First of *Regalecus russellii* (Cuvier, 1816) (Teleostei: Regalecidae) from the southwestern Thailand

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ABSTRACT

The oarfish, *Regalecus russellii* (Cuvier, 1816) is reported from Thai waters for the first time based on two specimens caught from the Andaman Sea off the coasts of Satun and Phuket provinces. The distribution range of the species extends circumglobally in tropical and temperate waters now including the Andaman Sea of Thailand, excluding northern Atlantic Ocean and Mediterranean Sea. The description of morphology of the specimens is provided showing similarities to *R. russellii* while a few characters are similar to those of *R. glense*. Genetic information clearly grouped these specimens with *R. russellii*. Combining two data sets, the specimens of oarfish in this study are clearly identified as *R. russellii*.

Keywords: DNA barcoding, first record, oarfish, Andaman Sea.

INTRODUCTION

The oarfish genus *Regalecus* Ascanius, 1772 is characterized by body distinctly slender, scaleless; premaxilla at right angle to axis of body; dorsal-fin origin above anterior rim of orbit; anterior six rays of dorsal-fin stout, distinctly elongated; pectoral fin small, fan-shaped; pelvic fin below and slightly behind pectoral fin with single ray very long; gill raker greater than 30 (Hayashi, 2002; Bray, 2008; Roberts, 2012). The genus consists of two valid species—*R. glesne* Ascanius, 1772 found in temperate seas worldwide and Mediterranean Sea, and *R. russellii* (Cuvier, 1816) found in tropical and warm temperate waters worldwide, including East China Sea and Sea of Japan (Roberts, 2012; Fricke *et al.*, 2024). The two species are very similar in external morphology including body shape, enlarge dorsal-fin rays and coloration that sometime causes misidentification. There are a few distinguishable characters such as the number of dorsal rays to vent, the number of gill rakers on the first gill arch and the number of vertebrae. Regalecid fishes are rather rare species, and their biology is poorly known because they are infrequently found alive and usually inhabit deep water ranging from 200 to 1,000 m. The specialized function of protractile toothless jaws and long gill rakers allows them to consume on euphausiid shrimps, small fish and squids (Roberts, 2012; Lee *et al.*, 2023)

Between January and February 2024, two specimens of *Regalecus russellii* were caught by Thai fishing boats trawling in the Andaman Sea off the coasts of Satun and Phuket provinces, Thailand (Figure 1). Both specimens were mostly intact, except for their bodies that were separated into 3–4 pieces, and their enlarge dorsal-fin, pectoral-fin, long pelvic-fin and caudal-fin rays were damaged. This study reports the first record of *R. russellii* in the Andaman Sea of Thailand and provides valuable information of its taxonomy and genetic diversity.



Figure 1. Map showing two collection localites of the oarfish *Regalecus russellii* in Andaman Sea, Thailand: A, THNHM-F24619, 2,311 mm SL, west of Satun, 07°01′51.6″N, 99°15′35.4″E, 3 January 2024; B, THNHM-F24625, 2,798 mm SL, *ca*. 28 km west of Phuket, 15 February 2024.

MATERIALS AND METHODS

Morphological method

Morphometric measurements and meristic counts followed the methods of Angulo and López-Sánchez (2017) except for the number of enlarge dorsal-fin rays and vertebrae were determined according to Roberts (2012). Technical terms "pectoral and pelvic insertions" and "vent" are the same meanings as "pectoral and pelvic origins" and "anus", respectively, in Angulo and López-Sánchez (2017). Morphometric and meristic measurements were taken on the left side wherever possible. Morphometrics were measured with digital calipers to the nearest 0.1 mm except for total length, standard length, and snout to vent length, which were measured with a tape measure to the nearest 1 mm. Meristics of vertebrae were observed using radiographs. These specimens are deposited at Thailand Natural History Museum, Thailand (THNHM).

Molecular analysis

Muscle tissues from 2 samples were digested with proteinase-K and extracted for genomic DNA using the commercial tissue extraction kit (NucleoSpin® Tissue Kit, Macherey-Nagel AG). The extracted DNA samples were amplified for a partial fragment of cytochrome c oxidase subunit I (COI) using F2 (5'– TCG ACT AAT CAT AAA GAT ATC GGC AC –3') and R2 (5'–ACT TCA GGG TGA CCG AAG AAT CAG AA–3') primers (Ward *et al.*, 2005). The polymerase chain reaction was performed following the protocol in Ward *et al.* (2005). The PCR products were visualized using 1.5% agarose gel. The products were purified using the commercial kit (NucleoSpin® Gel and PCR Clean-up) and then sent to sequencing using Sanger method with an ABI 3730 automated sequencer.

Two samples were obtained from the coast of the Andaman Sea of Thailand, but only one sample (THNHM-F24625) yielded the readable sequence of 651 base pairs (bp) (GenBank accession number: PP837671) while the other sample (THNHM-F24619) was degraded and resulted in the sequence of the deep-sea bacteria, *Shewanella* sp. The obtained sequence was edited in BioEdit version 7.2 (Hall, 1999). The fragment of COI was blasted in the NCBI database for comparison. Additional COI sequences of *Regalecus russellii* were also obtained from Barcode of Life Data System (BOLD). Certain COI sequences of *R. glesne* were selected for comparison due to potential mis-identification of some specimens (Lee *et al.*, 2023). Genetic distance among samples was calculated using Kimura-2-Parameter model (Kimura, 1980). The phylogenetic tree construction was performed in MEGA11 (Tamura *et al.*, 2021) using 1000 bootstrap replications based on K2 model. The COI sequence of *Agrostichthys parkeri*, which is the member in the same family, was used as an outgroup.

RESULTS

Regalecus russellii (Cuvier, 1816) [Thai name: Pla-pha-ya-nak] [English name: Oarfish] Figure 2 and 3; Tables 1 and 2



Figure 2. Two specimens of *Regalecus russellii* when fresh: A and C, THNHM-F24619, 2,311 mm SL, west of Satun, Thailand, Andaman Sea (left side, photo credit by Chayajit Deekrachang); B and D, THNHM-F24625, 2,798 mm SL, *ca*. 28 km west of Phuket, Thailand, Andaman Sea (flipped image of right side, photo credit by Sirikanya Chungthanawong).



0.10

Figure 3. The maximum likelihood tree based on 651 bp of COI fragments demonstrating the phylogenetic relationship between *Regalecus russellii* and *R. glesne*.

Character	acter R. russellii			
	THNHM-F24619	THNHM-F24625	Literature data*	Literature data*
Morphometrics				
Total length: TL (mm)	2329	2798	1,585–ca 7,300 ^k , ca 1,700–4,220 ^m , 3,379 ¹	1,500–ca 4,200 ^k , 1,549–3,190 ^g , ca1,940 ⁱ , 2,438 ^a , 3,805–4,570 ^j
Standard length: SL (mm)	2311	2786	$\begin{array}{c} 1,500-3,270^k,\\ 1,970-5,301^f,3,353^l,\\ 3,930^c,4,700^e \end{array}$	1,620–3,990 ^k
Snout to vent length: SVL (mm)	732	881	391–670 ^k , ca 450–1,380 ^m , 1415 ¹	420–1,660 ^k , 530–1,171 ^g , 812 ⁿ , 1,450–1,460 ^j
Head length: HL (mm)	174	207	108–285 ^m , 240 ^k , 265 ^c , 266 ^l , 373 ^e	$\begin{array}{c} 91{-}135^{g},137^{h},168^{n},\\ 191{-}200^{j},\!195{-}238^{k} \end{array}$
In % SL				
Snout to vent length	31.7	31.6	24.1–31.8 ^k , 42.2 ¹	25.7–49.6 ^k
Head length	7.5	7.4	6.1-8.6 ^f , 6.7 ^c , 7.9 ^{1,e}	$4.9-7.2^{k}$
Head depth	6.3	5.9	6.0-8.9 ^f , 7.2 ^l , 8.0 ^e	-
Greatest body depth	7.4	7.0	7.0 ¹ , 7.1 ^c , 9.6 ^e	-
Body depth at vent	5.7	6.1	5.0 ¹	-
Snout to dorsal-fin origin length	1.9	1.6	1.6°, 3.5 ¹	-
Snout to pectoral-fin insertion length	7.0	7.2	8.0 ¹	-
Pectoral-fin length	-	-	1.0°, 1.5–2.1 ^f , 2.2 ^l , 3.4°	-
Pectoral-fin base length	1.0	0.9	0.9^{1} , 1.1^{e} , $1.2-1.5^{f}$	-
Snout to pelvic-fin insertion length	8.1	8.3	9.2 ¹	-
Pelvic-fin length	-	-	9.7°	-
Pelvic-fin base length	0.5	0.3	<0.11	-
Snout length	1.9	1.7	2.9 ¹ , 3.5 ^e	-
Maxillary length	3.0	2.5	3.4°, 4.71	-
Maxillary depth	1.3	1.4	1.5 ¹	-
Orbit diameter	1.4	1.3	$1.0^{\circ}, 1.5^{\circ}$	-
Post-orbital head length	4.3	4.5	4.2 ¹	-
Head length	23.8	23.5	18.81	11.5–17.2 ^g , 13.2–13.7 ⁱ , 20.7 ⁿ
Head depth	19.9	18.6	16.0–17.6 ^m	$11.5-15.8^{\text{g}}, 12.1^{\text{j}}, 14.3^{\text{n}}$
Greatest body depth	23.4	22.2	19.6–21.4 ^m	14.3–17.5 ^g , 16.0 ⁿ , 20.5–21.7 ^j

Table 1. Morphometric and meristic data of two specimens of *Regalecus russellii* collected from Andaman Sea, Thailand, compare with *R. russellii* and *R. glesne* based on literature analysis.

Character		R. glesne		
	THNHM-F24619	THNHM-F24625	Literature data*	Literature data*
Body depth at vent	18.0	19.2	12.5-20.2 ^m	10.8 ⁿ , 13.4–16.2 ^g
Snout to dorsal-fin origin length	5.9	5.2	3.6-6.7 ^m	6.9 ⁿ
Snout to pectoral-fin insertion length	22.1	22.7	-	-
Snout to pelvic-fin insertion length	25.5	26.2	-	20.9–21.9 ^g , 21.6 ⁿ
Snout length	6.0	5.4	5.1-6.3 ^m	5.5 ^j
Orbit diameter	4.2	4.1	2.9-4.2 ^m	$\begin{array}{c} 2.4 - 2.7^{j}, \ 3.4 - 3.6^{g}, \\ 4.3^{n} \end{array}$
Post-orbital head length	13.5	14.2	11.6-13.3 ^m	-
In % HL				
Head depth	84.0	79.4	66.7–94.0 ^m , 90.6 ^l , 98.0–103.5 ^f , 101 ^e	${}^{69.0^{\rm n}}_{81.0^{\rm h}}, 80.2100.0^{\rm g}_{\rm g},$
Predorsal length	24.8	22.1	$40.2^{\circ}, 44.0^{\circ}$	16.1–51.9 ^g , 33.3 ⁿ
Snout length	25.1	23.1	21.3-30.5 ^m , 36.1 ¹	29.2 ^h
Maxillary length	39.2	33.9	42.4°, 59.41	36.4-44.4 ^g
Maxillary depth	17.3	18.2	18.4 ¹	15.4–22.2 ^g
Orbit diameter	18.1	17.2	12.5°, 12.6°, 14.0–17.6°°, 17.1°, 18.7 ¹	$\begin{array}{c} 17.5{-}19.5^{\rm j},\\ 20.5{-}29.6^{\rm g},20.8^{\rm n},\\ 24.8^{\rm h} \end{array}$
Post-orbital head length	57.1	60.2	53.0 ^l , 55.6–62.0 ^m , 60.4 ^c	49.6 ^h
Meristics				
Dorsal-fin rays (in total)	264	371	164°, 243–310°°, 323 ¹ , 357°	138 ^a , 164–366 ^g , 321 ^j , 340? ^b , ca422 ^d
Enlarge dorsal-fin rays	5	6	3?-7? ^k	$\begin{array}{c} 5?{-}14^{k}, 10{-}12^{d}, 12^{g}, \\ 13^{a} \end{array}$
Dorsal-fin rays to vent	65	76	58-82 ^k , 67-73 ^m	84–120 ^k , 99–111 ^g
Pectoral-fin rays	-	11	$11^{\rm e}, 11-12^{\rm f}, 11-13^{\rm k}, \\ 12-13^{\rm m}, 13^{\rm lc}$	${}^{11-13^{g}, 11-14^{k},}_{12^{abh}, 12^{-15^{j}}, 13^{d}}$
Pelvic-fin rays	1	1	1^{lcefm}	1 ^{bd}
Gill rakers on first gill arch	53(12+41)	51(13+38)	$46-49^{\text{f}},$ $49-54(11-12+38-40^{\text{m}},$ $51(14+27)^{\text{l}}$	$34-47(5-10+25-39^k, 35(6+29)^h, 27-42(5-0+22)^2, 26(6+29)^h, 27-42(5-0+22)^2, 26(6+29)^h, 27-40^h, 2$
(epibranchial+ ceratobranchial)			$51(14+57)^{k}$, $51-60(10-15+39-47)^{k}$, $55(13+42)^{c}$, 60^{c}	$5/-45(5-9+52-36)^{\text{E}}, 40^{\text{a}},$ $41(6+35)^{\text{i}}, 43(7+36)^{\text{d}},$ $43(9+34)^{\text{j}}$
Abdominal vertebrae	37	34	34–37 ^k	49?-52 ^k
Total vertebrae	97	114	-	-

Table 1. Morphometric and meristic data of two specimens of *Regalecus russellii* collected from Andaman Sea, Thailand, compare with *R. russellii* and *R. glesne* based on literature analysis. (continuous)

*Analysis from literature data based on specimens with estimate body length ≥1,500 mm SL: a, Grieg (1887); b, Waite (1899); c, Chávez-Ramos *et al.* (1985); d, Heemstra (1986); e, Castro-Aguirre *et al.* (1991) described as *Regalecus kinoi*; f, Salazar-Hermoso *et al.* (1999); g, Psomadakis *et al.* (2008); h, Schmitter-Soto (2008); i, Dulčič *et al.* (2009); j, Ruiz and Gosztonyi (2010); k, Roberts (2012); l, Angulo and López-Sánchez (2017); m, Lee *et al.* (2023); n, Galasso *et al.* (2024).

roctichthus narkeri New Zealand	galecus glesne USA-Atlantic Ocean F930351)	galecus glesne Belize R086908)	galecus glesne Atlantic Ocean Q870417)	galecus russellii iwan (ZOSKT1987-16)	galecus russellii wan (FTWS710-09)	galecus russellii iwan (KU943114)	galecus russellii uth Korea (OQ291175)	galecus russellii ailand-Phuket (THNHM-F24625)	galecus russellii P979107)	galecus russellii IC_081042)	mples
0 2371	0.2073	0.2104	0.2042	0.0043	0.0043	0.0043	0.0043	0.0000	0.0000	Ι	Regalecus russellii (NC_081042)
0.2371	0.2073	0.2104	0.2042	0.0043	0.0043	0.0043	0.0043	0.0000	Ι		Regalecus russellii (OP979107)
0.2371	0.2073	0.2104	0.2042	0.0043	0.0043	0.0043	0.0043	I			<i>Regalecus</i> <i>russellii</i> Thailand-Phuket (THNHM-F24625)
0.2307	0.2135	0.2166	0.2104	0.0043	0.0043	0.0043	I				<i>Regalecus russellii</i> South Korea (OQ291175)
0.2371	0.2135	0.2166	0.2104	0.0000	0.0000	I					<i>Regalecus russellii</i> Taiwan (KU943114)
0.2371	0.2135	0.2166	0.2104	0.0000	I						<i>Regalecus russellii</i> Taiwan (FTWS710-09)
0.2371	0.2135	0.2166	0.2104	I							<i>Regalecus russellii</i> Taiwan (ZOSKT1987-16)
0.2473	0.0022	0.0043	I								<i>Regalecus glense</i> Atlantic Ocean (OQ870417)
0.2506	0.0022	I									<i>Regalecus glesne</i> Belize (KR086908)
0.2506	I										Regalecus glesne USA-At- lantic Ocean (KF930351)
Ι											Agrostichthys parkeri New Zealand (MN123259)

Material examined. 2 specimens (2,311–2,798 mm SL)—THNHM-F24619, 2,311 mm SL, west off the coast of Satun Province, Thailand, Andaman Sea, 07°01′51.6″N, 99°15′35.4″E, 3 Jan 2024, trawling operated by Ko Thepcharoenporn fishing boat, Thanisorn Vasinopas; THNHM-F24625, 2,798 mm SL, *ca.* 28 km west of Phuket, Thailand, Andaman Sea, 15 Feb 2024, trawling operated by Po Masayanumchok fishing boat, Thanisorn Vasinopas.

Diagnosis. *Regalecus russellii* is distinguished from its congener, *R. glesne*, by the following characters in having head depth 16.0–19.0 % SVL, post-orbital length 53.0–62.0 % HL, first dorsal-fin crest with 3–6 rays united by fin membrane and second dorsal-fin crest with a single ray, 46–60 total gill rakers on first gill arch, fewer than 82 dorsal-fin rays to vent, and 34–37 abdominal vertebrae. (vs. head depth 11.5–14.3 % SVL, post-orbital length 49.6 % HL, first dorsal-fin crest with 6–8 rays united by fin membrane, and second dorsal-fin crest with 5–11 not united by fin membrane, 84–120 dorsal-fin rays to vent, 34–47 total gill rakers on first gill arch and 45–56 abdominal vertebrae in *R. glesne*) (Roberts, 2012; Angulo and López-Sánchez, 2017; this study).

Description. Morphometrics and meristics, including comparative data, are presented in Table 1. Body long and narrow, considerably compressed, ribbon-like with deepest at pectoral-fin insertion. Dorsal profile of body rising precipitously from snout tip to dorsal-fin origin and gradually lowering to caudal-fin base. Ventral profile from anterior tip of lower jaw to pelvic-fin insertion gradually declined, subsequently nearly straight and parallel with body axis to anal-fin base. Head rather small compared with body length, but maxilla and eye large compared with head length. Eyes rounded. Premaxilla narrow. Maxilla, lacrimal, preopercle, opercle, interopercle and subopercle broad, flat, and well exposed with diagonal-line sculpture. Mouth small, vertical, and projectable. Lower jaw well protractile. Nostril one pair. Teeth absent. Branchiostegal membrane divided from isthmus. Gill rakers elongated and flat. Lateral line complete and continuous, running down from upper margin of gill opening to 29th or 32nd dorsal-fin ray, subsequently almost straight parallel with lower half of body axis. Dorsal-fin base greatly long and continuous. Dorsal-fin origin anterior to center of eye but not reach anterior edge of orbit. First dorsal-fin crest with 4 or 5 rays and second dorsal-fin crest with a single ray (broken in both specimens), subsequently rays distinctly short. No anal fin. Pectoral fin located posteroventrally to head, level to 14th or 19th dorsal-fin ray (broken in both specimens). Pelvic fin thoracic, level to pectoral-fin insertion (broken in both specimens). Skins without scales, covered by dermal tubercles in whole body but distinctly larger on upper part of body and lower part behind anus.

Color when fresh: body silver pinkish gray with forehead bluish gray, ocular globe with iris creamy and pupil black. Dorsal, pectoral and caudal fins red, pelvic fin creamy gray. Longitudinal series of irregular dark markings 3–5 along the sides (see Figure 2).

Color in alcohol: whole body pale brown with longitudinal series of irregular dark markings, ocular globe become dark iris and white pupil.

Distribution: Circum-global in tropical and temperate waters except northern Atlantic Ocean and Mediterranean Sea (Castro-Aguirre *et al.*, 1991 (as *Regalecus kinoi*); Roberts, 2012; Angulo and López-Sánchez, 2017; Feeney and Lea, 2018; Lee *et al.*, 2023), including Andaman Sea, Thailand (this study).

DNA barcoding. The COI fragment of the sample in this study matched perfectly with that of *R. russellii* (NC_081042–unspecified location, OP979107–China). The sample also revealed similarity with other samples of *R. russellii* as well as some of *R. glesne* (AP012973, JF931947), which were possibly mistakenly identified (suggested by Lee *et al.*, 2023). Therefore, the sample was excluded from the construction of a phylogenetic tree. Genetic distance and the phylogenetic tree revealed great genetic differentiation between the two *Regalecus* species, which formed own monophyletic group (Table 2, Figure 4). Within *R. russellii* clade, there were two groups, i.e. one group belonging to the populations found in Taiwan and South Korea and the other from the Andaman Sea, China, and unspecified location.

DISCUSSION

Roberts (2012) revised the taxonomy of the genus Regalecus and provided a taxonomic key to species based on larger juveniles, subadults and adults that distinguished R. russellii from R. glesne in having fewer than 82 dorsal-fin rays to vent (vs. 90-120), 34-37 abdominal vertebrae (vs. 45-56), 113-122 total vertebrae in an intact fish (vs. 127-163), 333-371 total dorsal rays (vs. 414–449), 47–60 gill rakers on first gill arch (vs. 34–47) and first dorsal-fin crest with 3-6 rays united by fin membranes and second dorsal-fin crest with a single ray only not united by membrane to other dorsal-fin rays (vs. first dorsal-fin crest with 6–8 rays united by fin membranes, and second dorsal-fin crest with 5-11 rays with no fin membrane connecting them to each other or to other dorsal-fin ray). Other characteristics of the specimens were also congruent well with R. russellii in having head depth 18.6-19.9 % SVL, post-orbital length 57.1-60.2 % HL, 65-76 dorsal-fin rays to vent, 51-53 total gill rakers on first gill arch, 34–37 abdominal vertebrae, and first dorsal-fin crest with 4–5 rays united by fin membrane and second dorsal-fin crest with a single ray. However, the maxillary length 33.9–39.2 % HL of the examined specimens overlaps with that of R. glesne (36.4–44.4) (Table 1). Previous records of large fish ($\geq 1,500$ mm SL) revealed almost completely overlapping morphometric and meristic measurements between the two species (Grieg, 1887; Waite, 1899; Chávez-Ramos et al., 1985; Heemstra, 1986; Castro-Aguirre et al., 1991; Salazar-Hermoso et al., 1999; Psomadakis et al., 2008; Schmitter-Soto, 2008; Dulčič et al., 2009; Ruiz and Gosztonyi, 2010; Roberts, 2012; Angulo and López-Sánchez, 2017; Lee et al., 2023; Galasso et al., 2024). However, there are five characters that can distinguish R. russellii from R. glesne including head depth in % SVL (16.0–17.6 vs. 11.5–14.3), post-orbital length in % HL (53.0–62.0 vs. 49.6), dorsal-fin rays to vent (58–82 vs. 84–120), gill rakers on first gill arch (46-60 vs. 34-47) and abdominal vertebrae (34-37 vs. 49?-52). Noticeably several specimens of regalecids in fish collections were reported as incomplete specimens, mostly left side of the anterior haft of body (Psomadakis *et al.*, 2008; Roberts, 2012). This is impossible to compare differences of two species by using the proportion of standard length (SL) as conduct in general teleost fish. The snout to vent length (SVL) may be useful instead of the standard length for morphometric characters on anterior part of body.

The DNA barcoding of these two specimens grouped them with *R. russellii* from China and showed slight differences from samples found in Taiwan and South Korea. The genetic distance clearly differentiated *R. russellii* and *R. glesne* (Lee *et al.*, 2023). Based on the morphological and genetic data, the specimens captured in the Andaman Sea of Thailand are identified as *R. russellii*.

The specimens of *R. russellii* were considered rare findings from the Indian Ocean. Most individuals of this species have been reported from the Atlantic and Pacific Oceans (24 and 43 individuals, respectively), whereas only 3 individuals were known from the Indian Ocean (Roberts, 2012; Angulo and López-Sánchez, 2017; Feeney and Lea, 2018; Lee *et al.*, 2023). Although *R. russellii* was recorded from Thailand by Roberts (2012) based on a specimen deposited at the Chao Mae Lim Ko Niau Shrine in Pattani Province, southern Thailand, that specimen was actually collected from Sumatra near Singapore, Indonesia, Indian Ocean. The two specimens in this study, collected from Andaman Sea, represent the first record of *R. russellii* in Thai waters.

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