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Modified Organs of Air Breathing Fishes in Korea

By Chi-Hong Kim and Jong-Young Park^{1,*}

Central Inland Fisheries Research Institute, Cheongpyeong-ri Cheongpyeong-myeon Gapyeong-gun, Gyeonggi-do 477-815, Korea,
¹Department of Biological Sciences and Institute for Biodiversity Research, College of Natural Sciences, Chonbuk National University, Jeonju 561-756, Korea

ABSTRACT Modified organs for air respiration in Korean fish was reviewed in the following 6 Korean fishes: three mudskippers (*Boleophthalmus pectinirostris, Periophthalmus modestus* and *P. magnus-pinnatus*), two mud loaches (*Misgurnus mizolepis* and *M. anguillicaudatus*), and a torrent catfish (*Lioba-grus mediadiposalis*). Three mudskippers and a torrent catfish have a modified epidermis to in order to make up for the deficient oxygen supply. Their epidermis has abundant intraepithelial blood capillaries except dermal capillaries situated just beneath the stratum germinativum of the epidermis in *B. pectinirostris*. The epidermis was thick due to component of the following cells: two kinds of glands as a small mucous cells and a large club cells in *L. mediadiposalis*, voluminous cells (swollen cells) swollen by epidermal cells and a small mucous cells in *B. pectinirostris*, and only voluminous cells having no any glandular cells in *P. modestus* and *P. magnuspinnatus*. In Particular, the epidermis of the mudskippers appears to be a web-like structure due to the swollen epithelial cells. The dermal bulges are found in *B. pectinirostris* and they are situated at the skin covering the body, not appendage of all the fins and the sucking disc. Another modified organ in *M. mizolepis* and *M. anguillicaudatus* occurs in intestine and its mucosal epithelium has abundant blood capillaries.

Key words: Modified organ, air respiration, blood capillaries, Korean fish

INTRODUCTION

The fishes are able to constantly meet their oxygen demands by performing aquatic respiration with their gills. However, some fishes live in warm and stagnant reservoirs, ponds and rice fields undergoing periodic drought which causes a reduction of dissolved oxygen in the water. Mudskippers, known as amphibious fish, mostly inhabit mangrove and mudflat areas of intertidal zone which are exposed to the atmosphere during low tide. To adapt to such poor dissolved oxygen conditions, those fishes have developed additional respiratory apparatus to take up air to supplement their oxygen supply through the following organs: the intestine (Cobitidae), the skin (Cobitidae, Anguillidae, Nototeridae, Gobiidae), the branchial chambers (Anabatidae, Osphromenidae, Channidae), the swim bladder (Dipnoi), the labyrinthine organ (Anabas), and others (Liem, 1967; Johansen, 1970; Tamura et al., 1976; Niva et al., 1981; Munshi and Hughes, 1986; Whitear, 1986; Moitra et al., 1989; Low et al., 1990; Itazawa and Hanyu, 1991; Suzuki, 1992; Yokoya and Tamura, 1992; Ishimatsu et al., 1998; Zhang et al., 2000; Park et al., 2000; Park and Kim, 2001; Park, 2002; Park et al., 2003a, b; Park et al., 2004).

In Korean fishes, mudskippers as Boleophthalmus pectinirostris, Periophthalmus modestus and P. magnuspinnatus perform amphibious life which they move or skip briskly on the inter-tidal habitats and spend time in burrows filled with hypoxic water. Mud loaches, Misgurnus mizolepis and M. anguillicaudatus, live at the bottom of muddy, stagnant swamps and warm reservoirs or rice fields which are subjected to periodic drying. The Korean endemic torrent catfish, Liobagrus mediadiposalis, inhabits the rocky or stony bottoms of mountain streams or torrents which are low or lacking in oxygen in the drying season. Through those fish live in hypoxic environments, they have been adapted to their environments. Therefore, the purpose of this paper is to introduce and compare the histological features on their epidermis and intestine based on habitats of the above Korean fishes.

^{*}Corresponding author: Jong-Young Park Tel: 82-63-270-3344, Fax: 82-63-270-3362, E-mail: park7877@jbnu.ac.kr

1. Features of modified skin

1) Boleophthalmus pectinirostris

The epidermis of the amphibious mudskipper fish, B. pectinirostris, consists of three layers: the outermost layer, middle layer and stratum germinativum. The outermost layer consists of polygonal cells or rather flattened cells arranged in one to eight layers. In between these cells, round small cells and mucous cells are present. The round small cells are ovoid to round with an inclusion of fine granules, which are weakly positive to acid chemical reaction. The mucous cells are predominantly acid mucopolysaccharide in nature. The middle layer consists of 1 to 50 layers of small or voluminous cells swollen by epidermal cells. Owing to various sizes and layers of the swollen cell, the middle layer shows a web-like structure. The stratum germinativum consists of a single layer of cuboidal cells, or more or less columnar cells. A dermal bulge is located at each apical area of the epidermis of the body regions, but does not exist in all the fins or in the sucking disc. In the epidermis of the body regions, the dermal bulges are sparsely occupied by dermal tissue and have dermal capillaries just beneath the stratum germinativum. The value of the diffusion distance is the lowest in the top of the head (mean 5.5 μ m \pm 1.6) and the highest in the ventral region (mean $44.3 \pm 12.7 \,\mu\text{m}$).

2) Periophthalmus magnuspinnatus

A histological study was performed to know whether the skin covering the whole body (eight body regions) and the appendages (five on the fins and one on the sucking disc), is related actually with respiration and which region of skin is more effective. The structure of the epidermis was the same in all the regions which consisted of the superficial layer, middle layer and stratum germinativum. A large number of fine blood capillaries were mainly situated at the superficial layer and rare at the middle layer. The diffusion distance between the capillary endothelial cells and the surface of the epidermis ranged from mean 2.0 to 15.4 µm: the lowest value in the back (mean $2.0 \,\mu\text{m} \pm 0.8$) and the highest value in the base of the anal fin (mean 15.4 μ m \pm 9.7). Among 14 regions of the epidermis, it can surmise that the two dorsal fins toward the upper region may be more exposed to the air for long time than other regions during amphibious life of P. magnuspinnatus.

3) Periophthalmus modestus

Interestingly, the epidermis has no gland cell. The epidermis consists of three regions: outermost layer of one to five layers of flattened epithelial cells, middle layer of the swollen epithelial cells instead of glandular cells and stratum germinativum of cuboidal cells. There are numerous blood capillaries in the outermost layer of the epidermis and the diffusion distance between the blood of capillaries and the epidermis in the back is about 1.4 μ m (± 0.6). The middle layer of the epidermis appears to

be a web-like structure due to the swollen epithelial cells. The stratum germinativum has a well-developed lymphatic space containing lymphocytes. There are numerous blood capillaries and elliptical area with acid mucopoly-saccharides in stratum laxum of the dermis.

4) Liobagrus mediadiposalis

The epidermis of the torrent catfish, *L. mediadiposalis*, consists of three layers: the outermost layer, middle layer and stratum germinativum. The epidermis consists of two types of skin glands, a small mucus cell and voluminous club cell. The unicellular mucous cell is acid sulfomucins (some sialomucins) and the club cell, sometimes binucleate, is proteinaceous. Well-developed vascularization is one of the characteristics of epidermis of *L. mediadiposalis*. The distance between the vascular capillaries and the surface of epidermis is mean $169 \, \mu m \, (\pm 50.9)$, ranging from $22.5 \, \text{to} \, 220 \, \mu m$. Well-developed lymphatic spaces contain lymphocytes in the epidermis.

2. Features of modified intestine

1) Misgurnus mizolepis and M. anguillicaudatus

The straight intestinal tracts of the mud loach M. mizolepis and M. anguillicaudata are divided into an intestine and rectum which consisted of a mucosa (epithelial layer), lamina propria-submucosa, muscularis and serosa. The intestine and rectum have shorter mucosal folds and a thinner wall. Extensive vascular capillary networks were present near the luminal surface of the intestine and the rectum. The diffusion distance between the vascular capillaries and the viscus lumen in the intestinal and rectal mucosal epithelium was about 0.7 μ m (\pm 0.1) in M. mizolepis and 11.2 um (± 1.1) in M. anguillicaudatus. The intestine and rectum of M. mizolepis probably have a respiratory function to address the deficient oxygen supply within their environment. The epithelial mucous cells in the both loaches contain acidic or a mixture of acidic and neutral mucins, the former being the most common.

3. Comparison with other air-breathing fishes

The epidermal structures in related to aerial cutaneous respiration in some fishes, which inhabit such environments as a reduction of dissolved oxygen in the water or amphibious habitats life on water and land, exhibits relatively developed or modified structures for supply of deficient oxygen; that is, 1) the thick epidermis has large gland cells or a large swollen cell, 2) a large number of intraepithelial blood capillaries, 3) defined lymphatic spaces containing small lymphocytes in the stratum germinativum, 4) the dermis has a definite area with acid mucopolysaccharides and abundant blood vessels, and 5) a reduction or absence of scales (Jakubowski, 1958; Liem, 1967; Johansen, 1970; Mittal and Munshi, 1971; Mittal and Banerjee, 1974; Mittal et al., 1980; Whitear, 1986; Suzuki, 1992; Yokoya and Tamura, 1992; Park et

Table 1. Diffusion distance of skin and intestine in air breathing fishes

Species	Literatures	Air-breathing organs	Diffusion distances (μm)
Mastacembellus pancalus	Mittal and Munshi (1971)	Skin (back)	34.0
Amphipnous cuchia	Mittal and Munshi (1971)	Skin (back)	19.0
Liobagrus mediadiposalis	Park (2003b)	Skin (back)	169 ± 50.9
Periophthalmus. magnuspinnatus	Park (2002)	Skin (back)	2.0
P. argentilineastus	Zhang <i>et al.</i> (2003)	Skin (dorsolateral body)	2.0
P. chrysospilos	Zhang et al. (2003)	Skin (dorsolateral body)	3.0
P. modestus	Park et al. (2000)	Skin (back)	1.4 ± 0.6
Boleophthalmus pectinirostris	Park et al. (2003c)	Skin (back)	9.3
Lepidocephalichthys guntea	Moitra et al. 1989	Intestine	2.6
Misgurnus mizolepis	Park et al. (2003a)	Intestine	0.7 ± 0.1
Misgurnus anguillicaudatus	Park and Kim (2001)	Intestine	11.2 ± 1.1

al., 2000, 2001; Park, 2002; Zhang et al., 2003). Among the observed Korean 6 species, 3 mudskippers, Boleophthalmus pectinirostris, Periophthalmus modestus and P. magnuspinnatus and a freshwater torrent catfish, Liobagrus mediadiposalis, showed similar modified skin structures as the above features.

Among them, the epidermis was thick due to component of the following cells: two kinds of glands as a small mucous cells and a large club cells in L. mediadiposalis, voluminous cells (swollen cells) swollen by epidermal cells and glandular mucous cells in Boleophthalmus pectinirostris, and only voluminous cells having no any glandular cells in Periophthalmus modestus and P. magnuspinnatus. In the cutanous respiratory fishes, two glandular cells in the epidermis were well known in Misgurnus fossilis,, M. anguillicaudatus, Heteropneustes fossilis, Mastacembelus pancalus, Amphipnous cuchia (Mittal and Munshi, 1971), and Monopterus albus (Liem, 1967), except for Monopteru albus having large unicellular mucous cells. Also, swollen cells in the epidermis are of characters in amphibious mudskippers, Periophthalmus and Boleophthalmus (Whitear, 1986; Al-Kadhomity and Hughes, 1988; Yokoya and Tamura, 1992; Zhang et al., 2000; Park et al., 2000; Park, 2002).

Of interesting features in cutaneous respiration, it is numerous blood capillaries in the epidermis. In many cutaneous respiratory fishes, intraepithelial blood capillaries were rare but reported in the epidermis of the adult mudskipper, some fish of genus Periophthalmus (Zang et al., 2003), P. koehlreuteri (Whitear, 1986), and the epithelial surface of the fins and the yolk sac in the larvae of swamp eel, Monopterus albus before the gills have developed (Liem, 1967). Through it is not intraepithelial blood capillaries, B. pectinirostris has dermal capillaries just beneath the stratum germinativum, which is the apex of the dermal bulges. The diffusion distance between the blood of the capillaries and the surface of the skin was the lowest, mean 1.4 µm in P. modestus, meaning that is situated at the most superficial region of the epidermis, whereas the highest, mean 34.0 μm, in *L. mediadiposalis*, situating at the middle region of the epidermis (Table 1).

Table 2. Diffusion distance by the regional epidermis in *Boleophthalmus pectinirostris*: N=number of determinations (Park *et al.*, 2003c)

Body regions	N	Diffusion distance near dermal bulge (μm)	
		Mean ±SD	Range
Interorbit	50	11.3±3.8	5.0~17.5
Top of the head	50	5.5 ± 1.2	$2.5 \sim 7.5$
Upper jaw	50	8.3 ± 2.9	$2.5 \sim 10.5$
Lower jaw	50	22.5 ± 4.2	$17.5 \sim 27.5$
Chin	50	6.3 ± 2.3	$2.5 \sim 12.5$
Operculum	50	14.3 ± 7.3	$5.0 \sim 20.0$
Dorsum	- 50	9.3 ± 2.8	$5.0 \sim 15.0$
Lateral region	50	14.0 ± 6.2	$7.5 \sim 20.2$
Ventral region	50	44.3 ± 4.4	40.0~50.0

The dermal bulges are appeared in Boleophthalmus among air-breathing fishes and have numerous blood vessels and capillaries in just beneath the stratum germinativum of the epidermis. From our results on 15 epidermal regions of B. pectinirostris, the value of the diffusion distance showed a trend, becoming lower in order of the top of head, the chin, the upper jaw, the dorsum, the interorbit, lateral region, and the operculum, ranging average 5.5 to 14.3 µm (Table 2). On the basis of the value by regions, it can surmised that regions toward the upper regions of the body exposed to the air showed lower values than the lower regions as the ventral region or the lower jaw. It means that the blood capillaries in the upper regions are closely situated to the epidermis to take up air from atmosphere. While in the water, they expose the head and a part of the dorsum above the water (Murdy, 1989; Kim and Park, 2002). Therefore, these differences of the diffusion distance by regions seem to be clearly related to their amphibious mode. However, all the fins and the sucking disc have no dermal bulge, and consequently blood capillaries did not exist. We can consider that air respiration by skin in B. pectinirostris is performed through skin covering the body region, not the appendage. In contrast, *Periophthalmus* has no dermal bulges, and they have intraepithelial blood capillaries in body regions as well as the appendage of all the fins and the sucking disc (Park et al., 2004).

In the observed 6 Korean fishes, another modified organ for air respiration is an intestinal respiratory tract. The mucosal epithelium of the intestine and rectum of M. mizolepis and M. anguillicaudatus has abundant blood capillaries. The distance between the blood of capillaries and the intestinal lumen was mean 11.2 um in M. anguillicaudatus and mean 0.7 µm in M. mizolepis. In addition to 2 Korean mud loaches, Lepidocephalichthys guntea was known as an intestinal respiratory epithelium with mean 2.6 µm of diffusion distance (Table 1). These capillaries serve in the gas exchange process between the blood and swallowed air (Moitra et al., 1989). The intestine and rectum have a respiratory epithelium in order to make up for the deficient oxygen supply, and gas exchange probably occurs between the air taken from the intestine and blood circulating at the intestinal mucosa. In physiological experiments on the intestinal respiration of M. anguillicaudatus, it has been reported that ingested food reached the straight intestine by movements of the stomach and was pushed out of the anus by air swallowed for intestinal respiration (Itazawa and Hanyu, 1991). In the observed fishes, the mucous cells found the skin and intestine were acidic sulphated mucosubstances or acidic sulfomucins in nature, which is characteristic of air breathing fishes (Mittal and Munshi, 1971; Mittal and Banerjee, 1974; Mittal et al., 1980; Park et al., 2000). Mittal et al. (1980) showed that mucus has a remarkable power of precipitating mud held in suspension in water and that the mucus in air breathing fishes may also be used in keeping the skin or epithelium clear for respiration. Lymphatic spaces containing small lymphocytes in the stratum germinativum layer of the epidermis was known as features of cutaneous respiration, functioning in the supply of nutrition to the stratum germinativum and to protect the epidermis from microorganisms or foreign proteins for protection of the epidermis (Mittal and Munshi, 1971). Also reduction or absence of the scale was found in a burrowing and a mud-dwelling fish (Amphipnous, Monopterus) and it was considered as an adaptation to its peculiar mode of life related to cutaneous respiration (Liem, 1967; Mittal and Munshi, 1971; Whitear, 1986). In the observed 6 Korean fishes, B. pectinirostris, P. modestus, P. magnuspinnatus, M. mizolepis and M. anguillicaudatus had small scales embedded in the superficial layer of the dermis, whereas L. mediadiposalis had no scale.

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어류의 공기호흡 기관

김치홍・박종영1

국립수산과학원 중앙내수면연구소, '전북대학교 자연과학대학 생명과학과 · 생물다양성연구소

요 약:한국의 담수어류 중 짱뚱어, 큰볏말뚝말둥어, 말뚝망둥어, 미꾸리, 미꾸라지, 자가사리 등 모두 6종에서 공기로 호흡하기 위해 변형된 부속기관이 알려져 있다. 짱뚱어, 큰볏말뚝말망둥어, 말뚝망둥어, 자가사리 4종은 부족한 산소를 공기로부터 흡입하기 위해 호흡상피(respiratory epithelium)를 갖는다. 이러한 호흡상피는 풍부한 모세혈관과 2종류의 선세포(점액세포와 곤봉세포-자가사리, 점액세포와 거대세포-짱뚱어) 또는 1종류의 선세포로 구성된 팽대세포(말뚝망둥어와 큰볏말뚝망둥어)를 갖는다. 특히 거대세포(swollen cell)는 상피세포의 팽대로 인해 형성된 거미줄 구조(web-like structure)를 갖고 있다. 또한 짱뚱어의 진피성돌기(dermal bulge)는 진피성 모세혈관이 매우 풍부하며, 지느러미 등의 부속지를 제외한 피부에 존재한다. 또한 미꾸리와 미꾸라지는 장 상피 (intestinal epithelium)에 모세혈관이 아주 풍부하게 존재한다.