External morphological characteristics and histological structures of needle cuttlefish (*Sepia aculeata*) larvae from Thai waters

Praewa Kongmeephol^{a,b}, Xiaodong Zheng^a, Charuay Sukhsangchan^{b,*}

^a Key Laboratory of Mariculture, Ministry of Education, Ocean University of China, Qingdao 266003 China

^b Department of Marine Science, Faculty of Fisheries, Kasetsart University, Bangkok 10900 Thailand

*Corresponding author, e-mail: ffiscrs@ku.ac.th

Received 27 Aug 2024, Accepted 17 Apr 2025 Available online 28 May 2025

ABSTRACT: The internal and external anatomies of cuttlefish larvae in Thailand have rarely been studied. This information is valuable not only for understanding their early development but also for applications in cephalopod distribution and abundance surveys. Needle cuttlefish (*Sepia aculeata*) eggs were collected from Phetchaburi Province, Thailand. Sixty hatchlings were prepared to observe the external morphology (30 specimens) and internal histology (30 specimens). A notable external morphological characteristic of the larvae is a broad, sac-like mantle. The hatching gland (Hoyle organ) and Kölliker organs are not visible on the skin surface. Chromatophores are distributed across the body, head, arms, and tentacles but are absent from the fins and funnel. White spots are present on the dorsal side, not on the ventral side. Lateral lines appear on the head and arms. The fin length is 80% of the mantle length (ML), the eyes are large and round, and the funnel is stout, with a length 30% of the ML. The arm formula is IV>I>III>II. The optic lobe is large, rounded, and rectangular, filled with medullar neuropil surrounded by pericaryal islands. It is separated into the cortex and medulla. The cortex consists of three layers: outer granule cell layer, outer neuropil layer, and inner granule cell layer. The eye comprises a circular lens, iris, ciliary body, and curved retina. The obtained data provide essential insights into the early developmental stage of *S. aculeata* and serve as fundamental information for advancing research on cephalopod development, lifecycle, and phylogenetic relationships, particularly within Thai marine ecosystems.

KEYWORDS: external morphology, histological structures, cephalopod larvae, needle cuttlefish, Sepia aculeata

INTRODUCTION

Cuttlefish are marine mollusks of the family Sepiidae, class Cephalopoda. Cuttlefish have an oval body, Wshaped pupil, 4 pairs of arms, and a pair of tentacles. Cuttlefish possess an internal shell structure called a cuttlebone, which is used to control buoyancy. In Thai waters, there are 13 species of cuttlefish from 3 genera, including *Metasepia tullbergi*, *Sepia aculeata*, *S. arabica*, *S. brevimana*, *S. esculenta*, *S. kobiensis*, *S. latimanus*, *S. lycidas*, *S. pharaonis*, *S. prashadi*, *S. recurvirostra*, *S. stellifera*, and *Sepiella inermis* [1].

The life stages of cuttlefish in Thai waters have been studied in *Sepia pharaonis* and *Sepiella inermis*, with embryonic development consisting of 17 and 15 stages, respectively, and hatchling sizes of 0.680 ± 0.123 cm and 0.498 ± 0.119 cm, respectively [2, 3]. Differences in embryonic duration are influenced by egg size, nutrient reserves, and physical factors such as temperature. Embryos with greater nutrient reserves and a benthic lifestyle tend to have a longer developmental period, whereas those with lower reserves and exposed to temperatures above $28 \,^\circ$ C hatch earlier [3, 4]. However, studies on the embryonic development of cuttlefish remain limited, including for *S. aculeata*.

Many species of cuttlefish, including the needle cuttlefish (*S. aculeata*), are commercially important [5]. In Thai waters, S. aculeata is found along coastal areas at depths of up to 60 meters [6]. Efforts have been made in Thailand to gather and evaluate data on needle cuttlefish, but much information is lacking, particularly regarding the morphological characteristics and histological structures of larvae. The external characteristics of Sepiella inermis, Sepia pharaonis, and S. aculeata have been reported [7,8]. However, histological investigations of S. aculeata larvae have not been reported. Histological structures of the cuttlebone sac have been described in *S. esculenta* [9]. The histological structures of the suckers in S. esculenta and S. lycidas, the ink gland of S. pharaonis, and the muscle and liver of S. pharaonis under salinity stress have been studied [10-12]. Despite these studies, the histological structures of cuttlefish larvae remain poorly understood, except for a report on the eye and central nervous system of S. officinalis [13].

Information about the external morphology and histology of *S. aculeata* larvae is essential for determining the species distribution and abundance. Knowledge of the larvae is also essential for a full understanding of the cephalopod life cycle; the early life stages of cephalopods are of great interest and importance for fundamental research, especially in their biology and ecology [14]. The purpose of this research was to study and document the external morphology and histology of *S. aculeata* larvae to support the development of a

database of cephalopod larvae found in Thai waters.

MATERIALS AND METHODS

Sample collection

Eggs of *S. aculeata* were collected from Chao Samran Beach, Phetchaburi Province, Thailand, in May 2022, where they were attached to fishing gear. The eggs were then transported to the laboratory of the Department of Marine Science, Faculty of Fisheries, Kasetsart University, Thailand. They were placed in 10-l glass flasks with controlled water quality parameters: salinity 32–35 psu, temperature 29–33 °C, and pH 7.5–8.5. Constant aeration was provided with aerators until hatching. Hatchlings were prepared for observation of external morphological characteristics by anesthetizing them with 5% magnesium chloride and fixing in 10% formaldehyde, while the specimens for histological examination were preserved following the methods described below [15].

External morphology

Morphometric measurements

The eggs were measured in length and width on the days close to hatching (Fig. 1a). Hatchlings (n = 30) were collected from newly-hatched eggs and kept in 10% formaldehyde for observation under a stereo microscope and scanning electron microscope (SEM). Hatchling weights (W) were recorded using an analytical balance with four decimal places to ensure precise measurements. Measurements of specimens were recorded [16], including total length (TL), mantle length (ML), mantle width (MW), fin length (FL), fin width (FW), head length (HL), head width (HW), eye diameter (ED), funnel length (FL), arm length (AL), tentacle length (TcL), and club length (ClL) (Fig. 1b,c).

Various morphological indices were calculated to conduct proportional comparisons. Eye diameter indices were expressed as a percentage of HL, while all other measurements were standardized as a percentage of ML.

Scanning electron microscope (SEM)

Hatchlings were gradually dehydrated through an ethanol series of 75%, 80%, 85%, 90%, 95%, and 100%, with each concentration lasting 15 minutes. The dehydrated samples were air-dried using a K850 critical point dryer, mounted on metal stubs, coated with platinum using a Quorum Q150R ES sputter coater, and examined under a Hitachi SU8020 field emission scanning electron microscope (FE-SEM).

Histological examination

Thirty larvae were preserved in 10% formaldehyde and then dehydrated in a graded series of ethanol before being transferred to xylene and embedded in paraffin wax [15]. Serial tissue sections were cut using a rotary microtome (RMC Boeckeler, MR3, USA) with 5 μ m thickness. The tissue sections were placed on a glass slide and stained with hematoxylin and eosin (H&E), and then histological structures were observed and photographed under a light microscope.

RESULTS

Egg and larva characteristics

The eggs of *S. aculeata* are contained in individual capsules. The capsules are soft, clear, transparent, rounded at the ends, and are attached to the substrate by stalks. The 30 eggs examined ranged in length from 16.79 to 17.42 mm (mean 17.09 ± 0.19 mm) and width from 11.49 to 14.00 mm (mean 13.04 ± 0.78 mm). The mantle lengths of hatchlings ranged from 3.38 to 4.38 mm (mean 3.87 ± 0.24 mm), with a total length ranged from 6.38 to 8.13 mm (mean 7.22 ± 0.46 mm) and weights ranged from of 0.0233 to 0.0517 g (mean 0.0351 ± 0.0070 g) (Fig. 2; Table 1).

Diagnostic features: mantle is broad and sac-like, and length is greater than width. The hatching gland (Hoyle organ) and Kölliker organs are not visible on the surface skin. No rostrum protrudes from the posterior tip of the mantle (Fig. 2a). Fins are long (80% of ML) and located on the terminal region of the mantle. The posterior end of the fins is unconnected. Cuttlebone is oval, thin, and translucent, making the septa faintly visible, while the inner cone is not apparent and the rostrum is small (Fig. 2b). Head is large (84% of ML) and wider than mantle. Eyes are large and round (54 and 56% of HL for left and right eyes, respectively) (Fig. 3a,b). Funnel is stout (30% of ML in length). Arm formula is IV>I>III>II. Suckers are globular, equal in size, and closely packed. Arm pair 1 has 2-3 rows; arm pairs 2 and 3 have 3-4 rows, and arm pair 4 has 4 rows. Lateral lines appear on head and arms. (Fig. 3c) Tentacular clubs are short (20 and 18% of ML on left and right, respectively). Suckers on tentacular clubs are small, closely packed in 5-7 rows (Fig. 3d). Ink sac is large, round, and visible on the ventral surface. Chromatophores are spread over the body, head, arms, and tentacles, but not on the fin or funnel. Chromatophores are brown, black, and yellow spots. White spots are spread over the dorsal side, but not the ventral side.

Histology

Histological examination of the eye and optic lobe revealed intricate structures essential for visual processing. The eye comprises a circular lens, iris, ciliary body, and curved retina (Fig. 4a). The retina contains thick proximal segments with photoreceptors and supporting cell nuclei separated by a basal membrane. The basal segment is densely packed with pigment granules, while the distal photoreceptive segments are filled with rhabdomeres and pigment granules



Fig. 1 Morphological measurements of needle cuttlefish (*S. aculeata*): (a) egg; (b) hatchling-dorsal side; and (c) hatchling-ventral side. Morphometric characters measured: total length (TL), mantle length (ML), mantle width (MW), fin length (FL), fin width (FW), head length (HL), head width (HW), eye diameter (ED), funnel length (FuL), arm length (AL), tentacle length (TcL), and club length (ClL).



Fig. 2 Morphology of S. aculeata larva: (a) dorsal view and (b) cuttlebone. Scale is 1 mm.

(Fig. 4b). The optic lobe, consisting of two large, rounded, and rectangular lobes, is located near the eyes and is filled with medullar neuropil surrounded by pericaryal islands (Fig. 4c). Positioned between the eye and the optic lobe is the anterior chamber organ. The optic lobe of cephalopods is separated into the cortex and medulla. The cortex consists of three layers: outer granule cell layer, outer neuropil layer, and inner granule cell layer. The outer neuropil layer is thicker than the others, and the outer plexiform layer curves inward towards the outer granular layer (Fig. 4d).

DISCUSSION

The external morphological characteristics and histological structures of cuttlefish larvae remain limited in Thai waters, particularly for *S. aculeata*, which is a commercially important species. The external morphology of larvae has been described for only a few cuttlefish species to date (Table 2).

The eggs of *S. aculeata* resemble those of the pharaoh cuttlefish (*S. pharaonis*) [17, 18]. They are contained in individual capsules and are soft, transparent, white, and round with a tip stalk. They measure 17.09 ± 0.19 mm in length and 13.04 ± 0.78 mm in width, which is smaller than pharaoh cuttlefish eggs (29.08 ± 0.45 mm in length and 17.67 ± 0.17 mm in width) [19]. After hatching, the larvae of both species are benthic [17].

The external morphological characteristics of *S. aculeata* larvae observed in this study — including a broad, sac-like mantle; narrow, elongated fins that are not connected at the posterior end; and small, short tentacular clubs with sub-equal size of suckers — are consistent with a previous report [8]. However,

3

Table 1 Morphometric characteristics of needle cuttlefish (*S. aculeata*) larvae (n = 30). The two eye-diameter indices express length as a percentage of HL. All other indices express length as a percentage of ML.

Index	Mean	SD	Range		
			Minimum	Maximum	
Mantle length (mm)	3.87	0.24	3.38	4.38	
Mantle width index (MWI)	80.90	5.09	73.33	90.32	
Fin length index (FLI)	79.50	9.57	58.62	96.67	
Fin width index (FWI)	11.70	3.92	4.69	20.97	
Head length index (HLI)	46.50	5.61	35.29	62.07	
Head width index (HWI)	84.08	6.24	71.43	96.77	
Left eye diameter index (LEDI)	54.49	7.62	43.75	75.00	
Right eye diameter index (REDI)	55.98	7.32	44.44	75.00	
Funnel length index (FuLI)	30.35	5.07	20.59	40.00	
Arm length index (ALI):					
Arm I - Left	44.70	5.23	35.63	56.30	
Arm II - Left	25.63	4.27	16.00	33.55	
Arm III - Left	31.30	4.45	23.53	40.00	
Arm IV - Left	51.43	5.93	37.65	65.93	
Arm I - Right	45.37	5.38	33.33	58.00	
Arm II - Right	25.58	5.14	16.67	34.67	
Arm III - Right	31.00	6.06	22.00	44.67	
Arm IV - Right	51.69	9.11	35.00	70.15	
Left tentacle length index (LTcLI)	76.12	12.86	54.56	98.13	
Right tentacle length index (RTcLI)	76.57	20.05	50.34	115.20	
Left club length index (LClLI)	19.81	3.67	12.35	28.28	
Right club length index (RClLI)	18.23	3.19	13.53	28.67	



Fig. 3 SEM of needle cuttlefish (*S. aculeata*) larvae: (a) dorsal side; (b) lateral side; (c) lateral line (red arrow); and (d) suckers on tentacular club. Scale bar in (a)–(c) is 1 mm and in (d) is 200 μ m.

our study found the arm formula was IV>I>III>II, which differs from the previous report [8]. Chumdang [2] reported that in hatchling pharaoh cuttlefish (*S. pharaonis*), Hoyle organ disappears and the external morphology resembles that of the adults. The *S. aculeata* larvae examined in this study also had

no Hoyle organ on the skin, and the hatchlings were similar in appearance to adults but were smaller than the *S. pharaonis* hatchlings. In *S. officinalis*, Hoyle organ is an anchor-shaped structure located on the posterior dorsal mantle [20].

Another difference from previous research was the



Fig. 4 Histological tissues of *S. aculeata* larvae: (a) transverse section of eye, (b) transverse section of retina, (c) longitudinal section of head, and (d) longitudinal section of optic lobe. Labels: al = anterior part of eye lens, aco = anterior chamber organ, bm = buccal mass, bs = basal segment, cm = ciliary muscle, co = cornea, ctx = cortex, ds = distal segments of photoreceptors, ey = eye, ig = inner granular cells, ir = iris, le = lens, ll = lower eyelid, m = medullar, nol = medullar neuropil of optic lobes, og = outer granular cells, ol = optic lobe, on = optic nerve, oneu = outer neuropil, op = outer plexiform, pl = posterior part of eye lens, ps = proximal segments of photoreceptors, re = retina, rp = retinal plexus, ul = upper eyelid, and scn = supporting cell nuclei. Scale bar is 100 µm in (a,b) and 500 µm in (c,d).

Table 2	Comparison of	of the external	characteristics	of cuttlefish	larvae in the	genus Sepia.
---------	---------------	-----------------	-----------------	---------------	---------------	--------------

Species	Mantle form	Dorsal mantle margin	Fin shape	Suckers on arms	Arm formula	Cuttlebone	Spine	Hatchling ML (mm)	Source
S. aculeata	Broad, sac-like	Free	Long	Arms I: 2–3 rows Arms II, III: 3–4 rows Arms IV: 4 rows	IV>I>III>II	Oval	Present	3.87	Present study
S. aculeata	Broad, sac-like	Free	Narrow, long	Arms I–III: 2–3 rows Arms IV: 4 rows	I>IV>III>II	-	-	3.69	[7]
S. pharaonis	Oval-wide	Free	Narrow, long	Arms I: 18–22 suckers Arms II: 38–42 suckers Arms III: 18–24 suckers Arms IV: 16–22 suckers	IV>III>II>I	_	_	-	[2]
S. pharaonis	Broad, sac-like	Free	Narrow, long	Arms I–IV: 4 rows	IV>I>III>II	_	Present	7.1	[7]
S. officinalis	Forward into an obtuse angle behind head	-	Wide	_	_	Posterior widened	Present	8.3	[34]

No data.

discovery of lateral lines on the head and arms of the larvae. Nine cephalopod taxa, including Sepioidea, Teuthoidea, and Octopoda, are known to have epidermal lines or lateral lines [21]. These lateral lines function as mechanoreceptors, detecting local water movements, and are probably involved in prey detection [22–24].

The histological structures of S. aculeata larvae also have remained mostly unexamined, especially in Thai waters. In this study, the histological structure of the eye and optic lobe in S. aculeata was consistent with findings from previous research on the brain structure of Loligo and octopus [25, 26]. In addition, the histology of the eyes and brain of S. officinalis revealed eyes with a circular lens, curved retina, and rounded rectangular optic lobe, which are features similar to our observations in S. aculeata [13]. However, S. aculeata has smaller eyes than S. officinalis. S. officinalis has thicker distal photoreceptor segments (ds), while S. aculeata has thicker supporting cell nuclei. Although the eyes of hatchling cuttlefish are fully grown and have the same structure as adults, there are differences in the thickness and development of particular layers of the retina [27]. The dimensions of the brain lobes also can change from hatchling to juvenile or adult. The characteristics of these structures have been linked to physical, behavioral, and environmental changes [13, 27].

Although external morphological characteristics provide valuable information, they may not be sufficient to distinguish between species, especially in the larval stage where specific traits can be highly variable and difficult to define [20]. Histological examination offers information about internal structures and developmental phases that outward appearance alone cannot convey. Genetic studies, such as the use of COI, 16S rRNA, and 12S rRNA genes, have proven effective in differentiating closely related species and uncovering hidden diversity [28-31]. Combining these findings with genetic data will allow for more precise species identification and provide insights into the evolutionary relationships and ecological roles of these organisms. These genetic markers offer high resolution and are extensively represented in genomic databases, facilitating easy comparison and identification [32, 33]. However, to acquire a complete understanding, external morphological characteristics must be conducted in addition to genetic research.

CONCLUSION

The external morphology and histology of *S. aculeata* larvae were examined. The mantle was broad and sac-like with long fins that were not connected at the posterior end. At this stage of development, the Hoyle organ and Kölliker organs were not visible on the surface of the skin. According to SEM imaging, the lateral or arm line appeared as a longitudinal groove

www.scienceasia.org

on the larvae's head and arms. Histological structures were observed on the eyes and optic lobe. Our study is the first to document these histological structures in larvae of *S. aculeata*. These data can be added to the larval database to further our knowledge of the its lifecycle, biology, and ecology. Furthermore, the information may be applied to studies of cephalopod development and phylogenetic relationships.

Acknowledgements: This study was financed by China Scholarship Council (CSC) Scholarship and Dual-Degree Master's Program between Ocean University of China and Kasetsart University. The authors also thank Pawida Prasopsook and Sonthaya Phuynoi for laboratory support.

REFERENCES

- Nabhitabhata J, Tuanapaya S, Tongtherm K, Promdam R, Aungtonya C (2019) First record of seven neritic cephalopods (Sepiidae, Loliginidae, Octopodidae) new to Thai waters with taxonomic list of Thai Teuthofauna. *Phuket Mar Biol Cent Res Bull* 76, 41–62.
- Chumdang J (1998) Embryonic and paralarva development of pharaoh cuttlefish (*Sepia pharaonis* Ehrenberg, 1831) and big fin squid (*Sepioteuthis lessoniana* Lesson, 1830). MSc thesis, Kasetsart University, Thailand.
- Nabhitabhata J, Polkhan P, Kbilrum S (1984) Culture, growth and behavior of spineless cuttlefish, *Sepiella inermis* Fer. & d' Orb. Technical Paper 5/1984, Department of Fisheries.
- Boonprakob P, Siripoonya P, Yodyingyuad U, Satayalai O, Sithigorngul P (1977) Studies on embryonic development, karyotype, effects of physical factors on development and behavioral response to physical factors of cuttlefish *Sepiella inermis*. Research Paper, Chulalongkorn University, Thailand.
- Yamrungrueng A, Noranarttragoon P, Boonnjorn N, Thongsila K, Rungpatikorn N (2016) Cuttlefish fisheries from commercial trawler in the Gulf of Thailand. Technical Paper 10/2016, Department of Fisheries.
- Reid A, Jereb P, Roper CFE (2005) Cuttlefishes. In: Jereb P, Roper CFE (eds) Cephalopods of the World: An Annotated and Illustrated Catalogue of Cephalopod Species Known to Date Volume 1: Chambered Nautiluses and Sepioids (Nautilidae, Sepiidae, Sepiolidae, Sepiadariidae, Idiosepiidae and Spirulidae), FAO, Rome, pp 56–212.
- Sukhsangchan C, Sunthornket P (2015) Species identification of commercially important cephalopods of Thailand. *Fish People* 13, 37–42.
- Sukhsangchan C, Sunthornket P, Phuynoi S (2017) Morphological characteristics of paralarvae of cephalopods found in Thai waters. *Mar Biodivers* 47, 639–645.
- Zheng X, Xiao S, Wang Z, Wang R (2007) Histological and histochemical analyses of the cuttlebone sac of the golden cuttlefish *Sepia esculenta*. J Ocean Univ China 6, 393–397.
- Kimbara R, Nakamura M, Oguchi K, Kohtsuka H, Miura T (2020) Pattern of sucker development in cuttlefishes. *Front Zool* 17, 24.
- 11. Jiang M, Xue R, Chen Q, Zhan P, Han Q, Peng R, Jiang X (2020) Histology and ultrastructure of ink gland and melanogenesis in the cuttlefish *Sepia pharaonis*. *Invertebr Biol* **139**, e12306.

ScienceAsia 51 (3): 2025: ID 2025042

- 12. Yin SJ, Zhang L, Zhang L, Wan J, Song W, Jiang X, Park YD, Si YX (2018) Metabolic responses and arginine kinase expression of juvenile cuttlefish (*Sepia pharaonis*) under salinity stress. *Int J Biol Macromol* **113**, 881–888.
- Wild E, Wollesen T, Haszprunar G, Heß M (2014) Comparative 3D microanatomy and histology of the eyes and central nervous systems in coleoid cephalopod hatchlings. Org Divers Evol 15, 37–64.
- Rodhouse PG, Elvidge CD, Trathan PN (2001) Remote sensing of the global light-fishing fleet: An analysis of interactions with oceanography, other fisheries and predators. *Adv Mar Biol* **39**, 261–303.
- 15. Romratanapun S (2002) Animal Tissue Technique, Kasetsart University Press, Bangkok.
- Roper CFE, Voss GL (1983) Guidelines for taxonomic descriptions of cephalopod species. *Mem Natl Mus Vic* 44, 48–63.
- Nabhitabhata J, Nilaphat P (1999) Life cycle of cultured pharaoh cuttlefish, *Sepia pharaonis* Ehrenberg, 1831. *Phuket Mar Biol Cent Spec Publ* 19, 25–40.
- Samuel D, Patterson J (2015) Studies on the embryonic development of pharaoh's cuttlefish *Sepia pharaonis* Ehrenberg, 1831 under laboratory conditions. *Indian J Geo Mar Sci* 44, 519–527.
- Lee MF, Lin CY, Chiao CC, Lu CC (2016) Reproductive behavior and embryonic development of the pharaoh cuttlefish, *Sepia pharaonis* (Cephalopoda: Sepiidae). *Zool Stud* 55, 41.
- 20. Vidal EAG, Shea EK (2023) Cephalopod ontogeny and life cycle patterns. *Front Mar Sci* **10**, 1162735.
- 21. Hanlon RT, Messenger JB (1996) *Cephalopod Behaviour*, Cambridge University Press, UK.
- 22. Bleckmann H, Budelmann B (1990) On the senses of the cuttlefish: A lateral line system in cephalopods. *German Res* **3**, 8–10.
- Komak S, Boal JG, Dickel L, Budelmann BU (2005) Behavioural responses of juvenile cuttlefish (*Sepia of-ficinalis*) to local water movements. *Mar Freshw Behav Physiol* 38, 117–125.
- Budelmann BU, Riese U, Bleckmann H (1991) Structure, function, biological significance of the cuttlefish "lateral lines". In: Boucaud-Camou E (ed) *The Cuttlefish: First International Symposium on the Cuttlefish Sepia*, Centre de Publications de l'Universite de Caen, pp 201–209.
- 25. Young JZ (1974) The central nervous system of *Loligo*. I. The optic lobe. *Philos Trans R Soc Lond B Biol Sci* **267**,

263–302.

- 26. Anadón R (2019) Functional histology: the tissues of common coleoid cephalopods. In: Gestal C, Pascual S, Guerra Á, Fiorito G, Vieites JM (eds) Handbook of Pathogens and Diseases in Cephalopods, Springer, New York, pp 39–85.
- 27. Fernández-Gago R, Molist P, Anadón R (2019) Tissue of paralarvae and juvenile cephalopods. In: Gestal C, Pascual S, Guerra Á, Fiorito G, Vieites JM (eds) *Handbook* of *Pathogens and Diseases in Cephalopods*, Springer, New York, pp 87–109.
- Khatami S, Tavakoli-Kolour P, Valinassab T, Anderson FE, Farhadi A (2018) Molecular identification and phylogenetic relationships of Coleoidea (Mollusca: Cephalopoda) from the Persian Gulf and Oman Sea reveals a case of cryptic diversity. *Molluscan Res* 38, 77–85.
- 29. Tan HY, Goh ZY, Loh KH, Then AY, Omar H, Chang SW (2021) Cephalopod species identification using integrated analysis of machine learning and deep learning approaches. *PeerJ* **9**, e11825.
- Lincy A, Anil MK, Thangaraj M, Jose JJ (2021) Molecular identification and genetic variation studies in economically important cephalopods at Beypore Fishing Harbour (Kozhikode), South West coast of India. *Not Sci Biol* 13, 10862.
- Poungthong I, Sukhsangchan C, Zheng XD (2023) Phylogenetic and morphological characteristics of *Sepioteuthis lessoniana* (Cephalopoda: Loliginidae) in the South China Sea and Andaman Sea. *ScienceAsia* 49, 232–239.
- 32. Guarneros-Narváez PV, Rodríguez-Canul R, De Silva-Dávila R, Zamora-Briseño JA, Améndola-Pimenta M, Souza AJ, Ordoñez U, Velázquez-Abunader I (2022) Loliginid paralarvae from the southeastern Gulf of Mexico: abundance, distribution, and genetic structure. *Front Mar Sci* **9**, 941908.
- 33. Olmos-Pérez L, Pierce GJ, Roura Á, González ÁF (2018) Barcoding and morphometry to identify and assess genetic population differentiation and size variability in loliginid squid paralarvae from NE Atlantic (Spain). *Mar Biol* 165, 136.
- Zaragoza N, Quetglas A, Moreno A (2015) Identification Guide for Cephalopod Paralarvae from the Mediterranean Sea. ICES Cooperative Research Report, vol 324.