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Osteocranium anatomy of African catfish (*Clarias gariepinus* Burchell 1822) from cultured pond in Aceh, Indonesia

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Abstract

To date, information related the skeletal morphology of fish in Indonesia is still limited. Therefore, we first described the morphology of the cranium of African catfish (Clarias gariepinus) collected from an aquaculture pond in Aceh Province, Indonesia. In the present study, research methodology included the preparation of cranium, photographing, editing images, and identifying the terminology of the cranium. The cranium was prepared physically and chemically. Each part of the cranium was documented using a Canon EOS 700D camera and edited using Adobe Photoshop CS6. The cranium nomenclature was determined by comparing the similarity of the shape and location of each part of the fish cranium that has been studied previously. The cranium of African catfish was divided into two major parts, namely neurocranium (ossa neurocranii) and splanchnocranium (ossa splanchnocranii). Neurocranium had four regions belonging to ethmoidal, orbital, otic, and occipital, while splanchnocranium had five regions belonging to maxillaris, mandibularis, arcus mandibularis, arcus hyoideus, and apparatus opercular. The African catfish had a solid and thick neurocranium structure. however, orbital, arcus hyoideus, and opercular apparatus regions were not well developed. The results of this study could be used as a basis for further research, especially in taxonomy and the phylogeny of fish.

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Introduction

African catfish (*Clarias gariepinus*, Burchell 1822) is one of the important cultured fish species extensively consumed by Indonesian people (1). However, to date, studies related to African catfish in Indonesia are still limited to fish feed formulations (2,3), aquaculture techniques (4), genetic improvement (5), and processed products (6). Meanwhile, studies underlying the morphology, especially the skeleton of African catfish, have not been well documented. Morphologically, African catfish have an elongated body shape, smooth skin, slimy, and not scaly (7). It belongs to the order Siluriformes and is one of the representatives of Actinopterygii. Actinopterygian fishes show hyostylic jaw suspension. The shape of the head is flat, with a relatively wide mouth and accompanied by four pairs

of tentacles. On the pectoral fin (pinna thoracalis), a pair of hard barbel or protrusion is used to defend themselves. At the top of the gill cavity, there is an arborescent organ, which has a shape resembling a branching tree (8). The growth and development of the skeleton play an essential role that affects fish's overall health (9). The skeleton of a fish generally contains three major parts, namely the cranium (ossa cranium), the backbone (ossa vertebrae), and the appendicular (ossa appendicular) (10). The cranium has an essential function as a protector of the brain and sensory organs in the head (11). The backbone acts as a biomechanical anchoring muscle and flexibility, while the appendicular skeleton has a primary function to support acceleration and stability (12,13). The elasticity of the skeleton also plays a role in the regulation of the homeostasis of phosphorus (14). The fish cranium may be divided into two main parts, namely neurocranium (ossa neurocranii) branchiocranium/splanchnocranium and (ossa branchiocranii/splanchnocranii) (15-17). However, the morphology of the cranium of each fish species is influenced by the individual's genetic, feeding behavior, and water quality (18,19). Burress (20) and Square et al. (21) stated that the mechanism of breathing and eating patterns are some of the significant factors that could influence the shape of the cranial bones of fish. For example, predatory fish tends to have a solid and robust jawbone structure that could exert a considerable bite pressure on their prey during the jawclosing process (22). In contrast, benthos-eating fish generally has a mouth at the ventral position, good lip elasticity, and a blunt jaw (23) this study aimed at describing the morphology of the cranium of the adult African catfish. The results of fish osteology could be used as a basis for further research, especially in the field of taxonomy and phylogeny.

Materials and methods

Specimen collection

A total of five specimens of male African catfish total weight: 4.26 ± 0.34 kg, total length: 78.16 ± 1.66 cm were obtained from the catfish culture pond in Lipah Rayek village, District of Bireuen, Province of Aceh (5°13'26.7"N 96°40'18.4"E). The stages of the study included specimen preparation, cranium preparation, photographing, image editing, and identification of the nomenclature of the cranium. All stages of the research were carried out at the Mathematics and Natural Sciences Laboratory, Universitas Almuslim, Indonesia.

Cranium preparation

The preparation of cranium consisted of two stages: physical and chemical, according to the protocol described in Akmal *et al.* (15). The physical stages were started with exfoliation of the skin, and head muscles were attached to

the bone with hot water 80-90 °C. The skin and connective tissue that blister and blusterous were then carefully cleaned using a scalpel. The cranium was further separated into two parts, namely neurocranium (*ossa neurocranii*) and splanchnocranium (*ossa splanchnocranii*). All the bones that have been released were cleaned using a soft brush and tweezers. Chemical stages were started with soaking the cranium into 10% formalin solution for seven days and 100% ethanol solution for 24 hours. The preserved cranium was cleansed again using a smooth brush to remove the remaining fat attached to the bones. The cranium was dried in the sun for seven days and subsequently coated with a clear transparent paint spray and dried again for three days (15).

Image processing and cranium nomenclature

Every cranium that separated from the others was fixed into its original positions using a glue stick and superglue adhesive. Each part of the cranium was photographed using a Canon EOS 700D camera, while the image was edited using Adobe Photoshop CS6 version 13.0.1. Each part of the cranium was named by comparing the similarity of the shape and location of each part with the bone morphology of several fish species that Rojo had previously describedRojo (10), Lundberg *et al.* (24), and Birindelli (25). All observations were analyzed descriptively and presented in tables and figures.

Results

Neurocranium (ossa neurocranii)

The neurocranium of African catfish was divided into four regions, namely the ethmoidal, orbital, oticum, and occipital regions. These four regions were located in the dorsal portion of the skull bones. The Ossa neurocranium of African catfish had a sturdy structure with solid joints and thorn-like protrusions on the dorsal part.

The ethmoidal (olfactory) region was located in the dorsal anterior portion of the neurocranium. This region was composed of four investing and replacing bones, namely mesethmoidale, os ethmoideum lateral, os vemorale, and os nasale (Figure 1). The orbital region was located in the central and ventral parts of the neurocranium. This region was composed of 10 investing and replacing bones, namely os frontale, os orbitosphenoidale, os pterosphenoidale, os parasphenoidale, os suprasphenoidale, os lacrimal, and os infraorbital II-IV (Figure 1). Os frontale formed the roof of the skull while os lacrimal formed the anterior part of the orbit. The otic region was located on both lateral sides of the neurocranium, attached to the opercular apparatus, and the hyomandibulare os connected with the jaw joint. This region was composed of five bones, namely os sphenoticum, os pteroticum, os prooticum, os epioticum, os posttemporale (Figure 1). Os practicum was articulated with parasphenoid anteriorly, sphenoticum and pteroticum laterally, and exoccipitale posteriorly. Os epioticum was attached with the os exoccipital, supraoccipital, and pteroticum through its ventral region. Os pteroticum showed articulation with os sphenoticum anteriorly, os supraoccipital dorsally, and os prooticum and os exoccipitale ventrally.



Figure 1: Dorsal view of the cranium of African catfish. Epi: os epioticum; Ethl: os ethmoidal lateralis; Fro: os frontale; Inf: ossa infraobitale; Lac: os lacrimal; Max: os maxillare; Mes: mesethmoidale; Nas: os nasale; Post: os posttemporale; Pre: os premaxillare; Pte: os pteroticum; Soc: os supraoccipitale; Sph: os sphenoticum. Scale bar: 1 cm.

Os sphenoticum was attached with os pteroticum posteriorly, os supraoccipitale mediolaterally, os frontale anterolaterally, and os prooticum ventrally. The lateroventral part of the oticum region was more developed than other parts, and it was directly related to the respiratory accessory organ. The occipital region was located in the posterior neurocranium, directly connected to the columna vertebrae (spine). This region was composed of five bones, namely os supraoccipitale, os exoccipitale, os epioccipitale, os basioccipitale and os extrascapulare (Figures 2). Os supraoccipitale formed the roof of the cranium. Os supraoccipitale was articulated with os frontale anteriorly, os exoccipitale posteriorly, and os pteroticum and os epioticum laterally. Os basioccipitale united in front with the parasphenoid and laterally with os exoccipital and os prooticum.

Splanchnocranium (ossa splanchnocranii)

The splanchnocranium was divided into five regions, namely maxillary, mandibular, mandibular arcus, arcus hyoideus, and opercular apparatus. The maxillaries region was located anteriorly from os mesethmoidale, which protrudes to the lateral side. This region comprises two bones, namely os maxillare and praemaxillare (Figure 3). The bone-forming maxillary region tended to have rigid structures and did not have sturdy joints. The mandibular region was located in the anterior part of the splanchnocranium. This region comprises four bones: os dental, os anguloarticulare, coronomeckeli, and retroarticular (Figure 3). Os praemaxillare was articulated in front with the same outer side and terminated before the articulation of the mandible with the os quadratum behind.



Figure 2: Lateral view of the cranium of African catfish. Ang: os anguloarticulare; Aut: os autopalatinum; Cer: os ceratohyale; Crm: os coronomeckeli; Den: os dentale; Epi: os epioticum; Ethl: os ethmoidale lateralis; Fro: os frontale; Hyo: os hyomandibulare; Inf: ossa infraobitale; Int: os interoperculare; Lac: os lacrimale; Max: os maxillare; Mes: os mesethmoidale; Met: os metapterygideum; Ope: os operculare; Post: os posttemporale; Pre: os premaxillare; Preo: os preoperculare; Pte: os pteroticum; Qua: os quadratum; Rbra: Radii branchiostegii; Sph: os sphenoticum. Scale bar: 1 cm.

The mandibular arcus region was linked directly to the posteroventral portion of the mandibular region and the anterodorsal portion of the ethmoidal region. This region was composed of five bones that support the lower jaw, namely os aupalatinum, os metapterygoideum, os quadratum, os hyomandibulare, and os preoperculum (Figure 3). The inner half of os metapterygoideum was united posteriorly with the os hyomandibulare, and the outer half was continued behind to meet os quadratum and os hyomandibulare. Os quadratum was united anteriorly with os metapterygoideum, mediolaterally with os hyomandibulare, and posteriorly with os preoperculum. The mandibular arcus region tended to have relatively more minor bones compared to other ossa cranium bones.

The hyoideus arcus region was a collection of bones that covered the ventral part of the gills. This region composed four bones: the os hypohyalia, os ceratohyale anterior, os ceratohyale posterior, and radii branchiostegii. The hyoideus arcus region had a round, long, thick shape with a sharp angle in the anterior. The opercular apparatus region was the gill cover bone located in the lateral ossa cranium. The opercular apparatus region was composed of two bones: the os interoperculum and operculum (Figure 2). This region was linked directly to the dorsomedial part of the hyomandibulare.



Figure 3. Ventral view of the cranium of African catfish. Bac: os basioccipital; Can: os ceratohyale anterior; Cpo: os ceratohyale posterior; Crm: os coronomeckeli; Den: os dentale; Epc: os epioccipitale; Epi: os epioticum; Exc: os exoccipital; Exs: os extrascapulare; Hyp: os hypohyalia; Obs: os orbitosphenoidale; Pre: os premaxillare; Pro: os prooticum; Psp: os parasphenoidale; Pto: os pteroticum; Pts: os pterosphenoidale; Sph: os suprasphenoidale; Rbr: Radii branchiostegii; Vom: os vemorale. Scale bar: 1 cm

Discussion

African catfish osteocranium was divided into two main parts, neurocranium (*ossa neurocranii*) and splanchnocranium (*ossa splanchnocranii*). This result is identical to several fish species such as *Platyclarias machadoi* (Clariidae) (26), *Tor tambroides* (Cyprinidae) (12), and *Kryptoglanis shajii* (Kryptoglanidae) (24). According to Löffler *et al.* (27), the neurocranium has the primary function as the protector of the brain and other sensory organs, while splanchnocranium is often linked to helping the digestive and respiratory functions of fish.

The neurocranium of the African catfish was composed of bones with a strong structure, thick and equipped with thorn-like protrusions on the dorsal part. The shape of the neurocranium of African catfish is similar to fish from the Pimelodidae family, such as *Phractocephalus hemioliopterus* (28). However, there is a difference with the Cyprinidae family (15,29), Nemacheilidae family (30), and Cichlidae family (31) in the sense that fish from this family tend to have a non-rigid and thin with a smooth surface.

The morphological variation of the neurocranium of African catfish compared to another family might be closely related to the various characteristics of the habitat of fish. Pérez and Fabré (32) revealed that the morphological differences between the neurocranium and otoliths of fish emerged as an adaptive (evolutionary) response to various environmental conditions (selective pressure) encountered. Catfish spends most of its life inhabiting the bottom layer of the waters. Furthermore, the head of catfish tended to receive intense water pressure for an extended period. Thus, catfish require a sturdy and thick neurocranium structure accompanied by small bumps on the dorsal part to reduce water pressure and mud.

The ethmoidal region has a kinematic role in sustaining the opening of the maxilla (33). Moreover, this region acts as a chemosensory directly related to the olfactory system (26). Os mesetmoidale in African catfish seems to be more developed than fish from the family Cyprinidae and Cichlidae, characterized by a more significant shape relatively and experiences anterior stretching (15,29,31). This was thought to be influenced by African catfish, which rely on the olfactory system in finding food. Sarkar & De (34) reported that fish forage by relying on the olfactory system has the sensory cells of the olfactory that are more developed than other fish, so a greater ethmoidal region is needed to support the performance of the olfactory system.

The orbital and otic regions play a role in supporting the hearing and vision system in African catfish. African catfish had the morphology of the orbital and oticum regions similar to *Ameiurus nebulosus* from the family Ictaluridae and *Bathycetopsis oliveira* from the Cetopsidae family (10). However, the orbital regions of catfish tend not to develop when compared to fish from the Cyprinidae family, such as the *Tor tambroides. Tor tambroides* is reported to have an orbital region with a broader os lacrimal shape and is equipped with a V os infraorbital (15).

Os lacrimal and os infraorbital are the bones that support the eyeball (10). African catfish are known to have environmental conditions with high turbidity levels. Besides, African catfish tend to be actively foraged at night (nocturnal). This caused the African catfish's vision system to be less developed than other sensory systems. Schmitz & Wainwright (35) also stated that fish that foraged by not relying on the organ of vision generally had bones that makeup-less developed orbital regions.

The shape of splanchnocranium (*ossa splanchnocranii*) has a relationship with the feeding behavior and the respiratory system of fish (10). There were three regions of splanchnocranium directly related to African catfish feeding behavior: the maxillary region, the mandibular region, and the mandibular arcus region. Catfish tend to be carnivorous, so they require strong jaws with large mouth openings. During the mouth opening process, these three regions form a straight line to enlarge cavum buchalis. Thus, the process of sucking food could take place optimally.

The arcus hyoideus region and the opercular apparatus play a role in supporting the respiratory system of fish (36). African catfish have a different respiratory organ called the arborescent organ. This organ is derived from modifying the second to fourth-gill arch to take oxygen from water and air (37). The presence of arborescent organs causes the African catfish's arcus hyoideus region and the opercular apparatus to become undeveloped compared to other fish with other respiratory organs. The African catfish, the opercular apparatus region, has two bones, namely os interoperculum and os operculum, whereas, in *Tor tambroides* fish (Cyprinidae), this region was composed of five bones: os preoperculum, os interoperculum, os suboperculum, and os operculum (15).

Conclusion

The African catfish neurocranium is divided into four regions: ethmoidal, orbital, oticum, and occipital regions, while the splanchnocranium is divided into five regions, namely the maxillary, mandibular, arcus mandibular, arcus hyoideus, and opercular apparatus. African catfish have a solid and thick neurocranium structure. However, orbital, arcus hyoideus, and apparatus opercular regions are not well developed. We recommend further research to understand the relationship between cranium morphology and the respiratory system, feeding behavior of African catfish, and adaption to environmental factors.

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Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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تشريح جمجمة اسماك القرموط الأفريقي في أحواض تربية الأحياء المائية في آتشيه، أندونيسيا

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الخلاصة

لحد الآن، لا تزال المعلومات المتعلقة بشكل الهيكل العظمي للأسماك في إندو نيسيا محدودة. لذلك، وصفنا أو لاً شكل جمجمة اسماكَ القر موط الأفريقي (Clarias gariepinus) الذي تم جمعه من أحواض تربية الأحياء المائية في مقاطعة آتشيه بإندونيسيا. تضمنت منهجية البحث في هذه الدراسة إعداد الجمجمة، والتصوير، وتحرير الصور، والتعريف بمصطلحات الجمجمة. تم تحضير الجمجمة فيزيائيًا وكيميائيًا. تم توثيق كل جزء من الجمجمة باستخدام كاميرا Canon EOS 700D وتحريره باستخدام .Adobe Photoshop CS6 تم تحديد تسمية الجمجمة من خلال مقارنة التشابه في شكل وموقع كل جزء من جمجمة الأسماك التي تمت دراستها مسبقًا. قسمت جمجمة اسماك القرموط الأفريقي إلى قسمين رئيسيين، هما القحف العصبي العظمي (ossa neurocranii) والقحف الحشوي العظمى(ossa splanchnocranii). تضمن القحف العصبي العظمي أربع مناطق تنتمي إلى المصفاوي والحجاجي والسمعي والقذالي، بينما تضمن القحف الحشوي خمس مناطق تنتمى إلى الفك العلوي، والفك السفلي، وقوس الفك السفلي، والقوس اللامي، والجهاز الغطائي. تمتلك اسماك القرموط الأفريقي بنية قحف عصبي عظمي صلبة وسميكة. ومع ذلك، لم تكن مناطق الحجاجي، والقوس اللامي، والجهاز الغطائي متطورة بشكل جيد. يمكن استخدام نتائج هذه الدر أسة كأساس لمزيد من البحث، خاصة في تصنيف وتطور الأسماك.