Biology, Faculty of Science and Technology, Pibulsongkram Rajabhat University, Phitsanulok 65000, Thailand
[3]Aquatic Resources Research Institute, Chulalongkorn University, Bangkok 10330, Thailand
[4]Department of Anatomy, Faculty of Science, Prince of Songkla University, Songkhla 90110, Thailand
[5]Department of Pathobiology, Faculty of Science, Mahidol University, Bangkok 10400, Thailand
[6]State Key Laboratory of Cellular Stress Biology, and Parasitology Research Laboratory, School of Life Sciences, Xiamen University, Xiamen, Fujian, China
[7]Faculty of Medical Technology, Prince of Songkla University, Songkhla 90110, Thailand
[8]Department of Biology, Faculty of Science, Mahidol University, Bangkok 10400, Thailand
[9]Department of Marine Science, Faculty of Science and Fisheries Technology, Rajamangala University of Technology Srivijaya, Trang Campus, Sikao, Trang 92150, Thailand

Abstract

Histological information of digestive system of fish in Family Hemiraphidae has never been reported. Although the malayan halfbeak *Dermogenys pusilla* is a carnivorous hemiraphid native to Southeast Asia, the structure of its digestive information is not known. Here is the first report on basic histology of digestive system of *D. pusilla*, as a representative from Family Hemiramphidae. All specimens (n=20) were collected in October 2016 from Pranburi River estuary, Thailand. The digestive system of *D. pusilla* was divided into digestive tract and accessory organs (liver and pancreas). The mouth part of this fish was sub-terminal. It had elongated oral cavity. Epithelial mucosa of the oral cavity was covered with stratified epithelium with mucous secreting cells. The epithelial pharynx was similar to that of oral cavity with infiltration of prominent pharyngeal teeth. Although this hemiramphid fish had no stomach, the long intestine was divided into anterior and posterior intestines. All intestinal regions were lined with simple columnar epithelium and goblet cells; however, the goblet cell abundance in the posterior intestine was greater than in the anterior intestine. Several polyhedral hepatocytes of liver tissue appeared to be distinctive sinusoid, whereas pancreatic parenchyma usually contained clusters of pyramidal cells in the acini. The pancreatic cells also contained large eosinophilic zymogen granules. In the present study, we showed histological characteristics of digestive system of *D. pusilla*, implying carnivorous nature of this fish.

Keywords: Estuarine area, Hemiramphid fish, Intestine, Histology, Histomorphology

***Corresponding author:** Sinlapachai Senarat; Department of Marine Science, Faculty of Science, Chulalongkorn University, Bangkok 10330, Thailand. Email: Senarat.S@hotmail.com

Article history; *received manuscript: 21 January 2020, revised manuscript: 27 January 2020, accepted manuscript: 8 February 2020, published online: 11February 2020*
Academic editor; *Korakot Nganvongpanit*

INTRODUCTION

Morphological and histological studies of digestive system in teleosts are very useful to understand their biology from their structures and functions. This information will provide some insights on their ecology and feeding habitats, which could applied in further studies such as ultrastructure, histopathology, immunohistochemistry and taxonomy. Studies on histology of digestive system of many fish have been reported for example *Seriola dumerili* (Grau et al., 1992), *Stegastes fuscus* (Canan et al., 2012), *Puntius stoliczkanus* (Senarat et al., 2013a) and *Hemibagrus filamentus* (Senarat et al., 2013b). All results provided were of great importance to understand their structures and biology associated with different feeding habits and diet diversity.

The malayan halfbeak *Dermogenys pusilla* represents a species of hemiramphid fish, which is one of the most abundant and common fish in freshwater and brackish water of rivers and coastal regions in Southeast Asia (i.e., Indonesia, Malaysia, Singapore, and especially Thailand). *D. pusilla* is a popular aquarium species due to its small and colored performance (Kottelat, 2013). Adult fish can reach the size up to 6.9 cm (Senarat et al., 2019). Although, *D. pusilla* was believed to be a carnivorous fish and an important link in food chain and food web in an ecosystem, histology of digestive system of this fish has never been reported. In the current study, we observed to describe the histology of digestive system in juvenile *D. pusilla* using histological techniques.

MATERIALS and METHODS

In this study, a total of twenty female *D. pusilla* were collected on October 2016 from five stations in the Estuary Pranburi River, Thailand (N 12°24'16.5" / E 099°59'20.2", N 12°24'16.5" / E 099°59'20.2", N 12°24'06.3" / E 099°58'58.0", N 12°24'18.5" / E 099°58'36.0", and N 12°24'15.3" / E 099°58'28.6"). We followed the experimental protocol officially approved and granted by the Animal Care and Use Committee of Faculty of Science, Chulalongkorn University, Thailand (Protocol Review No. 1723004).

The fish (n=20) were euthanized by the rapid cooling method (Wilson et al., 2006). They (n=18 with total length of 3.5±0.97 cm) were longitudinally opened, dissected for the study of their morphological study of digestive tract and then fixed in Davidson's fixative about 24 hr at room temperature (Dietrich and Krieger, 2009). The samples of the smallest size of whole bodies of fish (n=2 with the total length of 2.2±0.66 cm) were fixed in the same fixative above. After that all samples were processed under standard histological techniques (Presnell and Schreibman, 1997; Suvarna et al., 2013) and sectioned at 4 µm thickness using a rotary microtome. These sections were progressively stained with Harris' hematoxylin and eosin (H&E), and photographed with Leica DM750 light microscope.

RESULTS and DISCUSSION

The digestive system of *D. pusilla* was divided into the digestive tract (oral cavity, pharynx, esophagus and intestine) and accessory organs (liver and pancreas). In term of the digestive tract was divided into oral cavity, pharynx, esophagus and intestine (Figures 1A-1B). Details are as following:

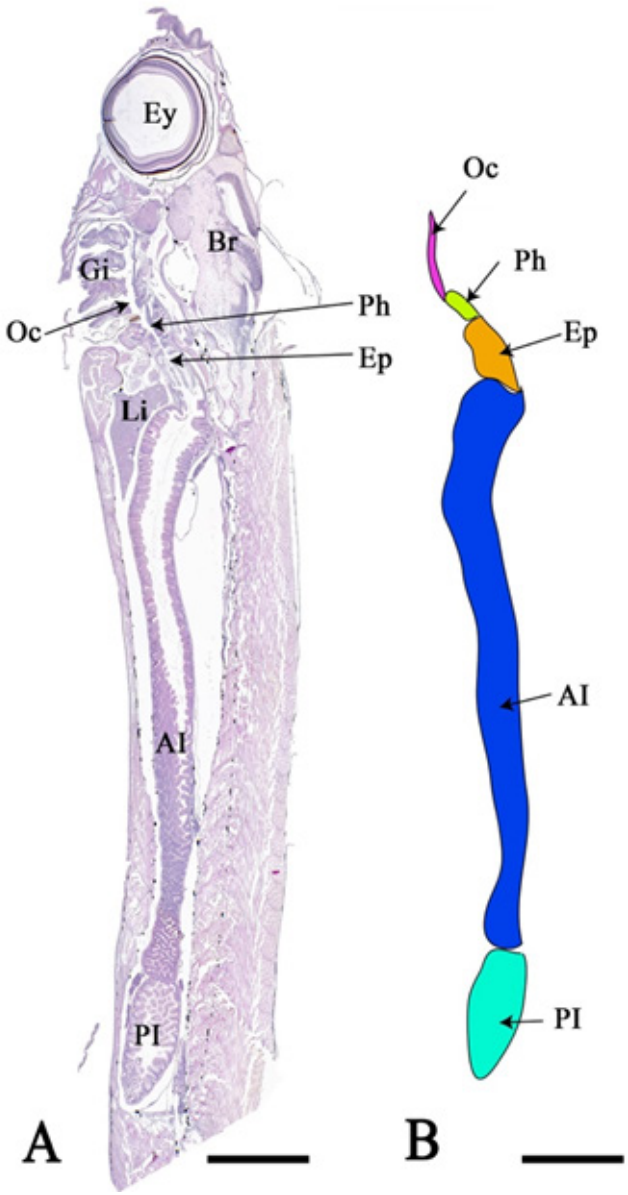

Figure 1 Light micrographs (A) and schematic diagram (B) of the whole digestive tract of *Dermogenys pusilla* including oral cavity (Oc), pharynx (Ph), esophagus (Ep), anterior intestine (AI), and posterior intestine (PI). Abbreviations: Br=brain, Ey=eye, Gi=gill. Scale bar = 1 mm

Oral cavity

The mouth of this fish was characterized with subterminal feature, having elongate sharp (Figure 2A). Histologically, the mucosal layer was covered with stratified polygonal squamous epithelium (Figure 2B). This histological arrangement agreed to that of other teleosts (Albrecht et al., 2001) i.e., *Cyprinus carpio* and *Gnathonemus petersii* (Genten et al., 2008), whereas the mucosa of *Squalus acanthias* was lined with a stratified cuboidal epithelium (Andrew and Hickman 1974). In addition, abundance taste buds appeared to be typical pear-shaped and secreting cells were observed as stratified epithelium (Figures 2C-2D). Submucosal layer was lined just beneath the mucosa and contained loose connective tissue (Figure 2C). A layer of smooth muscle tissue was observed in the muscularis layer.

Pharynx

Longitudinal section showed that pharynx was located between oral cavity and esophagus (Figure 2D). Prominent pharyngeal teeth were observed on upper jaw (Figure 2D). There were immature and mature teeth (Figure 2E). According to this pattern, several teeth penetrated through stratified epithelium (Figure 2E), similar to these of zebrafish (Menke et al., 2011). Both submucosal and muscularis layers were also present, as in oral cavity (Figures 2D-2E).

Esophagus

A narrowing structure of esophagus was found in a small region (Figure 2D). The presence of this structure indicated that food items can be directly passed into intestine with no anatomical boundaries, as supported by Stevens and Hume (1995). The same feature was observed in marine carnivorous fish for example *Lutjanus synagris*, *L. purpureus* and *Ocyurus chrysurus* (Morais et al., 2014). The esophageal wall was histologically unique and completely consisted of mucosa, submucosa, muscularis, and serosa (Figure 2F). The mucosa (epithelium and lamina propria) lined with a stratified epithelium forming longitudinal fold and protruded into esophageal lumen (Figure 2F). A number of goblet cells were also observed near apical epithelium (Figure 2F). These goblet cells may involve in immunological responses against bacterial infections and osmoregulatory functions (Albrecht et al., 2001; Senarat et al., 2015). The submucosa layer consisted of loose connective tissue, and had some blood vessels (Figure 1). The muscularis was beneath the submucosa (Figure 2F). The inner and outer muscularis layers contained the organized muscle fibers (Figure 2F), which were important in the translocation of food items (Cao and Wang, 2009). The outermost layer of esophagus was the serosa, which was lined with a simple squamous epithelium (Figure 2F).

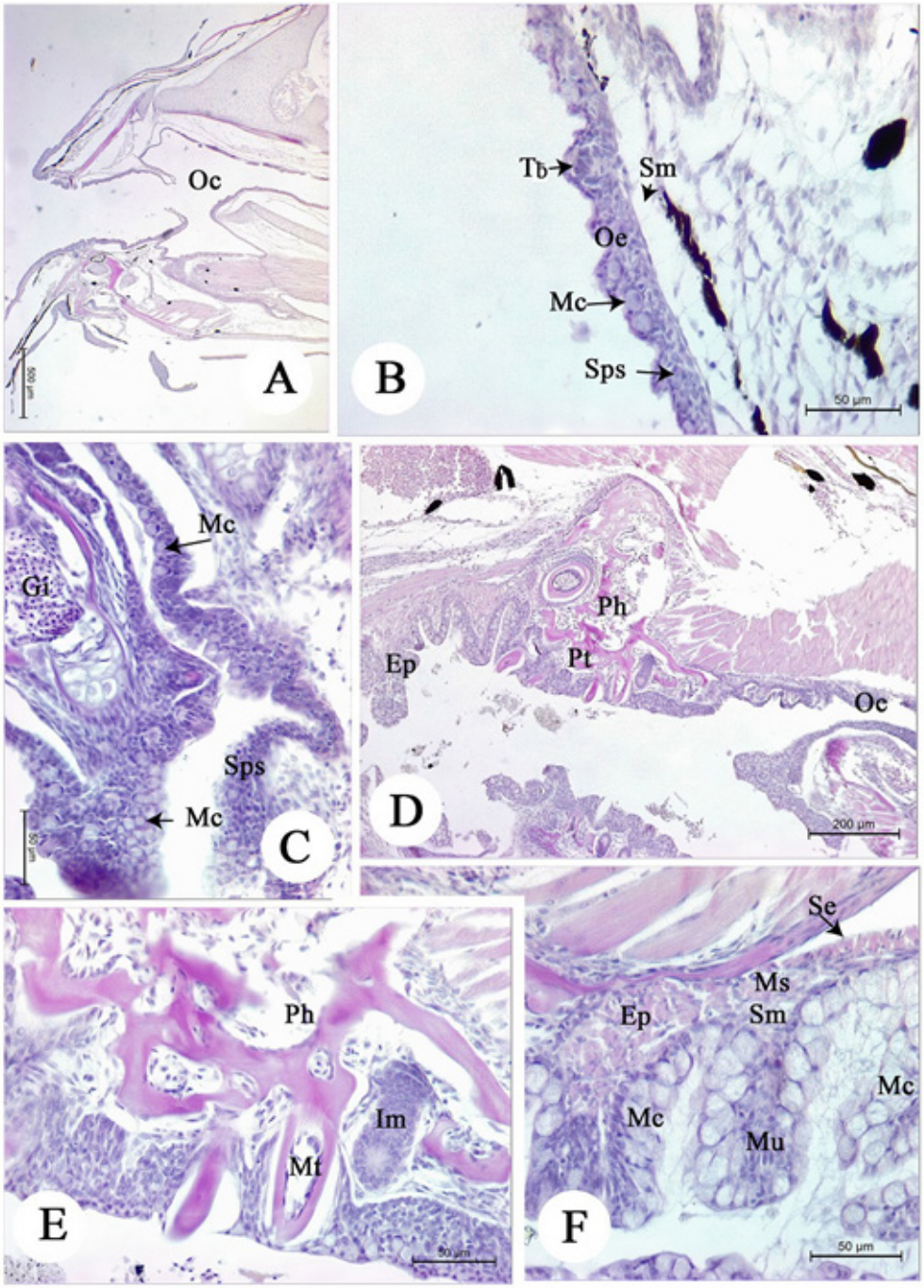

Figure 2 Light micrographs showing oral cavity (Oc) (A-C), pharynx (Ph) (D-E), esophagus (Ep) (F) of digestive tract of *Dermogenys pusilla*. Abbreviations: Gi=gill, Im=immature teeth, Mc=mucous cells, Mt=mature teeth, Ms=muscularis, Mu=mucosa, Oe=oral epithelium, Ph=pharynx, Pt=pharyngeal teeth, Se=serosa, Sps=stratified polygonal squamous epithelium, Sm=submucosa, Tb=taste bud.

Intestine

Our results showed that stomach of *D. pusilla* was absent, as in a number of fish species including cyprinid fish (carp), killifish (Cypridontidae), clingfish (Gobiesocidae) and gobies (Gobioidae) (Harder, 1975). According to their cellular and histological components, the intestinal structure could be further divided into anterior and posterior intestines (Figures 1A-1B), as likely seen in previous observations (Dai et al., 2007; Abdulhadi, 2005). The transition between the esophagus and the anterior intestine was histologically apparent, from the complex structure of esophagus to a simple layer of simple columnar epithelium (Figure 3A). Intestinal wall of the anterior intestine consisted of numerous mucosal folds with sparse goblet cells, forming as finger-like fold structure (Figure 3B). Each fold was covered with a simple ciliated columnar epithelium, forming thick brush border (Figure 3C). Each columnar cell contained a basal nucleus and was surrounded by an acidophilic cytoplasm (Figure 3C). Similar observation has been previously described in *Scorpaena porcus* (Nazlić et al., 2014) and *Merluccius merluccius* (Bočina et al., 2016). Submucosal, muscularis, and serosal layers were loosely formed and rarely separated (Figure 3C). In addition, we found that the structure of the posterior intestine was similar to that of the anterior intestine; however, the abundance of goblet cells in this region was greater than the anterior part of the intestine (Figure 3D-3E). This observation was similar in *Stegastes fuscus* (Canan et al., 2012) and *Rastrelliger brachysoma* (Senarat et al., 2015). The greater number of goblet cells in the posterior intestine may be related to the need for significantly increased mucosa protection and lubrication for fecal expulsion (Murray et al., 1994).

Liver

It was located between esophagus and anterior intestine (Figure 4). Several functions of liver specifically concerned to the assimilation of nutrients, detoxification and metabolic homeostasis (Genten et al., 2008). Parenchyma of liver in this species structurally composed of several polyhedral hepatocytes and a portal tract. Hepatocyte had a large cell with a spherical nucleus in the nucleus, which were separated by sinusoids (Figure 3F). This pattern had similar observed in *R. brachysoma* (Senarat et al., 2018a). Several bile ducts were seen among the liver parenchyma. The structure of bile ducts was lined by a simple cuboidal epithelium and surrounded by connective tissue (Data not shown).

Pancreas

Parenchyma of the pancreas was located along the intestines (Figure 4) and was histologically composed of many clusters of pyramidal cells in the acini and portal afferent veins. It had been similarly observed in *R. brachysoma* (Senarat et al., 2018b), *Gnathonemus petersii* and *Poecilia reticulata* (Genten et al., 2008). Characteristic of the pyramidal pancreatic cell was the presence of a prominent nucleus, surrounded by basophilic cytoplasm which contained various eosinophilic zymogen granules (Figure 4). It is well-known that the intracytoplasmic zymogen granules play an important role for the digestion of proteins, carbohydrates, fats, and nucleotides (Genten et al., 2008).

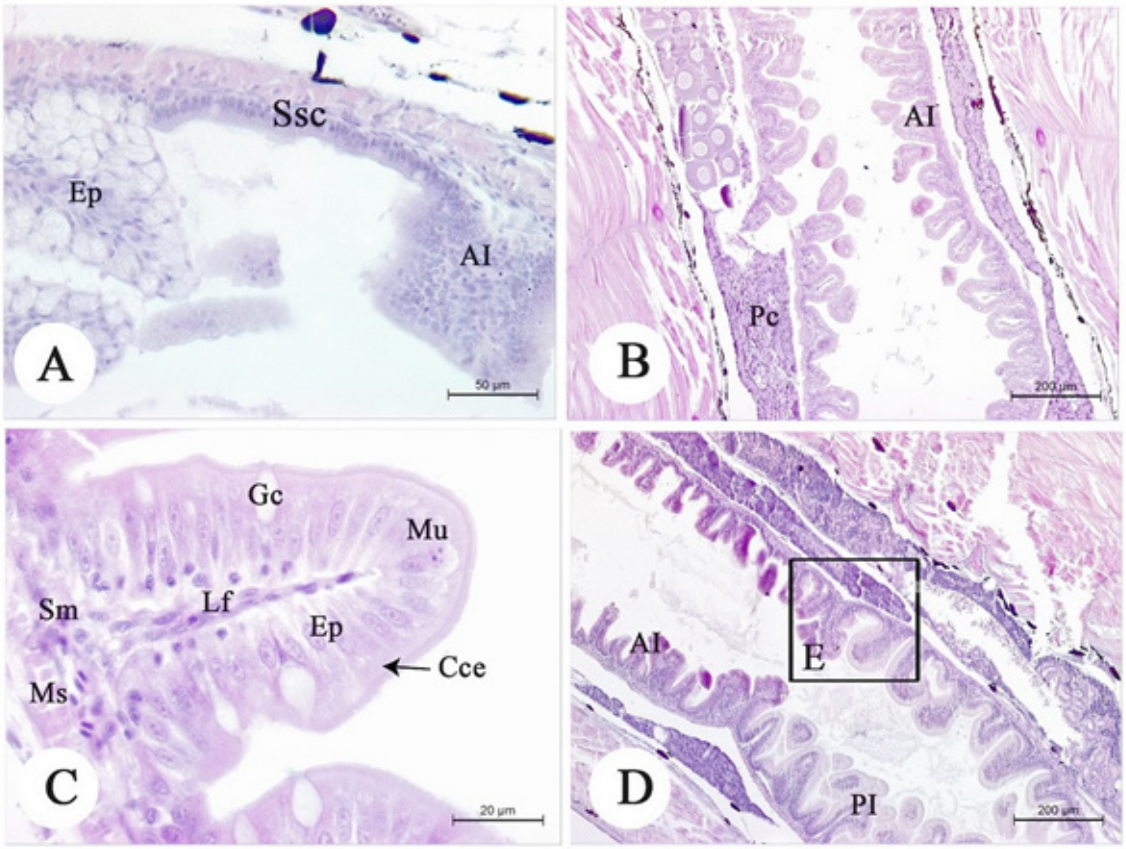

Figure 3 Light micrographs showing anterior intestine (AI) and posterior intestine (PI) of digestive tract of *Dermogenys pusilla* [A-E]. Abbreviations: Cce=ciliated simple columnar epithelium, Ep=epithelum, Gc=goblet cell, Lf=longitudinal fold, Mu=mucosa, Ms=muscularis, Sm=submucosa, Ssc=single layer of simple columnar epithelium.

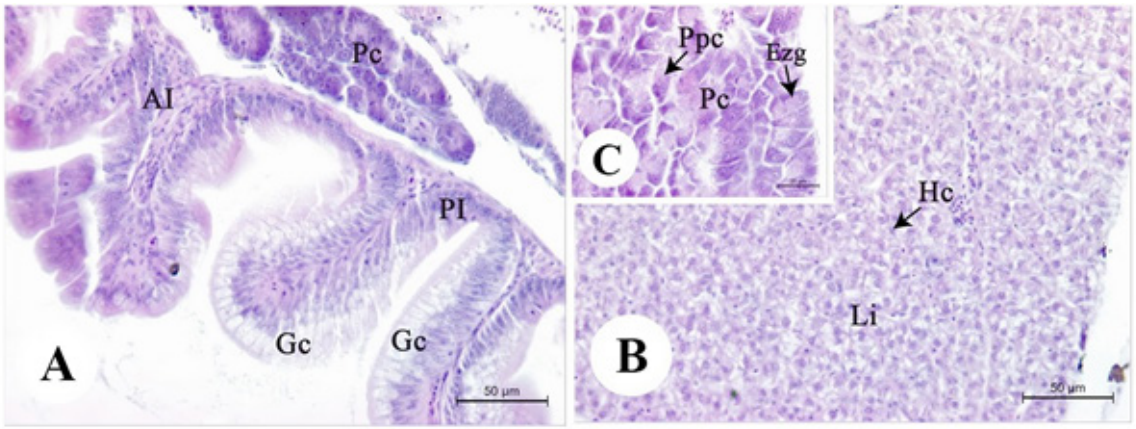

Figure 4 Light micrographs showing pancreas (Pc) and liver (Li) of *Dermogenys pusilla*. Abbreviations: AI=anterior intestine, Ezg=eosinophilic zymogen granules, Gc=goblet cell, Hc=hepatocytes, PI=posterior intestine, Ppc=pyramidal pancreatic cell.

CONCLUSION

We first described the digestive structure of *D. pusilla*. Surely, our data provided a better understanding of digestive histology of this species, which will be applied to deep knowledge for example histopathology and physiology of this fish and other members of Family Hemiramphidae.

CONFLICT of INTEREST

There is no conflict of interest.

REFERENCES

Albrecht M.P., Ferreira, M.F.N., Caramaschi, E.P., 2001., Anatomical features and histology of the digestive tract of two related neotropical omnivorous fishes (Characiformes; Anostomidae). J. Fish Biol. 58, 419-430.

Abdulhadi, H., 2005. Some comparative histological studies on alimentary tract of tilapia fish (*Tilapia spilurus*) and sea bream (*Mylio cuvieri*). Egypt. J. Aquat. Res. 31, 387-39.

Andrew, W., Hickman, C.D., 1974. Digestive system. – In: Andrew W., Hickman, C.D. (Eds.), Histology of the Vertebrates. Mosby, St. Louis., MO, USA, pp. 243-286.

Bočina, I., Ružić, S., Restović, I., Paladinm A., 2016. Histological features of the digestive tract of the adult European hake *Merluccius merluccius* (Pisces: Merlucciidae). Ital. J. Zool., 83, 26-33.

Canan, B., Nascimento, W.S., Silva, N.B., Chellappa, S., 2012. Morphohistology of the digestive tract of the damsel fish *Stegastes fuscus* (Osteichthyes: Pomacentridae). Sci. World J., 1-9.

Cao, X.J., Wang, W.M., Song, F., 2011. Anatomical and histological characteristics of the intestine of the top mouth culter (*Culteral burnus*). Anat. Histol. Embryol., 40, 292–298.

Dai, X., Shu, M., Fang, W., 2007. Histological and ultrastructural study of the digestive tract of ricefield eel, *Monopterus albus*. J. Appl. Ichthyol., 23, 177-183.

Dietrich, D.R., Krieger, H.O., 2009. Histological analysis of endocrine disruptive effects in small laboratory fish. John Wiley and Sons, New Jersey, U.S.A.

Genten, F., Terwinghe, E., Danguy, A., 2008. Atlas of fish histology. Science Publishers Enfield, NH, U.S.A.

Grau, A., Crespo, S., Saraquete, M.C., Gonzalez, M.L., 1992. The digestive tract of the amberjack *Seriola dumerili*, Risso: a light and scanning electron microscope study. J. Fish Biol. 41, 287-303.

Harder, W., 1975. Anatomy of fishes. Stuttgart. Schweizerbart'sche Verlagsbuchhandlung, Pt.1:612 p., Pt.2:132.

Kottelat, M., 2013. The fishes of the inland waters of Southeast Asia: A catalogue and core bibliography of the fishes known to occur in freshwaters, mangroves and estuaries. Raffles Bull. Zool., 27, 1–663.

Menke, A.L., Spitsbergen, J.M., Wolterbeek, A.P., Woutersen, R.A, 2011. Normal anatomy and histology of the adult zebrafish. Toxicol. Pathol., 39, 759–775.

Morais, A.L.S., Carvalho, M.M., Cavalcante, L.F.M., Oliveira, M.R., Chellappa, S., 2014. Características morfológicas do trato digestório de três espécies de peixes (Osteichthyes: Lutjanidae) das águas costeiras do Rio Grande do Norte, Brasil. Biota Amazônica. Macapá., 4, 51–54.

Murray, H.M., Wright, G.M., Goff, G.P., 1994. A comparative histology and histochemical study of the stomach from three species of pleuronectid, the Atlantic halibut, *Hippoglossus hippoglossus*, the yellow tail flounder, *Pleuronectes ferruginea*, and the winter flounder, *Pleuronectes americanus*. Can. J. Zool., 72, 1199–1210.

Nazli, Ć.M., Paladin, A., Bo, Č.I., 2014. Histology of the digestive system of the black scorpion fish *Scorpaena porcus* L. Acta Adriatica., 55, 65–67.

Presnell, J.K., Schreibman, M.P., 1997. Humason's animal tissue techniques, 5th ed. US, Johns Hopkins University Press.

Stevens, C.E., Hume, I.D., 1995. General characteristics of the vertebrate digestive system. In: Comparative Physiology of the vertebrate digestive system. Cambridge University Press.

Senarat, S., Jiraungkoorskul, W., Kettratad, J., Kaneko, G., Poolprasert, P., Para, C., 2019. Histological analysis of reproductive system of *Dermogenys pusilla* (Kuhl & van Hasselt, 1823) from Thailand: Sperm existence in ovary indicates viviparous reproductive mode. Maejo Int. J. Sci. Technol., 13, 185-195.

Senarat, S., Kettratad, J., Tipdomrongpong, S., Pengsakul, T., Jiraungkoorskul, W., Boonyoung, P., Huang, S., 2018a. Histopathology of kidney and liver in the captive broodstock (*Rastrelliger brachysoma*) during its juvenile stage. Vet. Integr. Sci., 16, 87-93.

Senarat, S., Kettretad, J., Poolprasert, P., Tipdomrongpong, S., Plumley, F.G., Jiraungkoorskul, W., 2018b. Health status in wild and captive *Rastrelliger brachysoma* from Thailand: Histopathology. Songklanakarin J. Sci. Technol., 40, 1090-1097.

Senarat, S., Kettratad, J., Jiraungkoorskul, W., Kangwanrangsan, N., 2015. Structural classifications in the digestive tract of *Rastrelliger brachysoma* (Bleeker, 1851). Songklanakarin J. Sci. Technol., 37, 561-567.

Senarat, S., Yenchum, W., Poolprasert, P., 2013a. Histological Study of the intestine of stoliczkae's Barb *Puntius stoliczkanus* (Day, 1871) (Cypriniformes: Cyprinidae). Kasetsart J. (Nat. Sci.)., 47, 247-251.

Senarat S., Kettratad, J., Poolprasert, P., Yenchum, W., 2013b. Histological structure of the esophagus and stomach in yellow mystus, *Hemibagrus filamentus* (Fang and Chaux, 1949). Thammasat J. Sci. Technol., 1, 46-54. (In Thai)

Suvarna, K.S., Layton, C., Bancroft, J.D., 2013. Bancroft's theory and practice of histological techniques. 7th ed. Canada, Elsevier.

Wilson, J.M., Bunte, R.M., Carty, A.J., 2009. Evaluation of rapid cooling and tricaine methanesulfonate (MS222) as methods of euthanasia in zebrafish (*Danio rerio*). J. Am. Assoc. Lab. Anim. Sci., 48, 785-789.

How to cite this article;

Sinlapachai Senarat, Pisit Poolprasert, Jes Kettratad, Piyakorn Boonyoung, Wannee Jiraungkoorskul, Shuaiqin Huang, Theerakamol Pengsakul, Pahol Kosiyachinda and Chanyut Sudtongkong. Histological observation of digestive system of malayan halfbeak, *Dermogenys pusilla* (Kuhl & van Hasselt, 1823) during juvenile stage from Thailand. Veterinary Integrative Sciences. 2020; 18(1): 33-41.