

Comparative Scanning Electron Microscopy Study on Scale Variations in Indonesian Cultivated Koi Fish (*Cyprinus rubrofuscus* Lacepede, 1803)

Krisna Noli Andrian¹, Hevi Wihadmadyatami², Nastiti Wijayanti³,
Srikanth Karnati⁴, Aris Haryanto^{1*}

¹Department of Biochemistry and Molecular Biology, Faculty of Veterinary Medicine, Universitas Gadjah Mada, Yogyakarta, Indonesia, ²Department of Anatomy, Faculty of Veterinary Medicine, Universitas Gadjah Mada, Yogyakarta, Indonesia, ³Department of Animal Physiology, Faculty of Biology, Universitas Gadjah Mada, Yogyakarta, Indonesia, ⁴Institute of Anatomy and Cell Biology, Julius-Maximilians-Universität of Würzburg, Würzburg, Germany.

*Corresponding author: arisharyanto@ugm.ac.id

Abstract

Koi (*Cyprinus rubrofuscus* Lacepede, 1803) is a highly favored ornamental fish due to its beauty and wide range of variations in Indonesia, categorized by color, patterns, and scales. Some variants are distinguished by color, while others, such as Ginrin, Doitsu, and Shusui, have unique scale types. Despite visible differences, microscopic scale variations remain unexplored. SEM studies in other fish species offer insights into scale ultrastructure, providing opportunities for comparison. Therefore, this study aimed to uncover the microscopic structure of four types of koi fish, namely Doitsu, Ginrin, Shusui, and common scale. Koi fish were obtained from breeders in Yogyakarta, Indonesia, and acclimatized in an aquarium. Scale extraction was conducted under anesthesia using MS-222 and the cleaned scales were then subjected to dehydration, fixation, and affixed to double-sided adhesive tape for SEM analysis. Furthermore, coating with conductive gold enabled observation using SEM at 10 kV, allowing examination of scale features such as focus, radii, circuli, and lepidonts at various magnifications. The results showed that based on SEM analysis, significant differences were observed in scale structures among koi variants. At low magnification, differences in tubercles and lepidonts were observed, particularly between common and Ginrin scale types. Shusui scale showed unique characteristics with a closer arrangement of circuli and distinctive lepidont shapes. At higher magnification, clearer details of radii, circuli, and lepidonts were observed, further highlighting the differences among koi variants. SEM provides crucial insights into the morphology of scales in koi fish variants, showing unseen macroscopic differences and distinct features such as tubercles and lepidont frequency.

Keywords: Doitsu, Ginrin, Koi fish, scanning electron microscopy, Shusui

Received: November 26, 2024

Revised: March 3, 2025

Accepted: April 12, 2025

INTRODUCTION

Koi (*Cyprinus rubrofuscus* Lacepede, 1803) is one of the most highly favored ornamental fish cultured in Indonesia known for the beautiful colors and patterns, which provide a calming feeling for enthusiasts. Furthermore, koi have many variations categorized based on color, specific patterns in markings, and scales (de Kock and Gomelsky, 2015; Andrian *et al.*, 2024). Some popular koi varieties in Indonesia, such as Kohaku, Sanke, Showa, and Shiro (Andrian *et al.*, 2023; Aysi *et al.*, 2022), are frequently cultivated and distinguished by color patterns. Other variants are distinguished by scale, such as the

sparkling scaled variant known as Ginrin, the scaleless type called Doitsu, chain-scaled types called Matsuba variant, and large-scaled types on the dorsal part called Shusui. With the numerous scale types, people also differentiate variants based on scale when the color pattern is the same. These differences eventually lead breeders and enthusiasts to give names based on their scales, such as Kohaku Doitsu type, Kohaku Ginrin type, or just Kohaku type if they have a common scale. The structure of fish scales plays a crucial role in various physiological functions, including protection (Ghods *et al.*, 2020; Murcia *et al.*, 2017), locomotion (Vernerey and Barthelat, 2010), and sensory perception (Scott *et al.*, 2023).

Understanding the morphology of koi scales is not only of academic interest but also holds practical significance for koi breeders and hobbyists. SEM analysis of scale structures provides valuable insights into the physical characteristics, surface patterns, and structural adaptations of koi fish. Investigating these variations helps to clarify the evolution of scale morphology and how different structural traits may influence hydrodynamics, protection, and overall function. This study enhances the current understanding of koi fish diversity, contributing to evolutionary biology. For koi breeders and hobbyists, scale traits are not only an aesthetic feature but also define the beauty, health, and value of the fish. Understanding scale morphology helps breeders make informed decisions, ensuring koi with strong, desirable traits while minimizing potential health issues.

Although the differences in scales can be observed visually at a macroscopic level, microscopic differences have not been identified to date. Scale visualization using Scanning Electron Microscopy (SEM) has been conducted in other fish species such as *Rutilus frisii* (Esmaeili and Gholami, 2011), *Channa* fish (Dey *et al.*, 2014; Dey *et al.*, 2015), *Neolissochilus hexagonolepis* and *Neolissochilus hexastichus* (Raffealla and Bhuyan, 2020), *Notopterus kapingratensis* and *Epiplatys suratensis* (Ansari *et al.*, 2021), and in elasmoid fish (*Garra shama*) (Al Jufaili *et al.*, 2023). SEM has offered valuable insights into fish scale ultrastructure, allowing detailed comparative analysis. Through such comparisons, both similarities and differences among the scales of different fish species can be identified. This can also be performed with koi fish, which have several types of scales in different variants. The use of SEM is expected to provide insights into the differences and similarities among scale variants in koi fish.

The differences in scales on koi fish are visible macroscopically, but to date, there have been no reports on their microscopic structure. Reporting on the microscopic structure using SEM in koi fish will provide new insights into the characteristics of each variant of koi fish scales. This study aimed to uncover the microscopic

structure of four types of koi fish: Doitsu, Ginrin, Shusui, and the scales commonly found on koi fish. Information on the microscopic characteristics of different koi scale variants can provide valuable additional insights into genetic diversity and evolutionary history.

MATERIALS AND METHODS

Ethical Approval

This experiment was carried out with ethical approval from the laboratory animals use research ethics committee of the Faculty of Veterinary Medicine, Universitas Gadjah Mada, Indonesia with reference No.043/EC-FKH/Int/2022.

Study Period and Location

This study was conducted in October–December 2023 at the Faculty of Veterinary Medicine, Universitas Gadjah Mada, and the University Central Laboratory (LPPT UGM).

Scale Extraction

Koi fish samples consisted of one individual from each of the four variants namely Shiro Doitsu, Shusui, Sanke Ginrin, and Sanke variants, as shown in Figure 1. Fish were obtained from koi fish breeders in the Yogyakarta area, Indonesia. Fish measuring 10–20 cm were used to ensure that scales were fully developed. Subsequently, acclimatization was carried out in a maintenance aquarium for one week, and feeding was carried out *ad libitum*. Scale extraction was performed after anesthesia using MS-222 (Sigma Aldrich, Missouri, USA) at a dosage of 50 mg/L of water, by placing fish in an anesthesia tank until they lost balance. Scales were extracted using anatomical forceps by pulling them in the direction of scale growth. Scales were collected based on the desired type: dorsal scale for the Shusui variant, which is unique in having specialized scale that appears only along the dorsal line; lateral scale for Ginrin variant; and lateral scale for common Sanke type. As for Doitsu variant, a 1×1 cm section of its lateral skin was excised using a scalpel blade while fish was under anesthesia. Scales were then washed with distilled water and potassium hydroxide (H₂O₂) (Merck-KgaA,

Darmstadt, Germany) until clean. After scale extraction, fish were immersed in water containing Sodium Chloride (NaCl) (Otsuka Indonesia, Jakarta, Indonesia) at a concentration of 0.2 g/L for 5 minutes before being returned to the maintenance aquarium.

Scanning Electron Microscope

The cleaned koi fish scales were dehydrated in a series of ethanol (Merck-KgaA, Darmstadt, Germany) concentrations, starting from 70%, then 80%, 90%, and finally 100%. This was followed by immersion in 4% glutaraldehyde (Merck-KgaA, Darmstadt, Germany) as a fixative for two days. Subsequently, scale were dried and affixed to double-sided adhesive tape, with the dorsal side facing outward while the ventral side adhered to the tape. The conductive gold coating was applied to the fish scales. The coated fish scales were then observed using a JSM 6510 LA scanning electron microscope (SEM) (JEOL, Tokyo, Japan) in secondary electron emission mode at an accelerating voltage of 10 kV. Scale examination was conducted at various magnifications to observe the Focus, Radii, Circuli, and Lepidont features of the scales.

Data Analysis

The results of this study are presented descriptively in the form of tables and figures.

RESULTS AND DISCUSSION

The Macroscopic Form

Koi fish used in this study were examined based on their scales, namely the common scale variant found in koi fish with a cycloid shape and a uniform color corresponding to koi fish variant (Sanke), Ginrin scale variant characterized by a shiny appearance on the fish scales, Shusui variant characterized by a symmetrical pattern of scales on its dorsal part while the rest is scaleless, and Doitsu variant characterized by the absence of scales entirely on the fish. As shown in Figure 1, the differences between koi variants used are clearly visible macroscopically, allowing enthusiasts to easily identify koi variant. The scales seen in Figure 2 exhibit a shape that is not

significantly different between the common scale variant found in koi fish and the scales of Ginrin variant, both being cycloid in shape. The difference is only visually apparent in Ginrin scales, which are shinier compared to the scales commonly found in fish. On the other hand, the scales of Shusui variant appear to have a shape resembling elongated wings found on dorsal fin of the fish. This difference in scale shape indicates that Shusui variant has unique characteristics in its scales because only the dorsal scales appear, while the rest are scaleless or Doitsu. In Doitsu fish type, there are no scales at all on the fish's body, giving the color pattern directly from skin as shown in Figure 2d. From this macroscopic appearance, the differences between koi fish variants are clearly visible, such as in shape, color, and the presence or absence of scales on the fish.

Koi fish are ornamental fish that currently have a very large number of variants, reaching up to 120 variants (de Kock and Gomelsky, 2015). The abundance of these variants is due to the many developments over several decades. One of the distinguishing factors among these variants is seen in the scales of koi fish. Although koi fish originally came from three types of variants, they have developed distinct characteristics up to the present day. This development is also evident in the variation of scales in koi fish, as reported in this study. Koi fish are part of the Cyprinidae family, which generally have cycloid-shaped scales like other cyprinid fish (Al Jufaili *et al.*, 2023; Esmaeili and Gholami, 2011; Raffealla and Bhuyan, 2020). Most koi fish variants have cycloid-shaped scales with colors varying depending on the variant, as seen in this study with the common scales in the Sanke variant, which were taken from black-colored areas on the fish's body, showing black-colored scales on its posterior field (Suciyono *et al.*, 2023). However, there are also Ginrin variants in koi fish, characterized by their shiny scales. The emergence of Ginrin variants originate from crossing non-Ginrin variants with Ginrin variants or among Ginrin variants, thus being inherited by their progeny (Gomelsky *et al.* 2015). Therefore, as seen in this study, despite having the same

variant type, such as Sanke with white, red, and black color patterns, the Ginrin variant has shiny scales, thus being called Sanke Ginrin. This demonstrates that koi fish have numerous variants due to various distinguishing criteria, despite being closely related (Andrian *et al.*, 2023). On the other hand, the Shusui variant is unique because it only has scales on its dorsal fin with a symmetrical shape, while other parts of the body have no scales. Meanwhile, in the Doitsu variant, the entire fish body lacks scales. The differences in scale structures in fish can be influenced by the environment in which the fish live, serving as one of the fish's defense mechanisms (Gu *et al.*, 2023). The structural variations can also impact physiological functions, such as osmoregulation,

hydrodynamics, and overall skin protection, affecting the fish's adaptability and survival in different habitats (Clark and Uyeno, 2024; Ujjaina and Jaiswar, 2024). It is also explained that koi fish with Ginrin scales tend to have a shorter lifespan compared to non-Ginrin types (Gomelsky *et al.*, 2015). This may also be influenced by the fact that Ginrin types more easily reflect light, making them more visible in nature. The uniqueness of each koi fish variant, although originating from the same ancestors, has developed into highly varied forms. This uniqueness influences the choices of enthusiasts and breeders regarding preferences for the variants to cultivate.

Table 1. Summary of differences in scale morphology among koi fish variants

Feature	Common Scale	Ginrin Scale	Shusui Scale	Doitsu Variant
Macroscopic Scale Shape	Cycloid	Cycloid	Elongated wings on dorsal fin	Scaleless
Color	Variant-dependent	Shiny, variant-dependent	Black symmetrical dorsal scales, orange and white in scaleless part of the body	Variant-dependent
Presence of Scales	Entire body	Entire body	Only on dorsal fin	No scales
Tubercles	Present in posterior field	Absent	Present in posterior field	N/A
Circuli	Rounded from the focus	Rounded from the focus	Closer together, elongated	N/A
Radii	Extending to anterior field	Extending to anterior field	Extending to posterior field	N/A
Lepidonts	Thin, elongated	Short, pointed	Thick, sharp, and dense	N/A
Reflectivity	Normal	High	Normal	Normal
Scaleless Area	None	None	Majority of the body	Entire body

SEM Analysis of Koi Fish Scales Reveals Structural Differences

The results of SEM at low magnification, as shown in Figure 3, show several important positions of scale structure such as focus, circuli, radii, anterior field, posterior field, lateral field, and tubercles on the scales. The focus is the

central part of fish scale where the initial growth occurs, which then grows away from the focus area. Circuli are growth patterns of scale, mostly circular with the center at the focus. Radii are radial lines extending outward from the focus toward the edge of scale. The anterior field is the front part of scale, the posterior is the back or tail

part of scale, and the lateral field is the side part of scale. Each part sometimes has shapes or characteristics that distinguish the fish. Tubercles are protrusions on scale, usually found on the posterior field. At this magnification, differences between the scales commonly found on koi fish in Figure 3a and Ginrin scale in Figure 3b are apparent, namely in the presence of tubercles in the posterior field of common scale, which are absent in Ginrin scale, even though they have relatively similar macroscopic shapes, i.e., cycloid, and no differences are visible unless examined directly on the posterior part of the scale. However, other parts of both types of scales have similarities, such as the focus located in the middle of the scale, circuli that tend to be rounded from the focus, and radii that lead towards the anterior field. On the other hand, on Shusui-type

scales shown in Figure 3c, the shape of the scale appears to be unique and has tubercles on its posterior field. The focus is on the middle of the scale, with radii leading towards the posterior field. The circuli of this scale type grow away from the focus, located between its radii. The significant differences in Shusui koi scales indicate that this variant has its own distinctive characteristics. In Figure 3d, the skin of Doitsu koi variant is visible, indicating that the fish does not have scales at all on its entire body, thus revealing the skin structure of the fish. With SEM at low magnification, clear differences are visible between the four fish variants, and the differences become apparent after SEM examination, especially between common type and Ginrin type, which have almost the same macroscopic shape but are different microscopically.

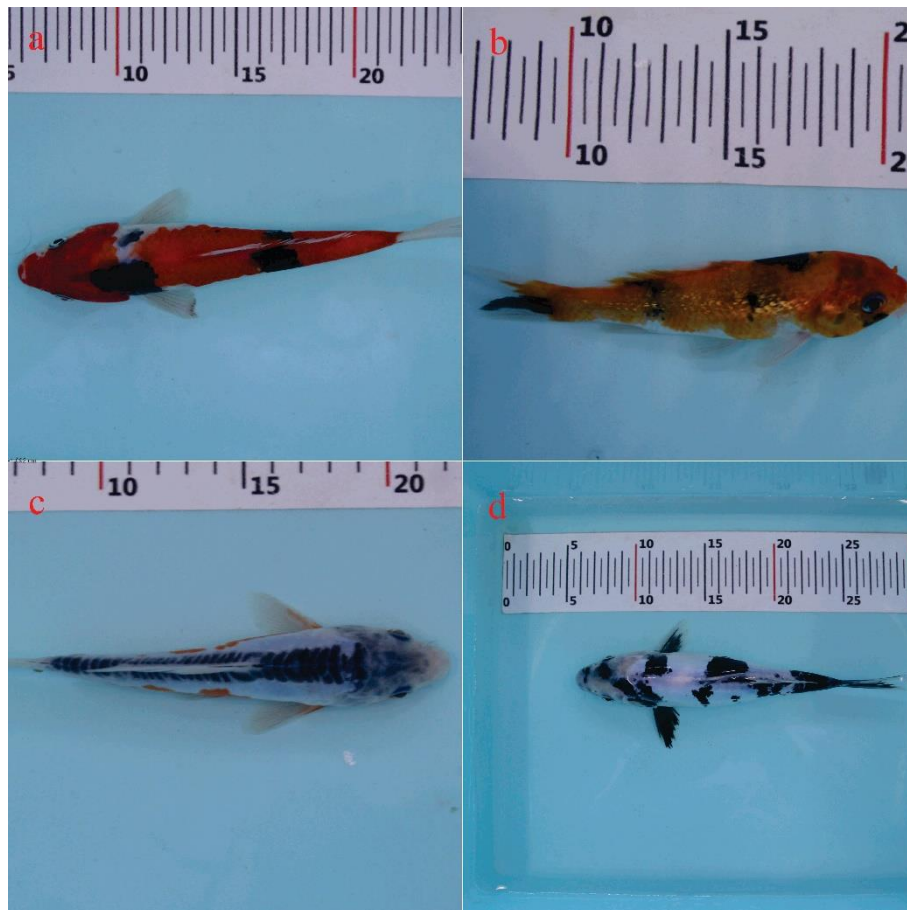


Figure 1. The koi fish variants used in this study consist of (a) Sanke with common scale, (b) Ginrin Sanke, (c) Shusui, (d) Shiro Doitsu.

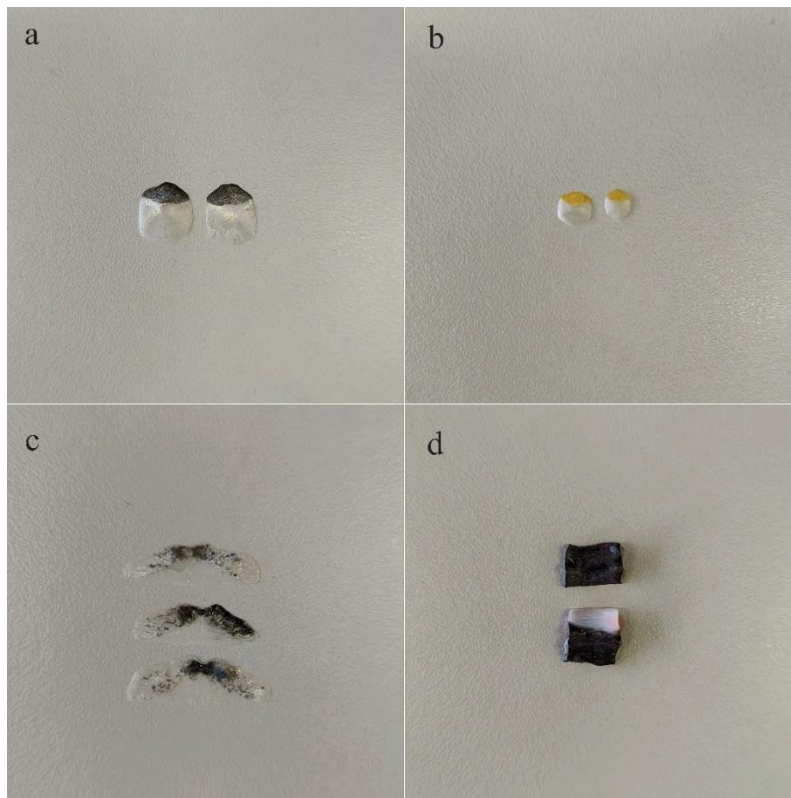


Figure 2. Detailed view of koi fish scales. (a) Common scale variant, cycloid in shape, (b) Ginrin scale variant, cycloid and shiny, (c) Shusui variant, scales resembling elongated wings on the dorsal fin, (d) Doitsu variant, completely scaleless.

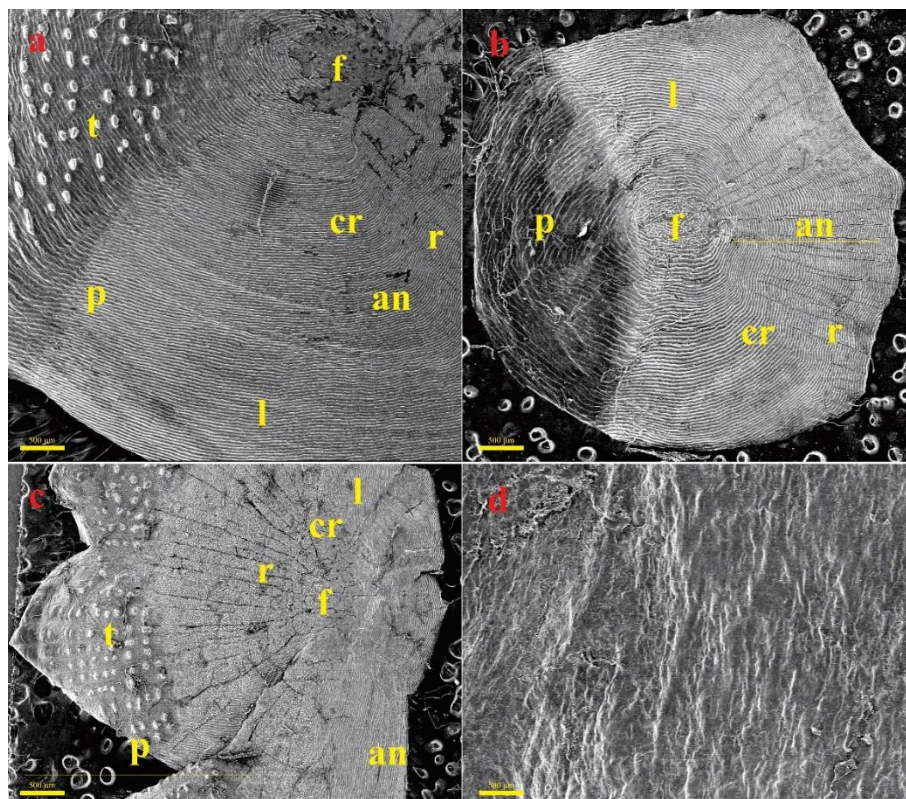


Figure 3. Low SEM magnification. (a) Sanke with common scale, (b) Ginrin Sanke scale, (c) Shusui dorsal scale, (d) Shiro Doitsu skin, (an) anterior field, (cr) circuli, (f) focus, (l) lateral field, (p) posterior field, (r) radii, (t) tubercles.

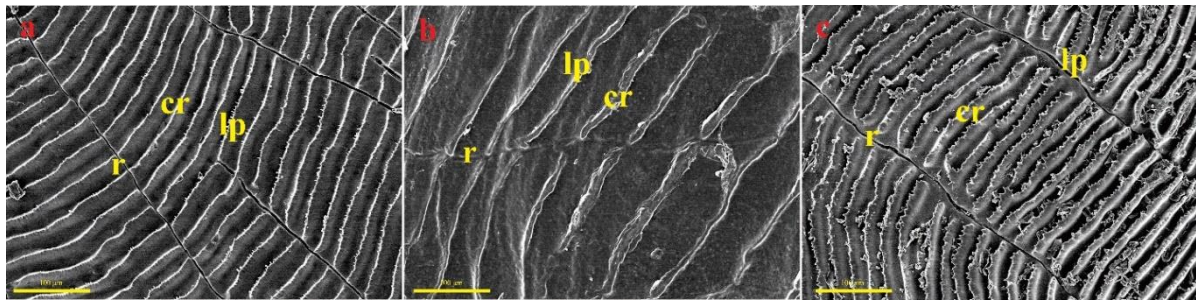


Figure 4. Circuli and radii structure. (a) Sanke with common scale, (b) Ginrin Sanke scale, (c) Shusui dorsal scale, (cr) circuli, (lp) lepidont, (r) radii.

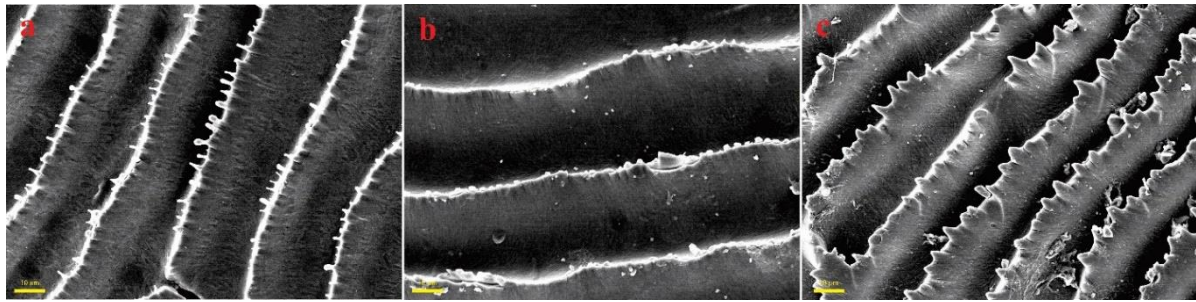


Figure 5. Lepidonts structure. (a) Sanke with common scale, (b) Ginrin Sanke scale, (c) Shusui dorsal scale.

The use of SEM in characterizing fish scale morphology provides a deeper insight into the characteristics of fish. Macroscopic differences that are not visible can be easily observed with SEM, as shown in this study. A notable difference is seen between common scale type and Ginrin scale type, which have almost the same macroscopic shape, but after imaging using SEM, differences can be observed due to the presence of tubercles on common scales and the absence of tubercles on Ginrin scales. Additionally, lepidonts on Ginrin scale are generally less common compared to the common type, which has many lepidonts. Another clear difference is observed in Shusui type, which has a unique shape compared to typical cycloid-shaped koi fish scales. Ultrastructurally, clear differences are also visible, especially in the circuli and lepidonts. The distance between circuli in Shusui type is closer compared to the common and Ginrin variants. The spacing between circuli indicates the growth phase of the scales, with closer spacing indicating faster scale formation. Lepidonts in Shusui variant also have distinctive sharp and dense shapes compared to the common variant, which tends to be thin but long. In other cyprinids such as *Neolissochilus hexagonolepis* and

Neolissochilus hexastichus (Raffealla and Bhuyan 2020), SEM can be used as a tool to compare the characteristics of scales between different species. When viewed using SEM, the structure of koi fish scales varies, but generally they still have the same structural composition as other cyprinids, such as having almost straight interradii circuli (Esmaili and Gholami 2011; Novindasari *et al.*, 2024). However, other differences with other cyprinids indicate variations between species. Even among koi fish variants, many differences in scale ultrastructure are found. Comparing the ultrastructure of koi fish scales shows that SEM can be used for characterizing scale morphology in koi fish and yields good results. Future studies can combine anatomical studies like this with molecular tests to determine if there are indeed significant differences between these variants. On the other hand, there are still many parts of koi fish that can be imaged using SEM to understand their ultrastructure and compare them between koi fish variants or with other cyprinids.

SEM Analysis Reveals Distinctive Radii, Circuli, and Lepidont Structures in Various Koi Fish Scale Variants

The structure of fish scales using higher magnification in Figure 4 shows clearer radii and circuli structures, and lepidonts are also visible on the circuli. In the common scale variant in Figure 4a, the circuli are more distinct compared to the circuli in Ginrin variant in Figure 4b, with almost the same circular shape moving away from the focus. In Shusui scale variant, the circuli appear closer together, with growth direction also moving away from the focus. All three types of scales also show clear radii in common and Shusui scales, while they are not very distinct in Ginrin type at this magnification. Lepidonts on common and Shusui scales are visible at this magnification, while they are not very visible on Ginrin scales. Lepidonts on the Shusui variant are very clear at the tip of the dorsal circuli, indicating significant differences. At higher magnification as shown in Figure 5, the lepidonts of each variant are clearly visible. Lepidonts on the common scale variant are thin and elongated, with many at each end of the circuli. In the Ginrin scale variant, lepidonts tend to be short and pointed, appearing less frequently than in the common type. In Shusui type, lepidonts appear thicker, sharper, and are found more frequently at the ends of the circuli. Additionally, at this magnification, the distance between circuli in Shusui variant appears to be closer compared to common and Ginrin variants. The differences in shape and number of lepidonts and circuli in each type of scale indicate significant differences among these variants.

The diversity of lepidonts is a critical aspect of ichthyological studies, providing valuable insights into taxonomic and phylogenetic relationships among fish species. Lepidonts, which are small denticles on fish scales, exhibit notable morphological variation influenced by species, body region, and developmental stage. This variation plays a key role in species identification and classification. Studies on other fish species have demonstrated how lepidont morphology differs significantly across taxa. For example, in Mullidae, lepidonts range from blunt to pointed forms (Echreshavi *et al.*, 2021;

Dhamayanti *et al.*, 2024), while in Cyprinion species, tetra-sectioned lepidonts serve as distinct taxonomic markers (Faal *et al.*, 2024). Taxonomically, lepidont characteristics have been used to distinguish fish species effectively. Similarly, studies on *Garra shamal* have linked lepidont morphology to higher taxonomic resolution (Al Jufaili *et al.*, 2023). Furthermore, ontogenetic variation influences scale morphology, including lepidont structure, as these features change with fish growth. This highlights the importance of considering developmental stages when using lepidonts for taxonomic classification (Al Jufaili *et al.*, 2021). Although lepidont morphology provides valuable taxonomic insights, it is also subject to environmental influences and individual variation, which may complicate its use in systematic studies. Nevertheless, the distinct lepidont structures observed in different koi scale types reinforce their significance in scale morphology research, supporting broader studies on koi fish scale evolution and structural adaptations.

Koi fish variants studied exhibit distinct characteristics in their scale morphology, as summarized in Table 1. Common scale variant features uniform cycloid scales that cover the entire body, while Ginrin scale variant shares the same cycloid shape but is distinguished by its shiny appearance. Shusui variant is unique with elongated wing-like scales limited to the dorsal fin, leaving the rest of the body scaleless. Doitsu variant, on the other hand, is entirely scaleless, exposing the skin's color pattern. Differences at the microscopic level are also evident; common scales have tubercles in the posterior field, which are absent in Ginrin scales, and Shusui scales exhibit tubercles and densely packed, sharp lepidonts. Circuli patterns vary, with common and Ginrin scales showing rounded patterns extending from the focus, while Shusui scales have closer, more elongated circuli. These distinctive morphological features, highlighted in Table 1, underscore the diversity within koi fish variants and the utility of SEM in identifying the differences.

The comparison between scaled and scaleless fish highlights distinct evolutionary trade-offs influencing their ecological roles. Scaled fish benefit from enhanced protection, improved hydrodynamic efficiency, and usefulness in taxonomic identification, but they face limitations in flexibility and vulnerability to scale loss. In contrast, scaleless fish exhibit increased flexibility and enhanced cutaneous respiration, allowing them to thrive in specific environments, though they are more susceptible to predation and ecological constraints (Gu *et al.*, 2023; Clark and Uyeno, 2024; Ujjaina and Jaiswar, 2024). However, it highlights genetic adaptations in scaleless fish, suggesting potential advantages in certain environments, while scales may provide protection and structural support. In other cyprinid fish, scale loss has been linked to changes in genes involved in scale formation and regulation (Fikri *et al.*, 2022; Ding *et al.*, 2024). In koi fish, the Doitsu variant may possess distinct genetic differences compared to the scaled koi variant; however, further research is required to identify the specific genes responsible and understand their regulatory mechanisms. Future studies should focus on genetic sequencing and functional analysis to uncover the molecular basis of scale loss in Doitsu koi, which could provide broader insights into evolutionary adaptations in koi fish.

CONCLUSION

The utilization of SEM for characterizing fish scale morphology offers valuable insights into the scales of these koi fish variants. Macroscopic distinctions are often imperceptible to the naked eye but become clear under SEM examination. This study revealed significant differences between common and Ginrin scale types, emphasizing the importance of SEM in revealing variations such as the presence of tubercles and the frequency of lepidonts. Furthermore, the unique morphology of Shusui scales underscores the diversity within koi fish variants, with ultrastructural analysis highlighting distinct characteristics in circuli and lepidonts. While koi fish share structural similarities with

other cyprinids, SEM enables precise comparisons revealing both commonalities and variations across species. The potential for SEM extends beyond characterization, offering a basis for future research to integrate anatomical and molecular approaches for comprehensive understanding. Overall, SEM emerges as a valuable tool for characterizing scale morphology in koi fish, providing valuable insights for hobbyists, breeders, and researchers.

ACKNOWLEDGEMENTS

The authors thank Universitas Gadjah Mada and Directorate General of Higher Education, Research, and Technology of the Ministry of Education, Culture, Research, and Technology of Republic of Indonesia. This study was supported by a research grant Pendidikan Magister menuju Doktor untuk Sarjana Unggul (PMDSU) fiscal year 2022–2024 from the Directorate General of Higher Education, Research, and Technology of the Ministry of Education, Culture, Research, and Technology of Republic of Indonesia, grant number 2065/UN1/ DITLIT/PT.01.03 /2024.

AUTHORS' CONTRIBUTIONS

All authors contributed significantly to this research. KNA conducted the study, performed the analysis, and drafted the manuscript. HW, NW, SK, and AH contributed to data analysis, critically reviewed the draft, and provided valuable feedback to enhance the manuscript. All authors have thoroughly reviewed and approved the final version of the manuscript.

COMPETING INTERESTS

The authors declare that they have no competing interests.

REFERENCES

- Al Jufaili, S. M., Masoumi, A. H., Esmaeili, H. R., Jawad, L. A., & Teimori, A. (2021). Morphological and microstructural characteristics of scales in longnose goby

- Awaous jayakari* (Teleostei: Gobiidae): Light and scanning electron microscopy approaches. *Microscopy Research and Technique*, 84(12), 3128–3149.
- Al Jufaili, S. M., Echreshavi, S., & Esmaeili, H. R. (2023). Scales surface topography: Comparative ultrastructural and decorative characteristics of a modern elasmoid fish scales in a cyprinid fish, *Garra shamal* (Teleostei: Cyprinidae) using digital optical light and scanning electron microscope imaging. *Microscopy Research and Technique*, 86(1), 97–114.
- Andrian, K. N., 'Aisy, N. R., Novindasari, B. B. M., Nurrahmi, I. A., Santi, M. D., & Haryanto, A. (2023). Random Amplified Polymorphic DNA-Polymerase Chain Reaction Analysis of Four Koi Fish (*Cyprinus carpio* var. *koi*) Variants from Yogyakarta, Indonesia. *IOP Conference Series: Earth and Environmental Science*, 1174, 1–7.
- Andrian, K. N., Wihadmadyatami, H., Wijayanti, N., Karnati, S., & Haryanto, A. (2024). A comprehensive review of current practices, challenges, and future perspectives in Koi fish (*Cyprinus carpio* var. *koi*) cultivation. *Veterinary World*, 17(8), 1846–1854.
- Ansari, S., Chavan, S., & Shaikh, Y. (2021). Scanning electron microscopy of scales in two edible fishes *Notopterus kاپirat* and *Etroplus suratensis* for its application in taxonomy. *International Journal of Fisheries and Aquatic Studies*, 9(4), 315–319.
- Aysi, N. R., Santi, M. D., Andrian, K. N., & Haryanto, A. (2022). Molecular fish sexing on Kohaku Koi (*Cyprinus carpio*) based on ArS.9–15 gene amplification by PCR method. *IOP Conference Series: Earth and Environmental Science*, 976(1), 1–6.
- Clark, A. J., & Uyeno, T. A. (2024). The biomechanics of fish skin. In *Encyclopedia of Fish Physiology*. Elsevier, pp: 476–498.
- de Kock, S., & Gomelsky, B. (2015). Japanese Ornamental Koi Carp: Origin, Variation and Genetics. In *Biology and Ecology of Carp*. CRC Press, pp: 27–53.
- Dey, S., Biswas, S. P., & Dey, S. (2015). Scanning electron microscopy of scale microstructures in *Channa barca*, a poorly known snake head fish in regard to taxonomy. *Microscopy*, 64(5), 351–359.
- Dey, S., Biswas, S. P., Dey, S., & Bhattacharyya, S. P. (2014). Scanning electron microscopy of scales and its taxonomic application in the fish genus *Channa*. *Microscopy and Microanalysis*, 20(4), 1188–1197.
- Dhamayanti, Y., Khairunnisa, H. K., Zahrudin, E., Bayram, M., & Suciyo, S. (2024). Immune responses of club cells in fish: A review. *Jurnal Medik Veteriner*, 7(2), 407–412.
- Ding, Y., Zou, M., & Guo, B. (2024). Genomic signatures associated with recurrent scale loss in cyprinid fish. *Integrative Zoology*, 00, 1–16.
- Echreshavi, S., Esmaeili, H. R., Teimori, A., Safaie, M., & Owfi, F. (2021). Hidden morphological and structural characteristics in scales of mullid species (Teleostei: Mullidae) using light and scanning electron digital imaging. *Microscopy Research and Technique*, 84(11), 2749–2773.
- Esmaeili, H. R., & Gholami, A. (2011). Scanning Electron Microscopy of the scale morphology in Cyprinid fish, *Rutilus frisii kutum* Kamenskii, 1901 (Actinopterygii: Cyprinidae). *Iranian Journal of Fisheries Sciences*, 10(1), 155–166.
- Faal, S. A., Esmaeili, H. R., & Teimori, A. (2024). Scale Characteristics of Six Fish Species of the Genus *Cyprinion* (Teleostei: Cypriniformes): A Microscopic Analysis. *Microscopy and Microanalysis*, 30(4), 771–792.
- Fikri, F., Wardhana, D. K., Purnomo, A., Khairani, S., Chhetri, S., & Purnama, M. T. E. (2022). Aerolysin gene characterization and antimicrobial resistance profile of *Aeromonas hydrophila* isolated from milkfish (*Chanos chanos*) in Gresik, Indonesia. *Veterinary World*, 15(7), 1759–1764.
- Ghods, S., Waddell, S., Weller, E., Renteria, C., Jiang, H. Y., Janak, J. M., Mao, S. S., Linley,

- T. J., & Arola, D. (2020). On the regeneration of fish scales: structure and mechanical behavior. *Journal of Experimental Biology*, 223(10), jeb211144.
- Gomelsky, B., Delomas, T. A., Schneider, K. J., Anil, A., & Warner, J. L. (2015). Inheritance of Sparkling Scales (Ginrin) Trait in Ornamental Koi Carp. *North American Journal of Aquaculture*, 77(3), 312–317.
- Gu, H., Wang, H., Zhu, S., Yuan, D., Dai, X., & Wang, Z. (2023). Interspecific differences and ecological correlations between scale number and skin structure in freshwater fishes. *Current Zoology*, 69(4), 491–500.
- Murcia, S., Lavoie, E., Linley, T., Devaraj, A., Ossa, E. A., & Arola, D. (2017). The natural armors of fish: A comparison of the lamination pattern and structure of scales. *Journal of the Mechanical Behavior of Biomedical Materials*, 73, 17–27.
- Novindasari, B. M., Nurrahmi, I. A., & Andrian, K. N. (2024). Molecular fish sexing on Taisho Sanshoku Koi (*Cyprinus carpio*) based on ArS.9-15 gene amplification using PCR method. *Jurnal Medik Veteriner*, 7(2), 255–263.
- Raffealla, N., & Bhuyan, R. (2020). Comparative study of fish scale using scanning electron microscopy in two Cyprinid fishes (*Neolissochilus hexagonolepis* and *Neolissochilus hexastichus*) found in Meghalaya, North-East India. *International Journal of Life Sciences*, 8(1), 77–82.
- Scott, E., Edgley, D. E., Smith, A., Joyce, D. A., Genner, M. J., Ioannou, C. C., & Hauert, S. (2023). Lateral line morphology, sensory perception and collective behaviour in African cichlid fish. *Royal Society Open Science*, 10(1), 221478.
- Suciyono, S., Mukti, A. T., Kenconoajati, H., Ulkhaq, M. F., Fasya, A. H., Lamadi, A., Imlani, A., & Rao Mariah, S. (2023). Color brightness and growth levels of goldfish (*Carassius auratus*) reared with different light spectrums. *Jurnal Medik Veteriner*, 6(2), 250–255.
- Ujjania, N. C., & Jaiswar, A. K. (2024). Scale morphology an additional tool for taxonomy and fish identification with reference to Nemipteridae fishes (*N. japonicus*, *N. bipunctatus* and *N. randalli*). *Flora and Fauna Jhansi*, 30(2), 11–18.
- Vernerey, F. J., & Barthelat, F. (2010). On the mechanics of fishscale structures. *International Journal of Solids and Structures*, 47(17), 2268–2275.
