

Tritonia iocasica sp. nov., a new tritoniid species from a seamount in the tropical Western Pacific (Heterobranchia: Nudibranchia)*

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Abstract During an expedition to a seamount at Caroline Plate in the tropical Western Pacific, a new species of *Tritonia* was sampled from the upper bathyal zone at depth of 970–1 262 m. This new species, *Tritonia iocasica* sp. nov., represents the first tritoniid nudibranch known to feed on octocoral of the family Melithaeidae. The species is up to 120 mm in length, pinkish in color; with the rhinophoral sheath divided into several petaliform extensions; veil with about 18 elongate digitiform processes; notal margin with 17–19 pairs of secondary gills; anus located below the 5th and 6th secondary gills, and the genitalia below the 3rd secondary gill on the right side of the body. Based on these external features, *T. iocasica* sp. nov. can be clearly distinguished from all previously described members of the genus. Phylogenetic analyses of two mitochondrial (COI, 16S rRNA) and a nuclear (H3) genes using Bayesian inference, maximum likelihood, and species delimitation analysis also support the separation of *T. iocasica* sp. nov. from its congeners.

Keyword: Tritoniidae; Mariana Trench; upper bathyal zone; Melithaeidae

1 INTRODUCTION

The genus *Tritonia* Cuvier, 1798 is a relatively diverse group of tritoniid nudibranchs that currently includes about 27 accepted species (MolluscaBase, 2021). The genus has a worldwide distribution, ranging from the Atlantic and Mediterranean (Marcus, 1983; Gosliner and Ghiselin, 1987; Silva et al., 2014; Trainito and Doneddu, 2014; Chimienti et al., 2020; Ortea and Moro, 2020), eastern Pacific (Bertsch, 2014; Valdés et al., 2018), Western Pacific (Baba, 1969; Smith and Gosliner, 2003), to Antarctica (Wägele, 1995; Schrödl, 2003; Ballesteros and Avila, 2006), from the intertidal zone to abyssal depths (2 550 m, *T. nigromaculata* Roginskaya, 1984). Most species are known or suspected specialist predators on octocorals, often limited to a single species or group of species of a single family (Smith and Gosliner, 2003; García-Matucheski and Muniain, 2011; Trainito and Doneddu, 2014; Chimienti et al., 2020). The genus has been relatively well studied in

the Atlantic Ocean, with nearly 20 species described. In contrast, however, the fauna in the Pacific region, especially in the Western Pacific, remains poorly studied, with only four described species.

Over the recent years, increasing explorations to deep-sea benthic ecosystems have yielded several novel species to the genus *Tritonia* (e.g. Valdés et al., 2017, 2018; Chimienti et al., 2020; Ortea and Moro, 2020), indicating that the knowledge of deep-water fauna of the genus is still incomplete. In 2019, the Institute of Oceanology, Chinese Academy of Sciences (IOCAS) conducted a scientific cruise to a

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seamount at the Caroline Plate in the tropical Western Pacific. During this investigation, three individuals of a sea slug were collected. Examinations of their external morphology, radular features, and gross internal anatomy indicated that they represent an undescribed species belonging to the genus *Tritonia*. Examination of gut contents revealed several coral fragments and sclerites that belong to the coral genus *Melithaea* Milne Edwards, 1857. This is the first record of a tritoniid species feeding on corals of the family Melithaeidae Gray, 1870. Phylogenetic analyses based on two mitochondrial (COI, 16S rRNA) and a nuclear (H3) genes using Bayesian inference, maximum likelihood, and species delimitation analysis also support the placement of the new species in the genus *Tritonia*, and its separation from related congeners.

2 MATERIAL AND METHOD

2.1 Specimens sampling and preservation

Specimens were photographed and then collected from the upper bathyal zone (970–1 262 m) during two dives of the ROV *Faxian* (mother ship R/V *Kexue*) (IOCAS) at a seamount near the Mariana Trench (Fig. 1a). The specimens were photographed alive on board. One specimen was preserved in 99.5% ethanol (here designated as holotype MBM285105) and two others were frozen at -80 °C for genomic study.

In the course of genomic study, one of the two specimens frozen at -80 °C was triturated thoroughly after dissecting its jaws, radula, and digestive gland. The jaws, radula, and digestive gland, together with a photograph of the living animal, are herein designated as paratype MBM285106 as a whole. The other specimen was entirely subjected to the genomic study, with no taxonomical observation and no gene fragments (i.e. COI, 16S, and H3) sequenced.

The holotype and paratype have been deposited at the Marine Biological Museum of Chinese Academy of Sciences (MBMCAS).

2.2 Morphological examination

External morphology and internal anatomy were observed under a Zeiss Stemi SV 11 Apo stereomicroscope. Drawings of the digestive and reproductive systems were made with the aid of a Zeiss camera Lucida. For scanning electron microscopy (SEM) studies, jaws and radula were extracted from buccal bulb by gross dissection, cleaned using 10% NaOH, rinsed in distilled water,

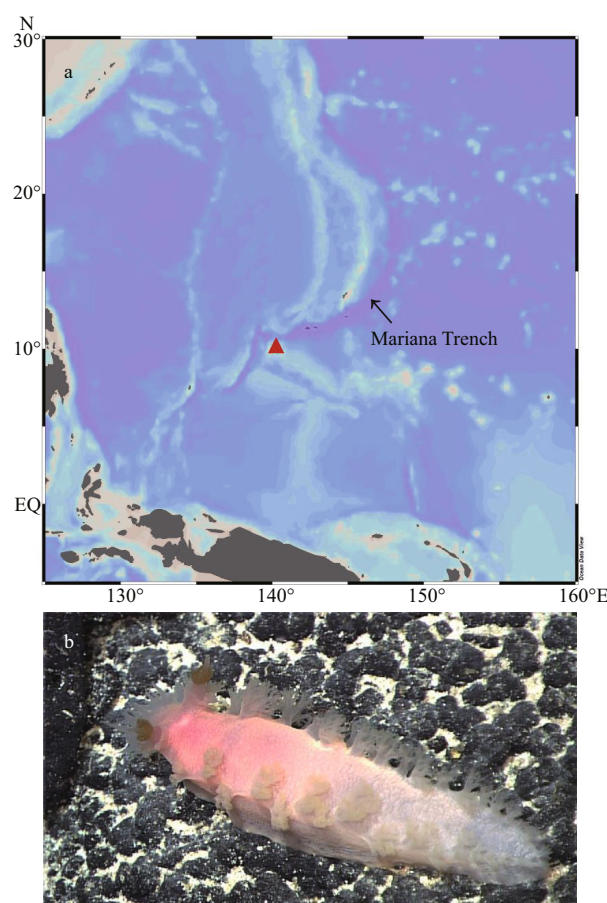


Fig.1 Geographical location of the type locality (a, solid red triangle) and the living specimen (paratype MBM285106) of *Tritonia iocasa* sp. nov. on a basalt rock at the type locality (b)

air-dried, coated with gold, and examined with SEM at an accelerating voltage of 5kV. Gut content and coral fragments were treated with undiluted commercial bleach in a centrifugal tube to obtain sclerites. These were washed several times with distilled water, transferred to aluminum stubs, coated with gold, and examined with Hitachi S-3400N SEM at an accelerating voltage of 5kV.

2.3 Molecular analyses

2.3.1 Extraction, amplification, and DNA sequencing

Holotype and paratype (see below) were selected for molecular studies. Genomic DNA from each individual was extracted with the Column Genomic DNA Isolation Kit (Beijing TIANGEN, China) according to the manufacturer's instructions. DNA was eluted in elution buffer and stored at -20 °C until use. PCR reactions were carried out in a total volume of 50 µL, including 2-µL DNA template, 1.5-mmol/L MgCl₂, 0.2 mmol/L of each dNTPs, 1 µL of both

Table 1 PCR primers used to amplify regions of the mitochondrial COI, 16S genes, and nuclear H3 gene

		Sequence of primers 5'→3'	Annealing temp. (°C)	Reference
COI	LCO1490 (F)	GGTCAACAAATCATAAAGATATTGG	42	Folmer et al., 1994
	HCO2198 (R)	TAAACT TCAGGGTGACCAAAAAATCA		
16S	16Sar-L (F)	CGCCTGTTTATCAAA AACAT	45	Palumbi, 1996
	16Sbr-H (R)	CTCCGGTTTGAACCTCAGATCA		
H3	HexAF (F)	ATGGCTCGTACCAAGCAGACGGC	45	Colgan et al., 1998
	HexAR (R)	ATATCCTTGGGCATGATGGTGAC		

forward and reverse PCR primers, 10× buffer and 2.5-U Taq DNA polymerase. Thermal cycling was performed under the following conditions: 94 °C for 3 min (initial denaturation), followed by 35 cycles of 94 °C for 30 s (denaturation), 40 s at primer-specific annealing temperatures, 72 °C for 60 s (extension) and a final extension at 72 °C for 10 min. PCR and sequencing primers for COI, 16S rRNA, and H3 genes are listed in Table 1. PCR products were verified on a GelRed-stained 1.5% agarose gel and purified with the Column PCR Product Purification Kit (Shanghai Sangon, China).

2.3.2 Phylogenetic analyses

Two specimens (MBM285105 and MBM285106) were subjected to molecular analysis. In addition, other sequences of *Tritonia* spp. were retrieved from GenBank and used for phylogenetic analyses (Table 2). We followed Korshunova and Martynov (2020) to include other tritoniid species in the analyses for comparison, and have used *Pleurobranchaea meckeli* (De Blainville, 1825) to root the tree.

Sequence alignments were generated with the MAFFT algorithm (Kato and Standley, 2013). COI and H3 sequences were translated into amino acids for confirmation of the alignment. Separate analyses were conducted for COI (567 bp), 16S (413 bp), H3 (288 bp), and concatenated data (1 268 bp). Ambiguously aligned fragments of 16S alignment was removed using Gblocks (Talavera and Castresana, 2007) with the following parameter settings: minimum number of sequences for a conserved/flank position (53/53), maximum number of contiguous non-conserved positions (8), minimum length of a block (10), allowed gap positions (with half). Maximum likelihood phylogenies were inferred using IQ-TREE (Nguyen et al., 2015) under Edge-unlinked partition model for 20000 ultrafast (Minh et al., 2013) bootstraps, as well as the Shimodaira-Hasegawa-like approximate likelihood-ratio test (Guindon et al., 2010). Bayesian Inference phylogenies were inferred

using MrBayes 3.2.6 (Ronquist et al., 2012) under partition model (2 parallel runs, 5 000 000 generations), in which the initial 25% of sampled data were discarded as burn-in. The best-fit models of evolution (GTR+I+G for COI and 16S, GTR for H3) were determined using PartitionFinder2 (Lanfear et al., 2017), with greedy algorithm and the Akaike information criterion AIC (Akaike, 1998). The *p*-distances within and among each species grouping were estimated with MEGA X (Kumar et al., 2018).

To discriminate species, we included 27 COI sequences for analyses based on the Kimura 2-parameter model. Bayesian inference (BI) was conducted using the software MrBayes v. 3.2.6 (Ronquist et al., 2012) based on the GTR+I+G model. The Metropolis-coupled Monte Carlo Markov chains were run for 5 000 000 generations, with sampling every 100 generations; split frequencies were less than 0.01 before the analyses were terminated. A 25% burn-in was applied before constructing the majority-rule consensus tree. Results were visualized using FigTree V. 1.4.3.

2.4 Species delimitation analysis

The Automatic Barcode Gap Discovery (ABGD) method (Puillandre et al., 2012) was used to assess the number of *Tritonia* species studied herein. The alignment from the fast-evolving COI gene was uploaded to the online server of ABGD. The analysis was performed with the model of Jukes-Cantor (JC69) with the following settings ($P_{\min}=0.001$, $P_{\max}=0.1$, steps=10, $X=1.0$, Nb bins=20).

3 RESULT

3.1 Phylogenetic analyses

Partial sequences (COI and H3) were obtained from the holotype (MBM285105) and paratype (MBM285106), while that of 16S was from the holotype only (see Table 2 for accession numbers). The two consensus trees inferred using Bayesian

Table 2 List of species and GenBank accession numbers of sequences used in the present study

Species	Voucher	GenBank Accession number			Reference
		COI	16S	H3	
<i>Tritonia iocasica</i> sp. nov.	MBM285105	MW086830	MW423688	MW424437	This study
<i>Tritonia iocasica</i> sp. nov.	MBM285106	MW086831	–	MW424438	This study
<i>Tritonia challengeriana</i>	CASIZ171177	HM162718	HM162643	HM162550	Pola and Gosliner, 2010
<i>Tritonia challengeriana</i>	CASIZ189419	KP153310	KP153277	KP153343	Hulett et al., 2015
<i>Tritonia exsulans</i>	ZMMU: Op-720	MW139259	MW144281	–	Korshunova and Martynov, 2020
<i>Tritonia exsulans</i>	–	KP764765	KP764765	–	Sevigny et al., unpublished
<i>Tritonia festiva</i>	CASIZ186478	KP153291	KP153258	–	Hulett et al., 2015
<i>Tritonia festiva</i>	SRR1950941	KX889748	MK100994	–	Goodheart et al., 2017
<i>Tritonia hombergii</i>	ZMMU: Op-724	MW139258	MW144280	MW158319	Korshunova and Martynov, 2020
<i>Tritonia tetraquetra</i>	SIO-BIC M12395	MH756139	MH756134	MH756145	Valdés et al., 2018
<i>Tritonia tetraquetra</i>	ZMMU:Op-719	MW139257	MW144279	–	Korshunova and Martynov, 2020
<i>Tritonia</i> cf. <i>psoloides</i>	CASIZ181055	KP153304	KP153271	KP153337	Hulett et al., 2015
<i>Tritoncula bayeri</i>	CPIC01540	–	MN162697	MN162696	Ekimova et al., 2019
<i>Tritoncula hamnerorum</i>	CASIZ181095	KP153292	KP153259	KP153325	Hulett et al., 2015
<i>Tritoncula hamnerorum</i>	CASIZ181090	KP153293	KP153260	KP153326	Hulett et al., 2015
<i>Tritoncula pickensi</i>	CASIZ175718	–	HM162642	HM162549	Pola and Gosliner, 2010
<i>Tritoniella belli</i>	N31D	GU227111	GU227002	–	Heimeier et al., 2010
<i>Tritoniopsis elegans</i>	CASIZ69928	KP153314	KP153281	KP153347	Hulett et al., 2015
<i>Tritoniopsis frydis</i>	CASIZ181156	KP153311	KP153278	KP153344	Hulett et al., 2015
<i>Marianina rosea</i>	CASIZ175746	HM162733	HM162656	HM162565	Pola and Gosliner, 2010
<i>Marionia abrahamorum</i>	MB28005053	MH892390	MH892386	MH892392	Silva et al., 2019
<i>Marionia arborescens</i>	CASIZ177578	HM162722	HM162646	HM162554	Pola and Gosliner, 2010
<i>Marionia blainvillea</i>	CASIZ176812	HM162721	HM162645	HM162553	Pola and Gosliner, 2010
<i>Marionia distincta</i>	CASIZ173317	HM162725	HM162648	HM162557	Pola and Gosliner, 2010
<i>Marionia elongoviridis</i>	CASIZ173308	HM162724	–	HM162556	Pola and Gosliner, 2010
<i>Marionia levis</i>	CASIZ192357A	KP153284	KP153251	KP153317	Hulett et al., 2015
<i>Tochuina gigantea</i>	ZMMU:Op-726	MW139260	MW144282	MW158321	Korshunova and Martynov, 2020
<i>Tochuina nigrigris</i>	LACM 3553	MH756138	MH756133	–	Valdés et al., 2018
<i>Pleurobranchaea meckeli</i>	–	FJ917499	FJ917439	EF133470	Göbbeler and Klussmann-Kolb, 2010

– means no data.

inference (BI) and maximum likelihood (ML) criteria were generally congruent (Figs.2 & 3). The genus *Tritonia*, including the new species, was recovered as monophyletic with strong support (posterior probability, PP=1; bootstrap value, BS=89). The analyses based on concatenated dataset (COI, 16S, and H3) showed that *T. iocasica* sp. nov. form a separate clade within the genus.

The BI analysis based on the concatenated COI, 16S and H3 markers does not solve (PP=0.62) the relationships among *T. iocasica* sp. nov., *T. challengeriana*, and the remaining *Tritonia* spp. included in this study, although the monophyly of the

genus is recovered. The phylogenetic tree inferred using BI criteria based on single gene COI (Supplementary Fig.S1) only support a joint relationship of *T. iocasica* sp. nov. and *T. challengeriana*, but does not recover the monophyly of *Tritonia*.

On the other hand, the ML analysis based on the concatenated COI, 16S, and H3 markers support a joint relationship among *T. iocasica* sp. nov. with *T. challengeriana*. These two species clustered together form a clade sister to other *Tritonia* spp. (BS=89).

These results support the systematic placement of

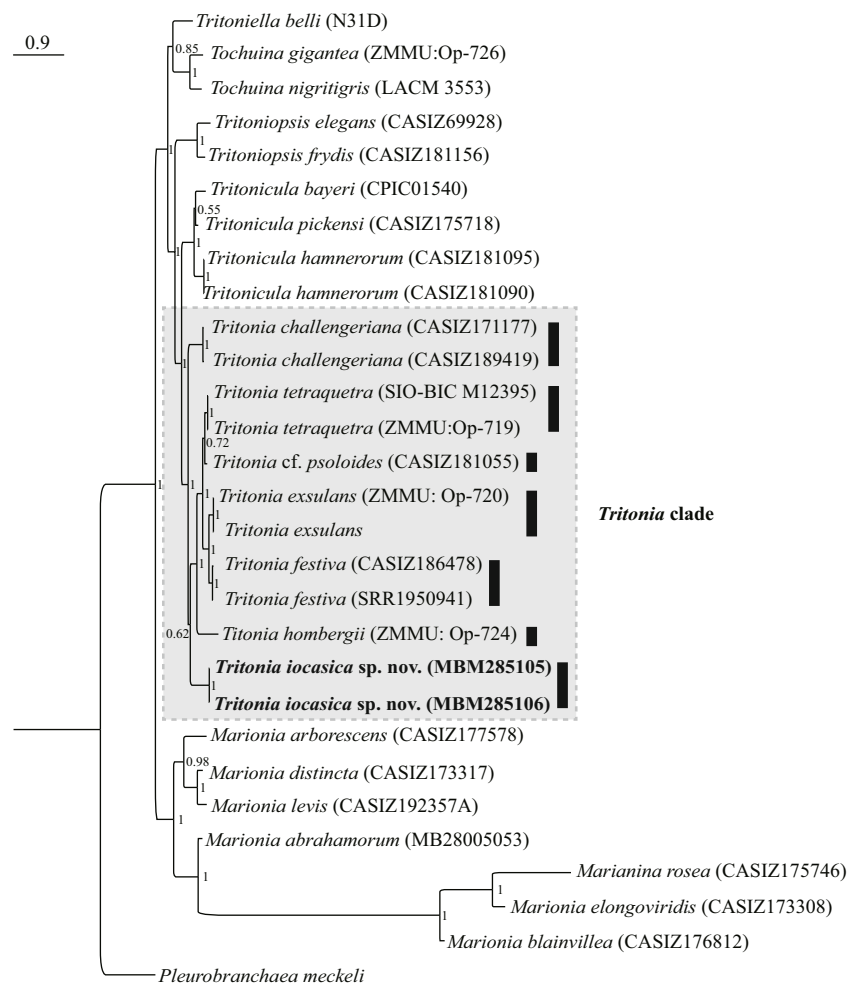


Fig.2 Phylogenetic tree inferred by Bayesian analysis (BI) based on concatenated dataset of COI, 16S, and H3 genes

Numbers adjacent to nodes refer to BI posterior probability (PP>0.5). PP≥90 was considered as enough support. Vertical bars indicate the results of ABGD species delimitation. Name of the new species are marked in bold.

the new species in the genus *Tritonia* and its separation from other congeners.

3.2 Species delimitation analyses

The ABGD analysis of the COI sequences resulted in the delimitation of seven species (Fig.2), with values of prior intraspecific divergence (P) being ≥ 0.001 . These groups correspond to clades recovered by BI and ML analyses. Within the genus, the two specimens of the new species are closest related to each other, with 0.3% pairwise distance; with the sequences studied herein, the pairwise distances among *T. iocasica* sp. nov. and other species of *Tritonia* are 17.8%–20.5% (Table 3).

These results provide additional support for the separation of *T. iocasica* sp. nov. from its congeners.

3.3 Systematics

Superfamily Tritonioidea Lamarck, 1809

Family Tritoniidae Lamarck, 1809

Genus *Tritonia* Cuvier, 1797

Type species: *Tritonia hombergii* Cuvier, 1803, subsequent designation by Lemehe (1961).

***Tritonia iocasica* sp. nov.** (Figs.1b, 4–6)

<http://zoobank.org/urn:lsid:zoobank.org:act:C29C82DB-08A5-490C-8512-3976CCD28132>

Material examined: Holotype MBM285105, live length 120 mm, dissected and sequenced, Station FX-Dive 212, (10°03'N 140°11'E) at depth of 1 262-m, 30 May 2019.

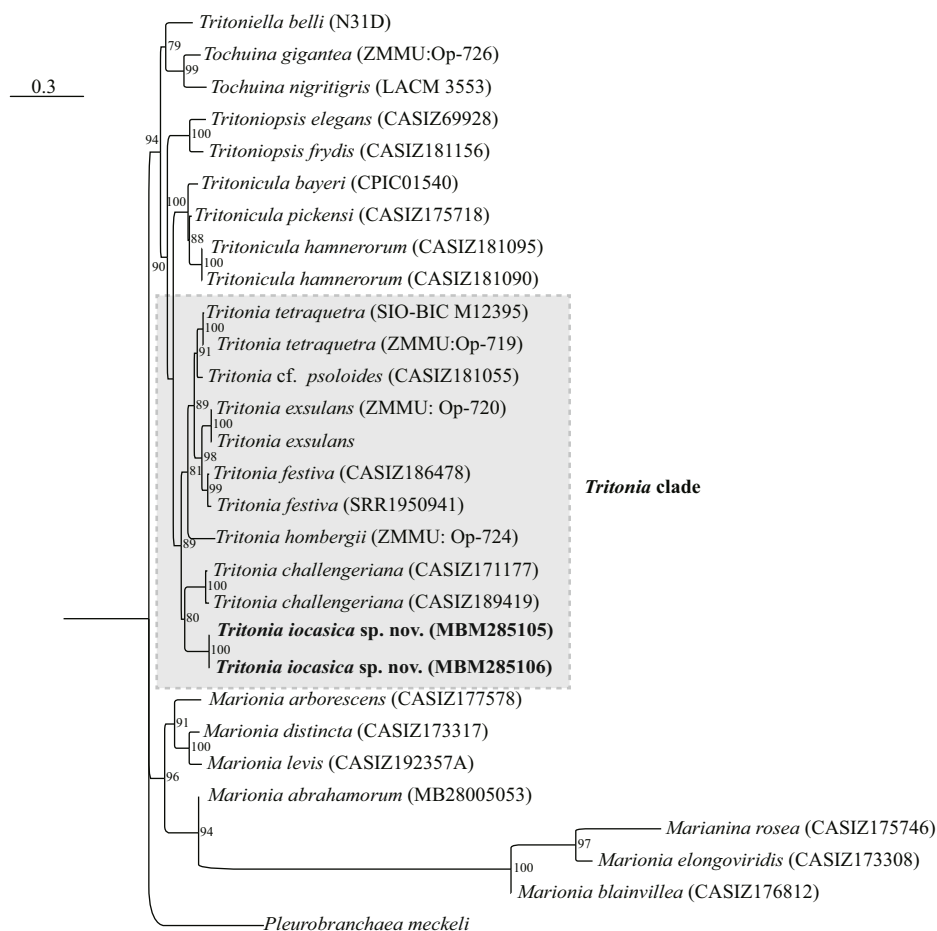
Paratype MBM285106, live length 120 mm, partly used for genomic analysis, with only jaws, radula, and digestive gland remaining for taxonomical study, Station FX-Dive 209, (11°36'N, 140°12'E) at depth of 970-m, 27 May 2019.

An additional specimen, live length 110 mm, entirely used for the genomic study, with no taxonomical observation and no gene fragments

Table 3 Estimates of *p*-distances of mitochondrial COI gene among *Tritonia* species and studied sequences

		Maximum distance between and minimum distance within species						
		<i>T. iocasica</i>	<i>T. challengeriana</i>	<i>T. exsulans</i>	<i>T. festiva</i>	<i>T. hombergii</i>	<i>T. tetraquetra</i>	<i>T. cf. psoloides</i>
<i>T. iocasica</i>	0.003							
<i>T. challengeriana</i>	0.178–0.186		0.029					
<i>T. exsulans</i>	0.193–0.195		0.179–0.187	0.003				
<i>T. festiva</i>	0.175–0.198		0.176–0.192	0.099–0.110	0.029			
<i>T. hombergii</i>	0.200–0.205		0.239–0.250	0.187–0.192	0.175–0.186	–		
<i>T. tetraquetra</i>	0.188–0.191		0.177–0.180	0.120–0.124	0.126–0.147	0.173–0.176	0.005	
<i>T. cf. psoloides</i>	0.181–0.182		0.173–0.175	0.122–0.124	0.131–0.138	0.178	0.080–0.087	–

Intraspecific distances in bold. – means no data.

**Fig.3** Phylogenetic tree inferred by maximum likelihood (ML) based on concatenated dataset of COI, 16S, and H3 genes

Numbers adjacent to nodes refer to ML bootstrap scores (BS>50). BS≥70 was considered as enough support. Name of the new species are marked in bold.

sequenced. Collected together with the paratype.

Type locality: A seamount at the Caroline Plate near the Mariana Trench, 10°03'N, 140°11'E.

Etymology: The new species is named after IOCAS (Institute of Oceanology, Chinese Academy of Sciences), in celebration of its 70th anniversary.

Description:

EXTERNAL ANATOMY (Figs.1b, 4a–c, paratype

MBM285106): Body elongate, slender. Dorsum covered with weak reticulate pattern of low ridges, giving dorsal surface a fish-scale appearance. Velum rounded, with about 18 elongate digitiform velar tentacles (Fig.4b). Notal margin with 17–19 pairs of secondary gills. Rhinophores long, located on anterior end of notum, bearing a cluster of pinnate processes that surrounds cylindrical apical papilla. Rhinophoral

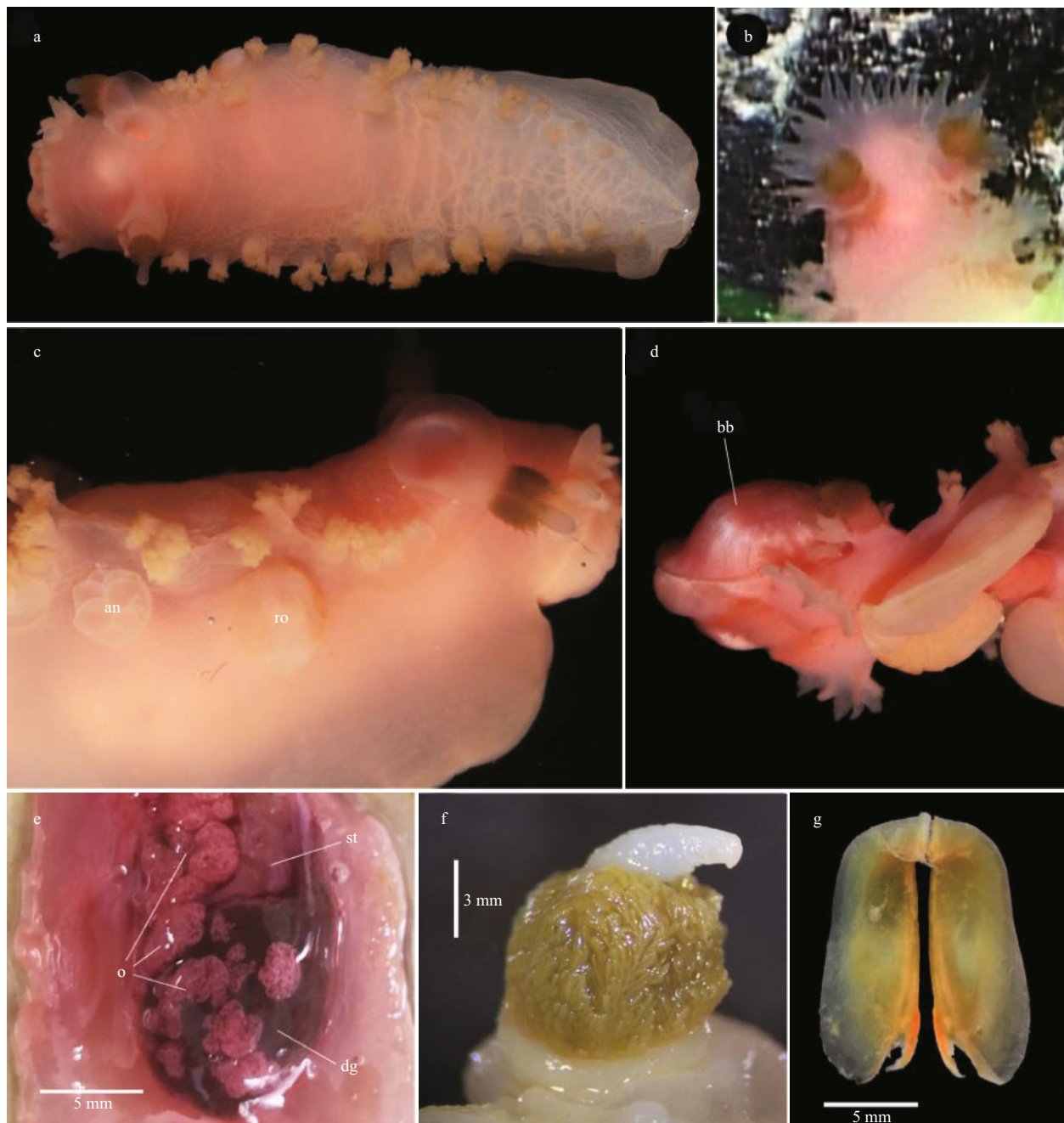


Fig.4 *Tritonia iocasica* sp. nov.

a–e & g. from the paratype (MBM285106); f. from the holotype (MBM285105) (a. fresh specimen on board, dorsal view; b. dorsal view of the velum; c. right view of anterior end; d. left view of anterior end, with anterior part of digestive tract evaginated; e. ventral view of partial visceral mass; f. rhinophores; g. jaws). Abbreviation: an: anus; bb: buccal bulb; dg: digestive gland; o: ovotestis; ro: reproductive opening; st: stomach.

sheath wide, divided into several (~10) petaliform lobes. Foot smooth, anterior end rounded, gradually tapering posteriorly. Genital, anal opening on the right side of body (Fig.4c). Anus situated below the 5th and 6th secondary gills, genital opening below 3rd secondary gills. Background color of living animal uniformly pinkish (same color as prey) except for pinnate processes on rhinophores which is brownish. Lighter pink on posterior 2/3 of the notum, secondary

gills, and velum.

INTERNAL ANATOMY: Digestive system (Fig.5a, holotype MBM285105) with thick, muscular buccal bulb (Figs.4d & 5a). Esophagus wide, anterior part connecting into buccal bulb. Posteriorly, it expands into a wider tube connecting to the stomach. The stomach oval in shape and its posterior part embedded ventrally into the digestive gland. Two long salivary glands connecting with the buccal bulb at

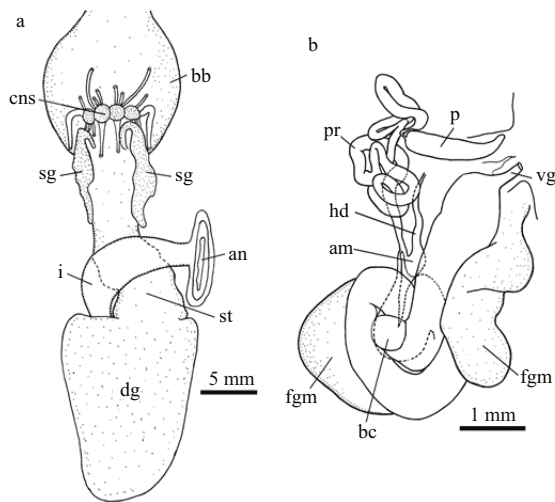


Fig.5 *Tritonia iocasica* sp. nov., holotype, MBM285105

a. digestive system; b. reproductive system. Abbreviations: an: anus; am: ampulla; bb: buccal bulb; bc: bursa copulatrix; cns: central nervous system; dg: digestive gland; fgm: female gland mass; hd: hermaphrodite duct; i: intestine; p: penis; pr: prostate; sg: salivary gland; st: stomach; vg: vagina.

each side of the esophageal junction. Intestine emerging dorsally from left side of digestive gland, forming simple loop towards right side of body, where it opens into the anus. Stomach simple, no stomach plates present. Digestive gland reddish brown, large (Fig.4e). Jaws (Figs.4g, 6a–c, paratype MBM285106) thin, elongate, amber-colored, inner margin (masticatory margin) terminating distally in small pointed process. The masticatory margin apparently worn at upper end; 1/3 lower part with about 18 or more rows of rodlets. Maximum length of rodlets about 200 μ m in length, and maximum width about 40 μ m. Most rodlets with irregularly pentagonal or hexagonal cross section. Radula (Fig.6d–h, paratype MBM285106) with a formula of 62 \times 100.1.100. Rachidian teeth tricuspid, with nearly rectangular bases. Central cusp sharply pointed and longer; outer cusps blunt and shorter, fused to margin of base. Distinct, deep incision between central cusp and right outer cusp (Fig.6f). First lateral teeth with thicker base, sturdy, hoodlike cusps (Fig.6g). Remaining lateral teeth sickle-shaped, with sharp cusps, becoming thin towards outer margins of radula (Fig.6h).

Reproductive system (Fig.5b, holotype MBM285105) androdiaulic. Ootestis covering most of surface of digestive gland, uniformly pinkish (Fig.4e). Ampulla short, simply convoluted, opening on female gland mass where prostate connects. Prostate narrow, elongate, forming several loops and ending into narrow deferent duct, which connects with conical penial sac. Penis simple, elongate.

Vagina elongate, broad, opening into the spherical bursa copulatrix.

Diet: Intestinal tract contained coral fragments and calcareous sclerites (Fig.7) of an unidentified coral species, *Melithaea* sp. (Melithaeidae).

4 DISCUSSION

The systematics of Tritoniidae has long been considered problematic despite the revisions by Odhner (1936, 1963) and Marcus (1983). Relationships among the genera are still unclear and the placement of some species remain tentative (Willan, 1988; Schrödl, 2003). The fact that previous authors utilized different characters to distinguish genera may account for the problem. Nevertheless, recent phylogenetic studies also showed that most tritoniid genera currently recognized are not monophyletic (Bertsch et al., 2009; Almón et al., 2018). A revision of the whole family based on more detailed anatomical information and molecular evidence is therefore needed to define generic boundaries (Korshunova and Martynov, 2020). Gosliner and Ghiselin (1987) treated all species of tritoniids with well developed secondary gills, with a tricuspid rachidian tooth, and without stomach plates as members of *Tritonia*. In the present study, we follow this classification, to assign the new species to the genus *Tritonia*. This assignation was supported by molecular evidence. In the phylogenetic tree reconstructed herein (Figs.2 & 3), *T. hombergii*, the type species of the genus, together with other *Tritonia* spp., was recovered as a monophyletic clade (PP=1, BS=89). The trees based on concatenated data and single COI gene all showed that *T. iocasica* sp. nov. form a separate clade sister to other *Tritonia* spp. The species delimitation and *p*-distance analyses strongly support the separation of *T. iocasica* sp. nov. from its congeners. Within the genus, *T. iocasica* sp. nov. is unique by its external morphology and distinct color pattern. At present, no other known species, to our knowledge, can be confused with the new species. In the Western Pacific region, there are only four other *Tritonia* species that have been described (Table 4), including *T. bollandi* Smith and Gosliner, 2003 from Okinawa, Japan and Indonesia (58–88-m depth), *T. festiva* (Stearns, 1873), an intertidal species native to Pacific coast of North America but also occurs in Japan Sea (Baba, 1969), *T. olivacea* Bergh, 1905 from Indonesia (32-m depth) and *T. tetraquetra* (Pallas, 1788) from Northwestern Pacific (1–700-m depth). *T. iocasica* sp. nov. thus represents the first of

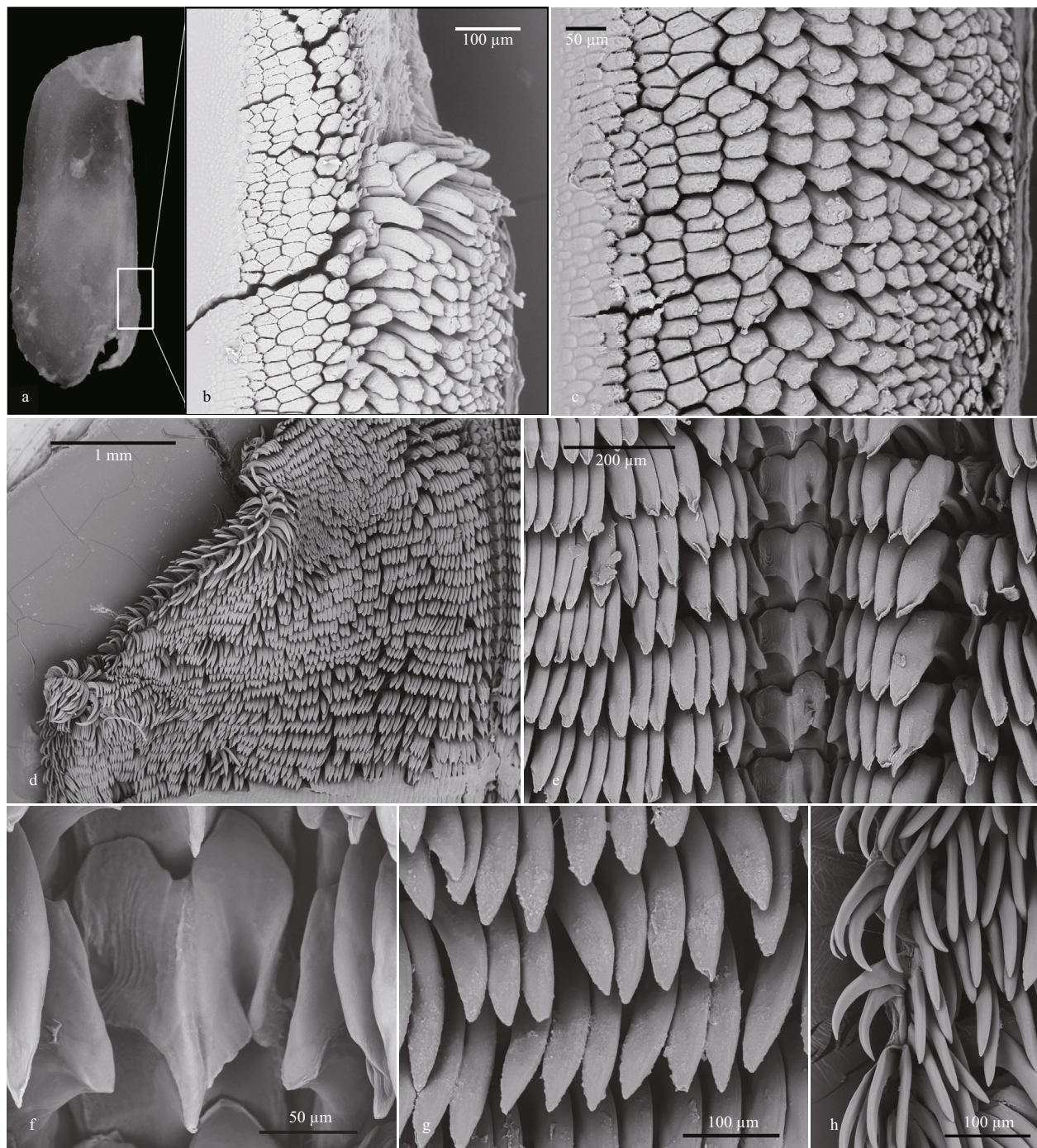


Fig.6 Micrographs of jaw and radula of *Tritonia iocasica* sp. nov., specimen MBM285106

a. left jaw; b, c. jaw masticatory border; d. general view of the left part of radula; e. rachidian and inner lateral teeth; f. rachidian and the first lateral teeth; g. middle lateral teeth; h. outermost lateral teeth.

Tritonia known from deep sea of the tropical Western Pacific, and, to our knowledge, the first for the family Tritoniidae. Unlike in the Atlantic and the eastern Pacific, the taxonomic knowledge of the genus in the Western Pacific is very poor, although several additional undescribed species were reported and illustrated by Gosliner et al. (2008, 2018). This

probably results from the fact that deep-sea fauna of the Western Pacific was relatively poorly investigated. Another explanation is that many recorded *Tritonia* species were unidentified or misidentified, as previous taxonomic studies were based primarily on few external characters that always result in taxonomic confusions and in turn, an inaccurate

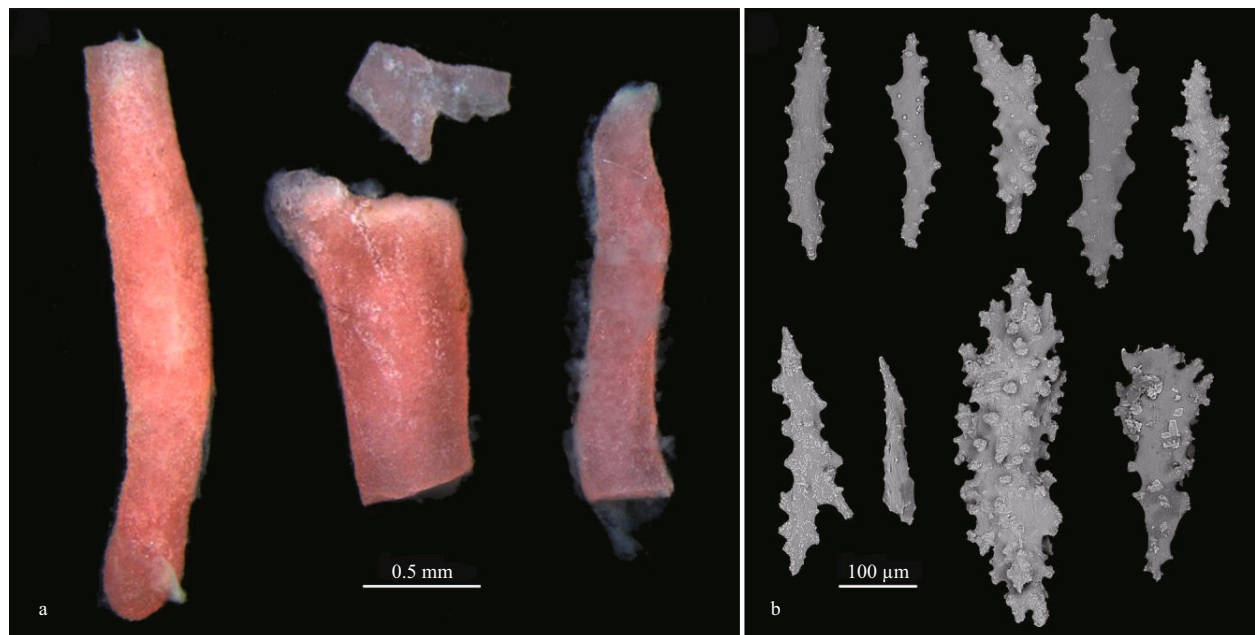


Fig.7 Coral fragments belonging to *Melithaea* sp. dissected from intestinal tract of specimen MBM285106 (a) and different forms of sclerites of coral (b)

Table 4 Comparison of the main characteristics of some related species of *Tritonia*

Species	Velar processes	Rhinophore sheath	Branchial plumes (pair)	Radula formula	Rachidian tooth	First lateral tooth	Penis	References
<i>T. challengeriana</i> Bergh, 1884	10–18	Smooth	≤30	31–46×37–63.1.1.1.37–63	Base broad; central cusp prominent	Broad, low	Digitiform to conical	Wägele, 1995; Schrödl, 2003
<i>T. bollandi</i> Smith and Gosliner, 2003	12–14	Undulate and flange-like	9–14	66×78.1.1.1.78	Base transversely wide; three cusps strong, triangular	Buttressed with a distinct peg-like tip	Filiform	Smith and Gosliner, 2003
<i>T. festiva</i> (Stearns, 1873)	8–12	Smooth	8–12	32–57×16–49.1.1.1.16–49	Base transversely wide; three cusps strong, triangular	Broad, with a dorsal ridge or projection	Conical, with an annulation near the apex	Baba, 1969; McDonald, 1983; Gosliner and Ghiselin, 1987
<i>T. olivacea</i> Bergh, 1905	12–16	Undulate and flange-like	9–15	70–80×35–80.1.1.1.35–80	Base roughly rectangular; three cusps strong, triangular	Oblique rhomboidal with an inner blunt spine	–	Baba, 1937
<i>T. tetraquetra</i> (Pallas, 1788)	>30	Bi- to tri-pinnate plumose	–	40–73×150–50.1.50–150	Base roughly rectangular; central cusp strong, outer cusp small	Broad, hooked	–	Korshunova and Martynov, 2020
<i>T. callogorgiae</i> Chimienti, Furfaro & Taviani, 2020	6	Simple chalice-shaped	4–5	50×110.1.1.1.108.	Base rectangular; central cusp short, outer cusp obsolete	Narrow, hooked	Simple, digitiform	Chimienti et al., 2020
<i>T. iocasica</i> sp. nov.	18	Petaliform	17–19	62×100.1.100	Base nearly rectangular; central cusp pointed, outer cusp blunt	Narrow, hooded	Simple, conical	This study

– means no data.

estimate of biodiversity. In central Pacific, an individual likely belonging to the genus was photographed by ROV *Deep Discoverer* (NOAA) from Musicians Seamounts (see <https://www.ncei.noaa.gov/waf/oceanos-animal-guide/Gastropoda030.html>). This individual is highly

similar to *T. iocasica* sp. nov. in coloration pattern, body shape, numbers of secondary gills and velar tentacles, and thus may be conspecific with the new species described herein. If this is the case, it would extend the distribution of *T. iocasica* sp. nov. to the central Pacific.

Internally, *T. iocasica* sp. nov. has a jaw armed with multiple rows of rodlets (Fig.5b & c). This type of denticulation has been only recorded for a single species in the genus (Chimienti et al., 2020). In the whole family, only two other species, *Marionia bathycarolinensis* Smith and Gosliner, 2005 and *Marionia tedi* Ev. Marcus, 1983, were known to have a jaw armed with rows of rodlets (Smith and Gosliner, 2005; Valdés, 2006). Compared to these three species, the rodlets in the new species is much longer. The type of this denticulation may facilitate the distribution of stress that might damage the thin, delicate inner edge of the jaw during cutting the coral stem.

Examination of the digestive tract of *T. iocasica* sp. nov. revealed that the gut contains several coral fragments and calcareous sclerites that belong to *Melithaea* sp. The three specimens studied herein were collected from a single seamount near the Mariana Trench during two ROV dives. Two of the specimens (Fig.1b) were discovered crawling on a platform of basalt rock. The entire area is partially colonized by primnoid octocorals, flabelliid hexacorals, the hydroid *Stegolaria* sp., and phoronematid sponges. Two or three groups of *Melithaea* sp. (Melithaeidae) were observed (not collected) from distance of about 1.5–2 m. The diet of the *Tritonia* species was reviewed by García-Matucheski and Muniain (2011); the members exclusively feed on octocorals, especially on the gorgonians, often limited to a single species or group of species within a single family. The present study represents the first record of predation by a tritoniid species on octocoral of family Melithaeidae, which expands our knowledge of the predation range of *Tritonia*.

The majority of *Tritonia* species live in intertidal or shallow subtidal waters, and only five have been reported down to 500 m (Bergh, 1899; Bouchet, 1977; Roginskaya, 1984; Valdés et al., 2017, 2018). Until now, the evolutionary history of species in *Tritonia* is still little known. Actually, it is difficult to make speculation as to the time of the *Tritonia* species originated from shallow water or deep sea, due to lack of a fossil record for these shell-less mollusks.

5 DATA AVAILABILITY STATEMENT

The authors declare that all the data supporting the findings of this study are available within the article.

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The data and samples were collected onboard R/V

Kexue. We would like to express our sincere thanks to the crews for their assistance during the survey. We are very grateful to Prof. Kuidong XU and Dr. Xuwen WU for their efforts in collecting the specimens. Dr. Yang LI identified the coral and helped in conducting the SEM study of the coral sclerites. Four anonymous reviewers provided valuable comments and suggestions that greatly improved the manuscript.

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Electronic supplementary material

Supplementary material (Supplementary Fig.S1) is available in the online version of this article at <https://doi.org/10.1007/s00343-021-0408-3>.