

# Bipolarity in sea slugs: a new species of *Doridunculus* (Mollusca: Nudibranchia: Onchidoridoidea) from Antarctica

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**Abstract** Bipolar distributions of benthic taxa have intrigued many biologists since the first Antarctic expeditions. Records of taxa, either at species or higher taxonomic levels, encompassing this peculiar distribution have been regularly reported since then. Moreover, the study of heterobranch mollusks from remote areas, such as Antarctica, often provides key taxa so far unknown, which might be essential for systematics. Here, we describe a new species of nudibranch from the eastern Weddell Sea using micro-computed tomography (micro-CT), namely, *Doridunculus punkus* n. sp. (Akiodorididae). Hitherto, the genus *Doridunculus* was only reported from the Arctic region. We provide an extensive description of morphological and anatomical characters of *D. punkus* n. sp., thereby offering new insights into the anatomy of the understudied Akiodorididae. A comparison of all Akiodorididae species shows that *D. punkus* n. sp. exhibits intermediate characters between some Onchidoridoidea families (i.e., hook-shaped innermost lateral teeth). Furthermore, the detailed study of its reproductive system suggests a close relationship of both Akiodorididae and Goniodorididae. We

suggest an Antarctic origin of Akiodorididae followed by dispersion and a subsequent speciation in the Arctic.

**Keywords** Akiodorididae · Phanerobranchia · Heterobranchia · Weddell Sea · Disjunct distribution · Micro-CT

## Introduction

A disjunct distribution of sister taxa covering the northern and southern hemispheres is a phenomenon known as bipolarity (Stepanjants et al. 2006). Bipolar distributions can occur either at the species, genus, or higher taxonomic levels (Allcock and Griffiths 2015). In mollusks, approximately 30% of living Antarctic bivalve and gastropod families are characterized by a bipolar distribution, including heterobranch genera such as *Philine* Ascanius, 1772 and *Toledonia* Dall, 1902 (Rudman 1972; Warén 1989; Dell 1990; Crame 1993). The wide fossil record of mollusks suggests at least three paleontological periods in which bipolar and/or amphitropical (i.e., on both sides of the tropics) events occurred, Late Jurassic (~150 Mya), Paleogene-Neogene (~23 Mya), and Neogene-Pleistocene (~2.6 Mya; Crame 1993). Current disjunct distributions might be the result of transequatorial dispersal during glacial maximum cooling or, alternatively, a prior cosmopolitan species isolated vicariantly in high latitudes during interglacial periods (Allcock and Griffiths 2015). Vicariant cases imply that species once placed in the tropics might have sheltered in deep waters during interglacial periods, a phenomenon called equatorial submergence (Stepanjants et al. 2006). This is applicable for *Philine* for example, which is distributed in deep waters of all oceans (OBIS 2016). However, knowledge of deep equatorial species is often lacking, probably due to the scarce sampling done so far in deep tropical waters (Allcock and Griffiths 2015).

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Antarctic glacial and interglacial periods during the early Cenozoic (~65 Mya) triggered species migration toward warmer temperatures in lower latitudes or in deep-sea shelters (Thatje et al. 2005). The Antarctic final breakup during the late Cenozoic allowed the formation of the Antarctic Circumpolar Current (~25 Mya). This thermal and hydrographic barrier hampered marine organisms' dispersion from north to south and above 1000 m depth at the Southern Ocean (SO; Barker and Thomas 2004). However, deep-sea organisms from the Weddell Sea may have dispersed through the Antarctic Bottom Water, which flows as part of the global thermohaline circulation system (Stepanjants et al. 2006; Pawlowski et al. 2007). Thereby, organisms with high dispersal capabilities may have colonized distant regions, either by planktonic larvae or by attaching to floating debris (Raguá-Gil et al. 2004), even contemporarily (Stepanjants et al. 2006; Pawlowski et al. 2007; Jun et al. 2012).

Wägele et al. (2008) suggested that basal members of some major Nudibranchia lineages may have an Antarctic origin. Several families and genera are only found in the Southern Ocean, and these might be crucial for understanding the evolution of heterobranch lineages. Nudibranchia, with approximately 35 described species from Antarctica, is the most speciose heterobranch lineage (De Broyer et al. 2016). Among them, the Onchidoridoidea Gray, 1827 of the SO are understudied phanerobranch dorids, characterized for being unable to fully retract their gills. This taxon comprises five families, namely, Akiodorididae Millen & Martynov, 2005; Calycidorididae Roginskaya, 1972; Corambidae Bergh, 1871; Goniodorididae Adams & Adams, 1854; and Onchidorididae Gray, 1827 (WoRMS Editorial Board 2015). Onchidoridoidea were formerly assigned to Suctorina due to the possession of a buccal pump (Bergh 1892). Akiodorididae is considered to be a basal family within Onchidoridoidea, presently related to Goniodorididae based on the reproductive system (Hallas and Gosliner 2015). In fact, Akiodorididae was recently established and includes genera previously placed in Goniodorididae and Onchidorididae (Millen and Martynov 2005). The family Akiodorididae currently contains five genera, all of them restricted to polar waters. *Akiodoris* Bergh, 1892 confined to the North Pacific, *Armodoris* Minichev, 1972 from the SO, and *Doridunculus* Sars, 1878 from the North Pacific and North Atlantic, each having two described species. *Echinocorambe* Valdés & Bouchet, 1998 in the Norwegian Sea and *Prodoridunculus* Thiele, 1912 from the Davies Sea are both monotypic. *Prodoridunculus* was described based on juvenile specimens, and it was suggested to be the senior synonym of *Armodoris antarctica* Minichev, 1972 (Millen and Martynov 2005). The synapomorphies of Akiodorididae are a smooth, thin, lip disk; two or more inner lateral teeth; and rectangular, reduced outer lateral teeth (Millen and Martynov 2005). When present, the rachidian tooth ranges from a small plate to a wide arch-

shaped structure, sometimes with a central cusp (Millen and Martynov 2005). The inner lateral teeth have a strong cusp directed downward, with several denticles along the margins. The marginal teeth are rectangular and progressively decrease in size toward the edge of the radula (Millen and Martynov 2005). Externally, these species have a spiculated notum covered by tubercles. The gills are dorsal and arranged in a semi-circle, with the anus lying posteriorly; gills are not found in a pocket as in Onchidorididae (Hallas and Gosliner 2015). However, in *Echinocorambe*, gills are reduced to one simple leaf and the anus is ventral (Valdés and Bouchet 1998).

The genus *Doridunculus* was first described by Sars (1878) from Risvær (Norway) at 100-m depth based on a single specimen, named *D. echinulatus* Sars, 1878. The species presents two dorsal keels lying side by side, and its dorsum is covered by conical spiculated tubercles. Later, Odhner (1907) described *D. pentabrancheus* Odhner, 1907, which was found from muddy bottoms of Skagerrak, Norway, at 335-m depth. This species differs from the type species only by having five gills instead of three. Therefore, Odhner (1922) himself further considered *D. pentabrancheus* as a probable junior synonym of *D. echinulatus*. Decades later, a new species, named *D. unicus* Martynov & Roginskaya, 2005, was described from abyssal waters (3000–3620 m) of the Sea of Japan (Martynov and Roginskaya 2005). Contrary to *D. echinulatus*, *D. unicus* lacks the dorsal keels and presents more gills, larger oral tentacles, and a rachidian tooth. Presently, these are the only described species of *Doridunculus*, both inhabiting exclusively the northern hemisphere (Martynov and Roginskaya 2005).

Here, we describe *Doridunculus punkus* n. sp., collected in the Antarctic eastern Weddell Sea, based on a single specimen collected in Austasen at 228-m depth in 2003. Since the analysis of a potentially new nudibranch species and its description require dissection, radula preparation, and examination of internal anatomy, usually, at least one specimen is destroyed in doing so. In order to save the holotype, we performed X-ray micro-computed tomography scans followed by 3D reconstruction to describe the new *Doridunculus* species. Micro-CT analysis allows non-destructive investigation of type material from regions that are difficult to survey, as is the case here (see Moles et al. 2016a). Moreover, we provide a comparative anatomical description of *Doridunculus* and the other Akiodorididae genera. A discussion on the bipolar distribution of this family is also included and extended to other examples found in the literature.

## Material and methods

### Sample collection

One specimen of *D. punkus* n. sp. was collected in the eastern Weddell Sea (Antarctica) during the Antarctic cruise ANT

XXI/2 (November 2003–January 2004) of RV Polarstern (Alfred Wegener Institute, Bremerhaven, Germany). The specimen was collected with Agassiz trawl in Austasen at 228-m depth (PS65/280–1), later photographed alive, and transferred to 70% ethanol for morphological analysis.

### Morphological analysis

For micro-CT analysis, *D. punkus* n. sp. was contrasted with 1% iodine metal (I<sub>2</sub>) dissolved in 100% ethanol (I<sub>2</sub>E) for 24 h. Subsequently, the specimen was transferred to a pipette tip containing 100% ethanol, in which the specimen was arrested. The pipette tip was then mounted on a pin with superglue. Three X-ray tomography scans were performed with an XRadia Micro XCT-200 (Carl Zeiss X-ray Microscopy Inc.). For the macro-scan, the 0.4× object lens unit was used, at 40 kV and 200 μA, with a pixel size of 15.35 μm. For scanning the anterior body region, the 4× object lens unit, at 90 kV and 88 μA, was used, with a pixel size of 3.70 μm. For the micro-scan of the radula and spicules, the 10× object lens unit, at 40 kV and 200 μA, was used with a pixel size of 2.02 μm. All scans were performed by using Binning 2 (summarizing 4 pixels) and subsequently reconstructed by using Binning 1 (full resolution) as given in Sombke et al. (2015). Tomography projections were then reconstructed using the reconstruction software XMReconstructor software (Carl Zeiss Microscopy GmbH), resulting in image stacks (Tiff format). We used the software platform Amira® 5.4. (FEL, Visualization Science Group) for image segmentation. Two graphical 3D PDF reconstructions of *D. punkus* n. sp. were performed using Deep Exploration. 3D PDFs can be opened in Adobe Acrobat Reader and activated by clicking on it (see Supporting Information).

## Results

### Systematics

Euctenidiacea Tardy, 1970.

Doridacea Thiele, 1912.

Onchidoridoidea Gray, 1827.

Akiodorididae Millen & Martynov, 2005.

*Doridunculus* Sars, 1878.

Type species. *Doridunculus echinulatus* Sars, 1878.

**Diagnosis of the genus** Color translucent white. Body elongated, notum rounded anteriorly, posteriorly tapered, slightly shorter than tail. Velum not clearly separated from ventral notum side. Oral tentacles small, conical, or folded. Notal longitudinal keel(s) present or absent. Notum covered by numerous conical tubercles. Tail ridged. Gills dorsal, minute, 3 to 10, disposed in semicircle. Network of elongated spicules

visible through epidermis. Radular formula 1–10.1.(0–1).1.10–1 (with or without rachidian); inner lateral hook-shaped or with pronounced, big cusp pointing downward; rectangular marginal teeth forming interlocking pointed plates, decreasing in size toward edge.

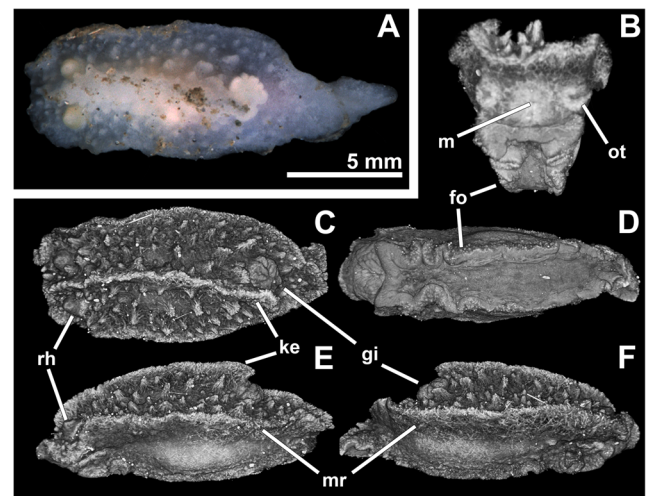
The genus contains only the following three species (Martynov and Roginskaya 2005): *D. echinulatus*, *D. unicus*, and *D. punkus* n. sp. described herein.

***D. punkus* n. sp.** (Figs. 1, 2, 3, 4, and 5) (See 3D PDF of the reconstructed anatomy of the whole specimen and a more detailed anterior part in Supplementary Material 1 and 2, respectively) <http://zoobank.org/NomenclaturalActs/69A41F49-FAEC-4A6F-ADD6-0657084A5745>.

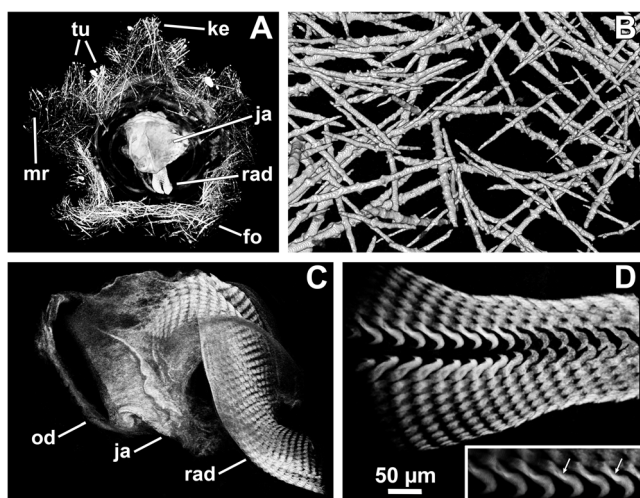
**Holotype** Adult specimen, 16 mm. Austasen, eastern Weddell Sea (71° 7.15' S, 11° 26.23' W) collected by Agassiz trawl on 29 December 2003, at 228-m depth (PS65/280–1). Deposited in SNSB Zoologische Staatssammlung München (catalog number ZSM Moll 2016113).

**Type locality** Austasen, eastern Weddell Sea (Antarctica).

**External morphology** (Figs. 1 and 2a, b) Live animal measured 16 mm (Fig. 1a), when preserved 15 × 6 × 6 mm (length:width:height). Color translucent white, brownish digestive gland seen postero-laterally by transparency. Body high, elongated, pentagonal in transverse section (Fig. 1b); mantle rim protruding; oral veil rounded, slightly folded laterally; oral tentacles grooved; notum rounded posteriorly (Fig. 1c); foot narrow, notched anteriorly, tapering posteriorly, mid-dorsal ridge present in tail (Fig. 1d). Posterior part not covered by notum. Rhinophores bearing seven diagonal

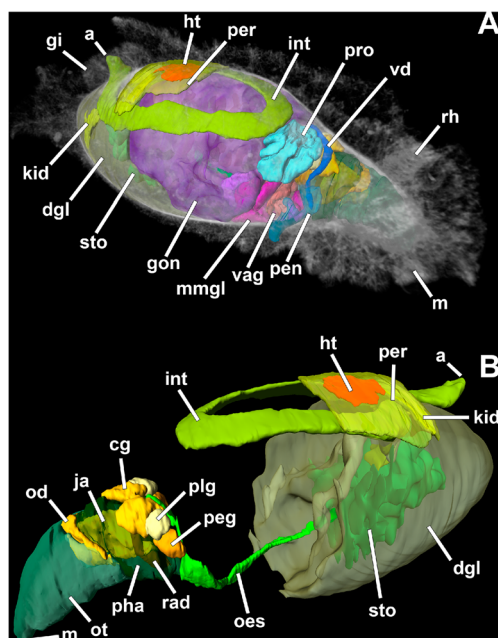


**Fig. 1** Live photograph (a) of *Doridunculus punkus* n. sp. and volume rendering (b–f) of the preserved animal. a Dorsal view. b Frontal view. c Dorsal view. d Ventral view. e Left-lateral view. f Right-lateral view. fo foot, gi gills, ke keel, m mouth, mr mantle rim, ot oral tentacles, rh rhinophores

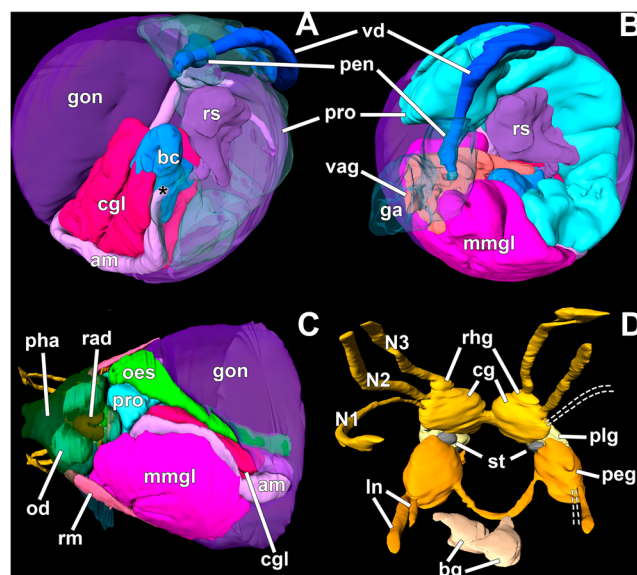


**Fig. 2** Micro-CT photographs of sclerotized structures of *Doridunculus punkus* n. sp. **a** Transverse section of the cephalic region showing spicules' arrangement in the body wall. **b** Detail of the notal spicules. **c** Lateral view of the buccal mass. **d** Distal part of the radula (approximate scale); close-up showing inner lateral teeth, note longitudinal denticles (white arrows). *fo* foot, *ja* jaw, *ke* keel, *mr* mantle rim, *od* oral disk, *rad* radula, *tu* tubercles

lamellae, retractile within smooth cavities. Notum dorsally keeled, starting right in front of rhinophores, extending posteriorly to gills, forming a protuberance tilted toward left side of gills (Fig. 1e); average height of keel about 2 mm. Dorsal

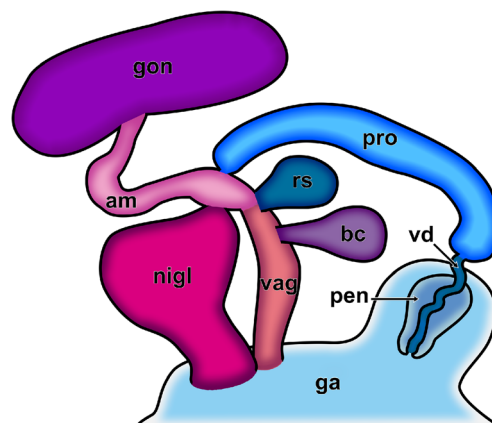


**Fig. 3** Macro-scan (0.4 $\times$ ) reconstruction of *Doridunculus punkus* n. sp. **a** Right-lateral, general view of all reconstructed organs. **b** Left-lateral view of the digestive system. *a* anus, *cg* cerebral ganglia, *dgl* digestive gland, *gi* gills, *ht* heart, *int* intestine, *ja* jaws, *gon* gonad, *kid* kidney, *m* mouth, *mmgl* membrane + mucus glands, *od* oral disk, *oes* oesophagus, *ot* oral tube, *pen* penis, *peg* pedal ganglia, *per* pericardium, *pha* pharynx, *plg* pleural ganglia, *pro* prostate, *rad* radula, *rh* rhinophore, *sto* stomach, *vag* vagina, *vd* vas deferens



**Fig. 4** Micro-CT reconstruction of the anterior body region of *Doridunculus punkus* n. sp. (4 $\times$ ). **a** Antero-lateral view of the reproductive system; membrane + mucus glands and vagina not depicted; asterisk showing connection between distal ampulla and prostate. **b** Frontal view of the reproductive system. **c** Ventral view of digestive, nervous, and reproductive systems. **d** Frontal view of the nervous system; missing nerves are depicted with white-dashed lines. *am* ampulla, *bc* bursa copulatrix, *bg* buccal ganglia, *cg* cerebral ganglia, *cgl* capsule gland, *ga* genital atrium, *gon* gonad, *ln* lateral nerves, *mmgl* membrane + mucus glands, *N1* oral nerve, *N2* labial nerve, *N3* rhinophoral nerve, *od* odontophore, *oes* oesophagus, *pen* penis, *peg* pedal ganglia, *pha* pharynx, *plg* pleural ganglia, *pro* prostate, *rad* radula, *rhg* rhinophoral ganglia, *rm* retractor muscles, *rs* receptaculum seminis, *st* statocyst, *vag* vagina, *vd* vas deferens

notum heterogeneously covered by pointed, conical, spiculated papillae; smaller in notal periphery and at sides of dorsal keel (Fig. 1c). Gills five, shortly pinnate, arranged in semicircle (Fig. 1a, c, f); anal papilla small, placed just behind gills. Genital pore small, situated antero-laterally beneath



**Fig. 5** Schematic outline of the reproductive system of *Doridunculus punkus* n. sp. *am* ampulla, *bc* bursa copulatrix, *ga* genital atrium, *gon* gonad, *nigl* nidamental glands (capsule, membrane, and mucus glands), *pen* penis, *pro* prostate, *rs* receptaculum seminis, *vag* vagina, *vd* vas deferens

notum rim. Spicules conspicuous, fusiform, irregularly multi-knobbed, sometimes with median crown of knobs (Fig. 2a); densely lying within notum, imparting rough texture, forming dense network toward epidermis (Fig. 2b).

**Digestive system** (Figs. 2c, d, 3, and 4c) Mouth lying ventrally, opening at base of oral veil, right in front of foot edge, in vertical furrow. Oral tube pyriform, surrounded by follicular oral glands. Oral disk thin, smooth, cuticular; demarcating transition into pharynx. Pharynx bulbous ventrally; two thick longitudinal muscles arise from postero-lateral part reaching gonad laterally (Fig. 4c). Jaws ear-shaped, without distinct ornamentation. Odontophore placed at rear part of pharynx. Radular formula  $41 \times 10.1.0.1.10$  (Fig. 2c, d). Innermost lateral hook-shaped, with one longitudinal denticle at middle part (see white arrows in Fig. 2d), flattened at base. Marginal teeth rhomboid in shape, presenting outer pointed cusp; decreasing in size toward lateral rim. Rachidian not observed. Salivary glands paired, saccular, small; lying behind pharynx, connected to it through small, thin, tubular ducts. Oesophagus thin, originating from posterior part of pharynx; passing through nerve ring, widening toward ventral side, reaching stomach far posteriorly; partly covered by gonad. Stomach completely surrounded by holohepatic digestive gland, situated in posterior third of viscera (Fig. 3a, b). Digestive gland presenting reticulated pattern, more pronounced toward periphery. Intestine thin, originating mid-dorsally from stomach, widening, becoming compressed, forming pronounced loop to right side, returning toward left side, ending in anal papilla.

**Reproductive system** (Figs. 4a–c and 5) Triaulic. Gonad placed in middle region of body in front of digestive gland; globular in appearance, presenting up to 60 follicles. Gonoducts large connecting in front of gonad, converging into thin, tubular ampulla. Ampulla lying on gonad, extending ventrally (Fig. 4a). Vas deferens originating from distal gonoduct close to ampulla, leading into big, folded prostate; prostate making counterclockwise loop alongside body wall, connecting dorsally to thick distal vas deferens, proximally making short loop (Fig. 4b). Penis small, short, apparently unarmed; placed inside large, globe-shaped, muscular, penial sheath, lying close to genital atrium in right antero-lateral side of body (Fig. 4a, b).

Vaginal duct originating from proximal oviduct, close to ampulla forming a very short uterine duct, entering saccular receptaculum seminis. Bursa copulatrix saccular, roundish, originating from thin duct distally from vagina, placed in central transverse position. Vagina relatively short, opening into genital atrium. Oviduct short, placed at distal gonoduct leading to capsule gland. Capsule gland extending far posteriorly into gonad. Membrane and mucus glands not clearly distinct from each other, embracing capsule gland, extending latero-

ventrally close to body wall (Fig. 4c). See overall schematic outline of reproductive system in Fig. 5.

**Nervous system** (Fig. 4d) Cerebral ganglia large, interconnected by short commissure; laterally oral nerve (N1) reaching mouth; labial nerve (N2) reaching anterior part of oral tube; rhinophoral nerve (N3) connected through small rhinophoral ganglia to cerebral ganglia, innervating rhinophores. Neither optical nerve, optic ganglion, nor eyes detected. Pleural ganglia attached to cerebral ganglia, without visible connectives. Statocysts present on ventral side of pleural ganglia, close to pharynx. Pedal ganglia large, interconnected by long commissure; with two lateral nerves running to ventral side. Two buccal ganglia, small, neighboring, lying at base of salivary glands, partly surrounded by them.

**Circulatory and excretory systems** (Fig. 3) Pericardium flattened, placed at posterior part of body, above kidney, intestine, and digestive gland. Heart placed in longitudinal axis, no obvious distinction between auricle and ventricle. Aorta and blood gland not observed. Kidney lying below pericardium, extending back behind digestive gland; no nephroduct observed.

**Ecology** The single specimen of *D. punkus* n. sp. was found in a detritus-rich benthic bottom at 228-m depth. The community was dominated by sessile phyla, such as sponges (*Cinachyra*, *Clathria*, *Isodyctia*, *Iophon*, *Tedania*), bryozoans (*Alcyonidium*, *Carbasea*, *Isoschizoporella*, *Notoplites*, *Reteporella*), gorgonians (*Thouarella*, *Primnoisis*), ascidians (*Aplidium*, *Cnemidocarpa*), and pterobranchs (*Cephalodiscus*). Vagile fauna such as nudibranchs (*Doris*, *Doto*, *Tritonia*, *Tritoniella*), sea cucumbers, and polychaetes were also collected at the same station.

**Etymology** *D. punkus* n. sp. is named after the Mohican hairstyle of the punks, referring to the presence of one keel in the dorsum.

## Remarks

*D. punkus* n. sp. differs from its congeners by the small folded oral tentacles, rounded (instead of bilobed) posterior notum, and by having a notal rim not covering the foot. The new species presents a single dorsal keel, while *D. echinulatus* presents two keels, more or less separated (Sars 1878; Odhner 1907), and *D. unicus* lacks them (Martynov and Roginskaya 2005). Both *D. punkus* n. sp. and *D. echinulatus* lack a rachidian tooth. In *D. unicus*, the rachidian is present and it is similar to that of *Akiodoris* and *Armodoris* (Millen and Martynov 2005). A hook-shaped inner lateral tooth is an autapomorphy of the herein described species, not found in

any of the described species of Akiodorididae. The stomach of *D. punkus* n. sp. is completely enveloped by the digestive gland, while it is fully free in *D. unicus* (Martynov and Roginskaya 2005). Cerebral and pleural ganglia are separated in *D. punkus* n. sp., similarly to *Armodoris* (Millen and Martynov 2005). In *D. punkus* n. sp., eyes were not observed; this character was not specified in *D. echinulatus* (Sars 1878; Odhner 1907), while *D. unicus* possesses eyes (Millen and Martynov 2005).

## Discussion

*D. punkus* n. sp. is the first record of the genus in the southern hemisphere, since the two other species were only recorded previously in the North Atlantic and the Sea of Japan (Sars 1878; Martynov and Roginskaya 2005). It seems plausible that the whole family is restricted to either polar and/or deep waters. We were able to reconstruct both hard and soft tissues of the single specimen collected using micro-CT techniques. The new species is placed within the genus *Doridunculus* since it conforms with most of the characters of the hitherto described species (see Table 1). For instance, *D. punkus* n. sp. and its congeners exclusively possess a dorsal ridge on the tail. Their notum does not cover the foot posteriorly, as in *Armodoris* and *Prodoridunculus* (Minichev 1972; Valdés and Bouchet 1998). However, *D. punkus* n. sp. differs from congeners in having a rounded notum posteriorly, like in *Prodoridunculus*, and not bilobed as in *D. echinulatus*, *D. unicus*, and *Echinocorambe*. Moreover, *D. punkus* n. sp. presents a notal rim extending laterally forming flaps, as in all other Akiodorididae. A single dorsal keel similar in position and height to the two present in *D. echinulatus* is also a diagnostic character of the new species (named after it). The gills are arranged in a semicircle in all akiodoridids (Millen and Martynov 2005). The lack of a branchial pocket to withdraw the gills, a feature also typical for the new species, was considered a synapomorphy of Akiodorididae, separating this group from other Onchidoridoidea (Millen and Martynov 2005).

In the buccal apparatus, a thin and smooth lip disk is shared among akiodoridids. The radular structure is quite different in *D. punkus* n. sp.; it lacks a rachidian tooth, although a rudimentary thin plate could have possibly been unnoticed in the micro-CT scan. In fact, the other species lacking the rachidian teeth were not analyzed using scanning electron microscopy (SEM), i.e., *D. echinulatus* and *P. gaussianus* (Sars 1878; Thiele 1912); thus, this absence is not well documented (Martynov and Roginskaya 2005). Remarkably, *D. punkus* n. sp. is the first member of the Akiodorididae that possess a large hook-shaped inner lateral tooth, suggesting that multiple inner lateral teeth evolved once within Onchidoridoidea (Hallas and

Gosliner 2015). Indeed, the inner lateral tooth of most akiodoridids still presents a strong curved cusp, thus resembling a rudimentary hook. Likewise, the whole family seems to have square marginal teeth decreasing in size toward the outer edge. The radula of *D. punkus* n. sp., as well as many Goniodorididae taxa, is similar to that of the Polyceridae, although the latter are lacking the innermost reduced lateral tooth (e.g., Vallès et al. 2000). Therefore, Wägele and Willan (2000) postulated that the innermost lateral tooth of the Goniodorididae actually could represent the second lateral of the Polyceridae. This might also be the case in *D. punkus* n. sp.

Our tomographic analyses reveal that the pharynx of *D. punkus* n. sp. is bulged and forms a buccal pump, as for all Akiodorididae, although the presence of two wide longitudinal retractor muscles at each side of the pharynx was never reported in this family before. The stomach is completely enclosed by the digestive gland in *D. punkus* n. sp., contrary to *Akiodoris* and *Armodoris* (Millen and Martynov 2005) and probably *D. unicus* as depicted by Martynov and Roginskaya (2005), albeit not reported therein. The intestine is dorsal and forms a pronounced loop forward, as in all studied species of Akiodorididae. The anus is dorsal in all species, except for the aberrant *Echinocorambe brattegardii* (Valdés and Bouchet 1998), where it is ventral, and thus, strongly resembling the species of the genus *Corambe* (Martynov and Schrödl 2011). The digestive gland is overlapping the gonad in *Doridunculus*, *Akiodoris*, and *Armodoris* (Martynov and Roginskaya 2005; Millen and Martynov 2005; Valdés et al. 2011). However, only in *D. punkus* n. sp., the gonad is restricted to the mid-longitudinal section of the animal, which could be attributed to the ontogenetic stage. In the new species, the ampulla is thin and tubular and connects with a receptaculum seminis by a short uterine duct, similar to *Akiodoris* (Millen 1985). The ampulla leads to a vaginal duct which has a saccular bursa copulatrix placed distally. The prostate can be tubular and club-shaped, and it is always wide and voluminous in Akiodorididae. The penis is normally unarmed, except in *Akiodoris* and *Armodoris*, where it is densely covered with spines (Millen and Martynov 2005).

In conclusion, *D. punkus* n. sp. shares most of the characters with Akiodorididae. However, it also looks externally similar to *Aegires albus* Thiele, 1912 found in the same waters (Wägele 1987a). Both species are of similar color, size, and shape, presenting dorsal, irregularly scattered tubercles and spicules (Wägele 1987b). Nevertheless, *D. punkus* n. sp. presents a rather squared anterior part, the mantle rim protruding laterally, and a dorsal keel. On the other hand, the external appearance of the new species (i.e., elongated notum, trailing ridged foot) strongly resembles that of the genus *Diaphorodoris* Iredale & O'Donoghue, 1923 (Millen 1985). *Diaphorodoris* is presently assigned to the family Calycidorididae together with the Arctic monotypic genus *Calycidoris* Abraham, 1876 (Hallas and Gosliner 2015).

**Table 1** Comparative table of diagnostic characters of the Akiodorididae genera, including all the species of *Doridunculus*

	<i>Doridunculus punktus</i> n. sp.	<i>Doridunculus echinulatus</i> G.O. Sars, 1878	<i>Doridunculus unicus</i> Martynov & Roginskaya, 2005	<i>Akiodoris</i> Bergh, 1892	<i>Armodoris</i> Minichev, 1972	<i>Echinocorambe</i> Valdés & Bouchet, 1998	<i>Prodoridunculus</i> Thiele, 1912
Notum	Elevated	Elevated	Elevated	Elevated	Elevated	Flattened	Flattened
Rim	Expanded laterally	Expanded ventrally	Expanded ventrally	Expanded	Expanded ventrally	Expanded ventrally	Expanded ventrally
Tail	Not covered	Not covered	Not covered	Not covered	Covered	Not covered	Covered
Posteriorly	Rounded	Bilobed	Bilobed	Rounded	Rounded	Bilobed	Rounded
Dorsal tubercles	Conical	Elongated, conical	Elongated, cylindrical-conical	Elongated or rounded	Rounded	Elongated	Small and big, conical in four longitudinal rows
Ridge	One, mid-dorsal	Two, mid-dorsal	Absent	Absent	Absent	Absent	Absent
Spicules	Rod-like, tuberculate	Present	Rod-like, normally hollow	Rod-like or quadrate	Rod-like, straight	Absent	Present
Rhinophores	Conical	Large	Large	Conical	Short, wide	Conical	n.s.
Sheath	Smooth	Smooth	Few tubercles	Tuberculated	Tuberculated	Smooth	n.s.
Gills	5, semicircle	3–5, semicircle	6–10, semicircle	4–17, semicircle	5, semicircle	1	n.s.
Position	Dorsal	Dorsal	Dorsal	Dorsal	Dorsal	Ventral	n.s.
Shape	Pinnate	Pinnate	Unipinnate and bipinnate	Bipinnate, tripinnate, or quadripinnate	Unipinnate	Smooth	n.s.
Tentacles	Small, folded	Club-shaped	Large, flattened	Rounded or flap-like	Rounded	Large	n.s.
Radular formula	10.1.0.1.10	1–6.0.6–1	4–9.1.1.1.1.1.9–4	3–13.2.1.2.1.3–3	4–8.4.6.1.4–6.8–4	4.3.1.3.4	2.2.0.2.2
Rachidian	Absent	Absent	Trapezoidal, long cusp	Wide, arch-shaped, or very small	Plate-like, with or without central cusp	Cuspless plate	Absent
Inner lateral(s)	Hook-shaped	Inner cusp, 4–5 denticles	Inner cusp, 2–6 denticles	Inner cusp, 2–3 denticles	Inner rounded cusp, 2–4 denticles	Inner cusp, 3–4 denticles	Inner cusp, 3–4 denticles
Marginal	Squared, one large outer cusp	Squared, one large outer cusp	Squared, one large outer cusp	Squared	Inner cusp, 6–0 denticles	Squared, one large outer cusp	Squared, one large outer cusp
Stomach	Fully free from digestive gland	n.s.	Partly covered by digestive gland?	Fully free from digestive gland	Partly covered by digestive gland	n.s.	n.s.
Reproductive system	Triaulic	n.s.	Triaulic	Triaulic	Triaulic	n.s.	n.s.
Penis	Unarmed	n.s.	Unarmed	Armed	Armed	Unarmed	n.s.
Ampulla	Tubular	n.s.	Voluminous, club-shaped	Tubular	Tubular or voluminous bean-shaped	n.s.	n.s.
Gonad	Free, not covered by digestive gland	n.s.	Partly covered by digestive gland?	Partly covered by stomach and digestive gland	Partly covered by stomach and digestive gland	n.s.	n.s.
Bursa copulatrix	Saccular	n.s.	Flattened, saccular	Wide, saccular	Oval, large	n.s.	n.s.
Receptaculum seminis	Saccular	n.s.	Long and narrow	Small	Small or long	n.s.	n.s.
Cerebro-pleural ganglia	Separated	n.s.	Fused	Fused	Separated	n.s.	n.s.
Eyes	Not observed	n.s.	Present	Present	Present	Absent	n.s.
Distribution	Eastern Weddell Sea (Antarctica)	Norwegian Sea	Sea of Japan	Sea of Okhotsk and British Columbia (North Pacific)	Davies Sea, Ross Sea, and South Shetland Islands (Antarctica)	Norwegian Sea	Davies Sea (Antarctica)
Depth range (m)	228	80–100	3000–3620	10–780	25–40	2538–3016	n.s.
Reference	Present study	Sars 1878	Martynov and Roginskaya 2005	Millen and Martynov 2005, Valdés et al. 2011	Millen and Martynov 2005, Valdés et al. 2011	Valdés and Bouchet 1998	Thiele 1912

n.s. not specified

But, *D. punkus* n. sp., as well as all akiodoridids, lacks a semicontractible branchial pocket into which gills can be retracted, a feature typical for Calycidorididae and some Onchidorididae (Fahey and Valdés 2005; Martynov et al. 2009). Morphological and phylogenetic studies placed Akiodorididae as sister group of Goniodorididae, sharing a receptaculum seminis that is connected to the uterine duct (Hallas and Gosliner 2015). Instead, in Calycidorididae, the uterine duct is situated independently on the vagina, and in Corambidae and Onchidorididae, the receptaculum is twice connected to the vagina and the uterine duct (Millen 1985; Fahey and Valdés 2005; Millen and Martynov 2005; Martynov and Schrödl 2011). Overall, all these facts support the placement of the new species within Akiodorididae and altogether within Onchidoridoidea.

Our finding of a representative of *Doridunculus* in the SO strongly suggests a bipolar distribution of the genus. Current bipolar disjunct distributions may have been the result of periods of dispersal and/or vicariant isolation, which have occurred several times in Earth's history (Crame 1993). Meridional deep flows were stronger in periods of climate cooling and may have formed dispersion bridges of animals from temperate and cold zones of one hemisphere to another (Vinogradova 1997). Alternatively, a prior cosmopolitan distribution in cooler times may have caused vicariant isolation during interglacial periods (Crame 1993; Allcock and Griffiths 2015). Nudibranchs, as well as other heterobranch taxa, may have originated in Antarctica (Wägele et al. 2008; Göbbeler and Klussmann-Kolb 2010; Moles et al. 2016b). This assumption is based on the presence of basal members of several heterobranch lineages in these waters. Therefore, it is plausible to assume an Antarctic origin of Akiodorididae, and the later dispersion out of Antarctica in glacial periods, a common pattern observed for other different taxa, such as cnidarians, priapulids, polychaetes, amphipods, copepods, isopods, tanaidaceans, holothuroids, and ophiuroids (Vinogradova 1997; Stepanjants et al. 2006; Brandt et al. 2007; Clarke 2008; Allcock and Griffiths 2015). Glacial periods implied periods of environmental stress for Antarctic fauna, resulting in a dramatic decrease in diversity (Zinsmeister 1982). Shelf fauna was completely impoverished by grounded ice masses during glacial maxima, inducing the sheltering migration into marine oases (polynyas) and deep-sea waters (Thatje et al. 2005, 2008). Consequently, species could have migrated using deep-water gateways, such as the Antarctic Bottom Water, as a part of the global thermohaline circulation system (Stepanjants et al. 2006; Pawlowski et al. 2007). The lack of molecular data of Akiodorididae precludes answering the mechanisms of such distribution. Therefore, a thorough taxon sampling in remote areas and deep-sea waters as well as molecular clock analyses are essential for revealing the phylogeographic history, including origin, dispersion, and speciation of Akiodorididae.

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