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#### Research article

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# Systematic revision of the family Pleioplanidae Faubel, 1983 (Polycladida, Acotylea): new genus and combinations

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**Abstract.** New morphological information, reconsiderations and the first combination of a generic name based on a previously established species are presented in the current revision of the family Pleioplanidae (Polycladida, Acotylea). Species belonging to this family are briefly presented and *Laqueusplana bocki* gen. et sp. nov. is described. An identification key to all valid species of Pleioplanidae is provided. Furthermore, results from phylogenetic analyses of the species treated herein are discussed.

Keywords. Platyhelminthes, flatworms, polyclads, descriptions, key.

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## Introduction

The family Pleioplanidae belongs to the superfamily Leptoplanoidea within the suborder Acotylea (Order Polycladida). Faubel (1983) established Pleioplanidae based on the anatomical reconstruction of *Notoplana atomata* (O.F. Müller, 1776) and the inner organization of its prostatic vesicle, which is divided into numerous tubular chambers that are not directly attached to the central ejaculatory duct. The prostatic vesicle organization of *N. atomata* ("atomata-type") is considered the main apomorphy justifying the formation of Pleioplanidae. Therefore, *N. atomata* was designated as the type species of the family and renamed *Pleioplana atomata*.

The genus *Notoplana* Laidlaw, 1903 (Acotylea, Polycladida) is one of the largest and most revised genera of Polycladida. *Notoplana*, traditionally included within the family Notoplanidae, comprised more than 70 species prior to Faubel's (1983) revision. Following this revision, the genus was reduced to 34 species. As mentioned, one of Faubel's major actions was to combine and delimit a specific group of *Notoplana* species into the family Pleioplanidae.

Two attempts to reorganize the numerous species of *Notoplana* were made prior to Faubel (1983). The first attempt was carried out by Bock in 1913 who established three clades: group A with *Notoplana evansii* Laidlaw, 1903 as the main species, group B based on *N. atomata* (O.F. Müller, 1776) and group C with *N. alcinoi* (Schmidt, 1861) as the primary species. Later, Marcus & Marcus (1966) divided the genus into nine groups of species based on the presence or absence of nuchal tentacles and specific characteristics of the male copulatory system. However, tentacles in preserved specimens may be poorly defined or not visible or recognizable.

The family Pleioplanidae currently comprises 4 genera and 13 species: *Pleioplana* Faubel, 1983, the largest genus of the family, currently with eight valid species, *Melloplana* Faubel, 1983 with two species, *Izmira* Bulnes, 2010 with two species and *Persica* Maghsoudlou, Bulnes & Rahimian, 2015 with only one species.

The type species of the family Pleioplanidae, *P. atomata*, was first described by O.F. Müller (1776) as *Planaria punctata*, but subsequently designated as *Planaria atomata* in the original description. Later, Oersted (1844) included *Planaria atomata* within *Leptoplana*, but Bock (1913) reassigned this species to the genus *Notoplana*. Some *Notoplana* species described during the late nineteenth and the twentieth centuries (*Notoplana californica* (Plehn, 1898), *N. delicata* (Yeri & Kaburaki, 1918), *N. mortenseni* (Bock, 1913), *N. inquieta* (Heath & McGregor, 1912) and *N. megala* (Marcus, 1952)), were assigned to the genus *Pleioplana* by Faubel 1983. Recently, *P. bosphorensis* Bulnes, Kalkan & Karhan, 2009 and *P. okusi* Bulnes, Kalkan & Karhan, 2009 were placed in *Pleioplana*.

Furthermore, the genus *Melloplana* as described by Faubel (1983) comprises two species previously assigned to the genus *Notoplana*. Both species, *Melloplana ferruginea* (Schmarda, 1859) and *M. japonica* (Kato, 1937), are characterized by the presence of an unarmed male copulatory organ without a stylet. The other two genera of Pleioplanidae are *Izmira* (*I. cinari* Bulnes, 2010 and *I. turkeyi* Bulnes, 2010), which was recently described from the Bosporus (Turkey), and *Persica* (*P. qeshmensis* Maghsoudlou *et al.* 2015) described from the Persian Gulf in Iran.

The description of new genera and species in the family since its establishment in 1983 and the abundance of new morphological information necessitate this current revision. Here, we review known characters, add new characters and perform a phenetic analysis of the family Pleioplanidae, taking into account new combinations and diagnostic features.

#### Material and methods

The examined material includes specimens collected, with the aid of the Grupo de Estudo do Medio Mariño (GEMM), from the north-western coasts of Galicia (Spain), mainly from the Ría de Arousa. Specimens were collected mainly by scuba diving, covering a bathymetric range that extends from the intertidal zone to the maximum accessible depth for scuba diving at approximately 40 metres. Most species were found under stones and rocks or on algal stalks and empty mussel shells, all of which are characteristic habitats for polyclads. Specimens were first photographed in their natural surroundings, then collected by hand using a brush or net and stored in containers according to specimen size. One specimen of *Pleioplana atomata* found in the intertidal zone at Los Chalanos Beach, Muros de Nalón (Asturias, Spain), was also studied, as were specimens of *Pleioplana megala* collected from the northern coast of Ubatuba, São Paulo (Brazil), kindly provided by the Swedish Museum of Natural History.

Specimens were first anaesthetized in a 7.5% magnesium chloride solution, then fixed with temperate Bouin following the Newman & Cannon method (2003) or placed directly on frozen fixative (Bouin), which causes them to become lethargic and immobile, and thus easily positioned. As the solution melts, specimens become submerged in the fixative. Prior to fixation, a lateral piece of tissue was extracted from each specimen and stored in absolute ethanol intended for future molecular studies.

Specimens were subsequently embedded in paraplast, serially sectioned between 7 and  $10 \mu m$  and stained with AZAN (trichrome staining method). Reconstructions of the internal anatomy were derived from serial sagittal sections. Measurements were obtained from both living and preserved material.

In the present article, the literature was extensively revised. Using morphological information taken from this review and the analysis of new specimens, a list of morphological characters and states was compiled with the Delta software (Dallwitz *et al.* 1993) to produce a key, update the descriptions and generate the matrix (Table I) used for the phylogenetic analysis.

#### **Abbreviations used**

cg = cement glands

ed = ejaculatory duct

ev = external vagina

fp = female pore

iv = internal vagina

lv = Lang's vesicle

mp = male pore

ov = oviduct

ph = pharynx

pv = prostatic vesicle

s = sucker

st = stylet

sv = seminal vesicle

vd = vasa deferentia

#### **Character list**

- 1. Body shape: 1. elongated. 2. oval
- 2. Colour: 1. cryptic. 2. aposematic
- 3. Dorsal appearance: 1. translucent. 2. opaque
- 4. Longitudinal median line: 1. not outstanding. 2. highlighted by a concentration of spots
- 5. Mottled background: 1. absent. 2. homogeneous. 3. heterogeneous

- 6. Mottled type with: 1. dots. 2. spots and dots. 3. maculae or patches.
- 7. Dorsal surface: 1. smooth. 2. with papillae. 3. with spines
- 8. Nuchal tentacles: 1. absent. 2. present
- 9. Tentacular knobs: 1. absent. 2. present
- 10. Marginal eyes: 1. absent. 2. present on the anterior margin. 3. present around the entire body margin
- 11. Tentacular eyes: 1. absent. 2. at the base of or within the tentacles. 3. rounded and compact clusters present instead of tentacles
- 12. Frontal dorsal eyes (located between the cerebral ganglia and the anterior body margin): 1. absent. 2. in groups of two or four. 3. dispersed. 4. in a fan-like pattern
- 13. Frontal ventral eyes: 1. absent. 2. in two dense clusters. 3. in four dense clusters
- 14. Cerebral eyes (located above the cerebral ganglia): 1. absent. 2. in one cluster. 3. in two clusters. 4. horseshoe-shaped (both clusters join frontally). 5. a pair of eyes
- 15. Pharynx: 1. at the end of the first body third. 2. in the second or last body third
- 16. Pharynx shape: 1. strongly ruffled. 2. slightly ruffled
- 17. Orientation of the pharynx: 1. frontally oriented. 2. ventrally oriented. 3. caudally oriented
- 18. Oral pore position: 1. in the first body third and after the cerebral ganglion. 2. in the first body third and below the cerebral ganglia. 3. in the first body third and before the cerebral ganglion. 4. in the second body third and in the middle of the pharynx. 5. in the second body third and in the posterior region of the pharynx
- 19. Intestine: 1. with numerous branches, without anastomosis. 2. with numerous branches and anastomosis; 3. without ramifications
- 20. Intestinal vesicles: 1. absent. 2. present
- 21. Distance between male and female gonopore: 1. very close to each other (in the same body third). 2. clearly separated. 3. located in a common pore
- 22. Male gonopore: 1. single. 2. double. 3. multiple
- 23. Male copulatory apparatus: 1. not enclosed in a muscular bulb. 2. enclosed in a muscular bulb
- 24. Orientation of male copulatory apparatus: 1. backwards oriented. 2. forward oriented. 3. vertically (dorsal-ventrally) oriented. 4. oriented towards the lateral body sides
- 25. Location of male copulatory apparatus: 1. in the last body third. 2. in the second body third. 3. in longitudinal rows on either side of the pharynx. 4. in the first body third (either anterior or posterior to the pharynx)
- 26. Ejaculatory duct: 1. with a straight stylet. 2. unarmed, without a stylet (with a penis rod in *Izmira*). 3. with a spiral-shaped stylet (forming a loop between the prostatic vesicle and the pharynx)
- 27. Prostatic vesicle: 1. absent. 2. interpolated. 3. free
- 28. Connection between ejaculatory duct and prostatic vesicle: 1. with projection. 2. without projection
- 29. Internal glandular lining of prostatic vesicle: 1. smooth. 2. folded. 3. fingered. 4. ridged. 5. with tubular chambers (of citrus-like appearance in transverse section)
- 30. Prostatic secretion: 1. intravesicular-epithelial (produced by the lining epithelium). 2. intravesicular-muscular (produced by gland cells in the muscular wall of the vesicle). 3. extravesicular (produced by glands located outside the vesicle and penetrating into the interior of the vesicle by long necks)
- 31. Prostatic glands: 1. located at the proximal end of the prostatic vesicle. 2. covering the whole prostatic vesicle. 3. absent
- 32. Prostatoid organs: 1. absent. 2. present
- 33. Seminal vesicle shape: 1. simple or spindle-like. 2. tripartite or anchor-shaped
- 34. Spermiducal bulbs (heavy muscularized ends of the sperm ducts, next to the copulatory apparatus. Accessory seminal vesicles after Lang 1884, Bock 1913): 1. absent. 2. present
- 35. External (or false) seminal vesicle: 1. absent. 2. present

- 36. Vagina: 1. an elongated, tube-like organ, looping towards the male complex. 2. a short tube-like organ directed posteriorly (a short posteriorly directed vagina is a synapomorphy of Cotylea.). 3. an elongated tube-like organ slanted towards the posterior end of the body
- 37. Lang's vesicle: 1. bulbous. 2. enlarged. 3. extending to both sides. 4. absent
- 38. Uterus: 1. absent. 2. paired. 3. common uterine duct or medial sac
- 39. Uterine vesicles: 1. absent. 2. one pair of uterine vesicles. 3. multiple uterine vesicles
- 40. Cement chamber: 1. tube-like. 2. sac-like
- 41. Genital sucker. 1. absent. 2. present
- 42. Eggs: 1. ectolecithal. 2. endolecithal

#### Phylogenetic analysis

To better understand the evolutionary relationships and observed similarities between the different genera and species of the family Pleioplanidae, a phylogenetic analysis was performed using the following methodology and parameters: optimality criterion = parsimony; bootstrap with heuristic search; number of bootstrap replicates = 1000; number of characters resampled in each replicate = 45. Character-status summary of 45 total characters: all are of type "unordered", and all have equal weight. Of these, 19 characters are constant; 6 variable characters are parsimony-uninformative; 20 characters are parsimony-informative; and gaps are treated as "missing". Starting tree(s) obtained via stepwise addition. Addition sequence: simple (reference taxon = *Pleioplana atomata*; number of trees held at each step = 1. Branch-swapping algorithm: tree-bisection-reconnection (TBR) with reconnection limit = 8; initial 'Maxtrees' setting = 100; branches collapsed (creating polytomies) if maximum branch length is zero; no topological constraints in effect. *Leptoplana tremellaris* was designated as the outgroup (Fig. 7).

Also a neighbour joining (NJ) analysis was performed using character-state optimization with accelerated transformation (ACCTRAN).

#### Results

## Key to the valid pleioplanid species

1.	Ejaculatory duct with a straight stylet and directed backwards
_	Ejaculatory duct unarmed, without stylet and directed backwards
_	Ejaculatory duct with a spiral-shaped stylet (forming a loop between the prostatic vesicle and the
	pharynx) and directed forward
2.	Dorsal appearance translucent
	Dorsal appearance opaque5
3.	Spermiducal bulbs absent; body shape oval
_	Spermiducal bulbs present; body shape elongated
4.	Nuchal tentacles absent; tentacular knobs present; tentacular eyes in rounded and compact clusters in place of the tentacles; male copulatory apparatus not enclosed in a muscular bulb; Lang's vesicle present
_	Nuchal tentacles present; tentacular knobs absent; tentacular eyes at the base of or within the tentacles; male copulatory apparatus enclosed in a muscular bulb; Lang's vesicle absent
5.	Body shape elongated6
_	Body shape oval

6. Pharynx located at the end of the first body third; spermiducal bulbs present; oral pore in the second body third and in the posterior region of the pharynx; male and female gonopores clearly separated - Pharynx in the second or last body third; spermiducal bulbs absent; oral pore in the second body third and in the middle of the pharynx; male and female gonopores located in a common pore 7. Spermiducal bulbs absent; connection between ejaculatory duct and prostatic vesicle without Spermiducal bulbs present; connection between ejaculatory duct and prostatic vesicle with 8. Cerebral eyes absent; spermiducal bulbs present; vagina slanted towards the posterior end of the body......9 Cerebral eyes in two clusters; spermiducal bulbs absent; vagina looping towards the male complex 9. Dorsal longitudinal median line not outstanding; oral pore in the second body third and in the Dorsal longitudinal median line highlighted by a concentration of spots; oral pore in the second 10. Male and female gonopores located very close to each other (in the same body third); connection between ejaculatory duct and prostatic vesicle with projection ...... Male and female gonopores clearly separated (in different body thirds); connection between ejaculatory duct and prostatic vesicle without projection ..... 

#### **Taxonomy**

Polycladida Lang, 1883 Acotylea Lang, 1884 Leptoplanoidea Faubel, 1983

Pleioplanidae Faubel, 1983

## Type genus

Pleioplana Faubel, 1983

#### **Diagnosis**

Pleioplanidae (after Faubel 1983, emended): Leptoplanoidea with varying body outline. Ruffled pharynx arranged centrally or anteriorly. Male copulatory apparatus directed backwards or forwards, posterior to the pharynx. Prostatic vesicle with numerous chambers giving a citrus-like (*atomata*-type) appearance in cross section. Lang vesicle present or absent.

#### Laqueusplana gen. nov.

<u>urn:lsid:zoobank.org:act:DFBD1007-F9CA-46D8-8A8A-9827B9045040</u> Figs 1–4, 6A–B

## Type species

Laqueusplana bocki gen. et sp. nov.

#### **Diagnosis**

Pleioplanidae with tentacular and cerebral eye clusters; without tentacles. Male copulatory apparatus directed forward. Seminal vesicle and/or spermiducal bulbs present; penis duct forms a loop between prostatic vesicle and pharynx. Penis stylet long and pointed. Female apparatus usually with vagina bulbosa; Lang's vesicle present.

#### **Etymology**

The name *Laqueusplana* refers to the term "*laqueus*", Latin for loop. This is in reference to the loop that the stylet makes between the prostatic vesicle and the pharynx in this genus.

## Other species of the genus

*Pleioplana megala* (Marcus, 1952) also has a forward directed male copulatory system and shows a loop between the prostatic vesicle and the pharynx. Therefore, it must be transferred to the new genus *Laqueusplana* gen. nov.

## *Laqueusplana bocki* gen. et sp. nov. urn:lsid:zoobank.org:act:0467FB7F-9254-49E8-AE5F-D02BC2CE89B6

Figs 1–3, 6A

Notoplana atomata O.F. Müller, 1776 in Bock 1913 (partim): 202, table X, fig. 3 (individuals with a spiral-shaped stylet).

#### **Etymology**

The name of the species is dedicated to Sixten Bock, famous Swedish invertebrate zoologist (1884–1946).

#### Material examined

#### Holotype

SPAIN: sagittally sectioned, stained with Azan, Jan. 2012 (MNCN 4.01/1118 a 1150, 34 slides).

#### Paratype

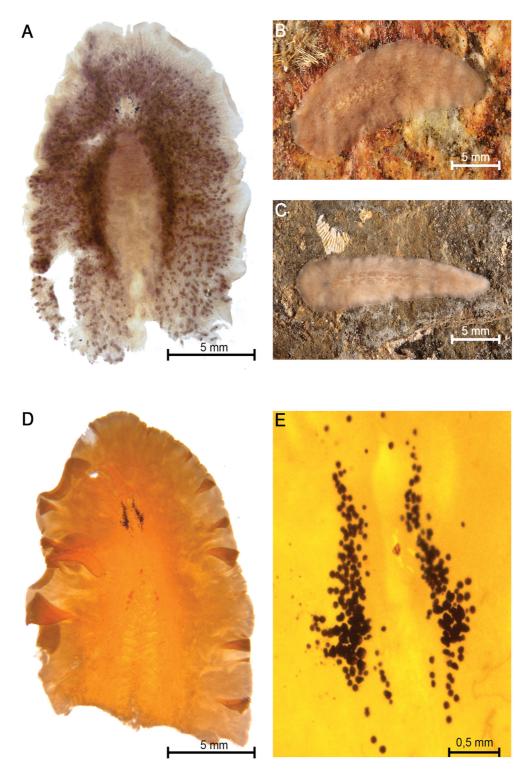
SPAIN: 1 sagittally sectioned specimen, stained with Azan, among mussels at a depth of nearly 6 meters, Dec. 2013, 42°33.7760′ N, 08°59.3090′ W (MNCN 4.01/1151 a 1189, 40 slides).

#### **Type locality**

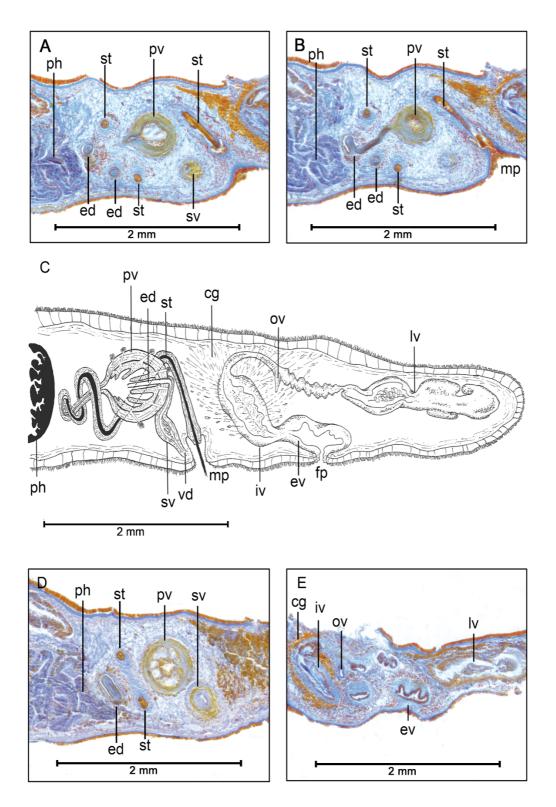
SPAIN: Galicia, Club Náutico de Riveira, among mussels on a pontoon between half to one meter depth, 42°33.7670′ N, 08°59.2860′ W.

## **Description**

Living worms 25 mm long and 7 mm wide. Body shape elongated, of fleshy consistency, broader anteriorly, then stretching to form a slightly pointed posterior end; few smooth marginal undulations (Fig. 1A–C). Dorsal surface with pale beige ground colour and evenly distributed dark brown spots,

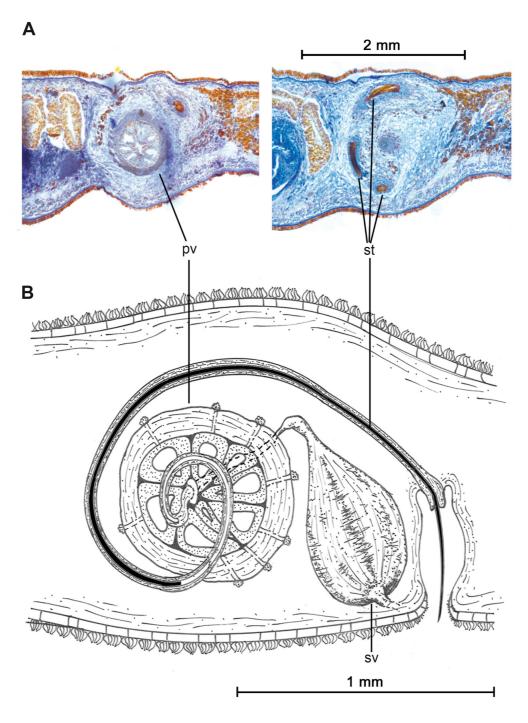


**Fig. 1.** Species of *Laqueusplana* gen. nov. **A.** Dorsal view of a fixed specimen of *Laqueusplana bocki* gen. et sp. nov. **B–C**. Dorsal views of living specimens of *L. bocki* gen. et sp. nov. **D**. Dorsal view of a fixed specimen of *L. megala* (Marcus, 1952) comb. nov. **E**. Higher magnification view showing the eye arrangement of *L. megala* comb. nov.



**Fig. 2.** Laqueusplana bocki gen. et sp. nov. **A–B**. Histological sections of the reproductive system (anterior end to the left). **C**. Sagittal reconstruction of the reproductive system. **D–E**. Histological sections of the male organ.

except in pharyngeal and genital regions; body margin transparent and spot-free (Fig. 1A). Tentacles absent; instead tentacular eyes present in two rounded clusters. Cerebral eyes form two elongated groups (between 15 and 17 eyes) next to tentacular eyes; marginal or frontal eyes lacking (Fig. 1A). Ventral surface pale brown, almost transparent. Bilobated brain with two symmetrical lobes located ventral to eyes. Pharynx occupies middle third of body, with oral pore opening ventrally in its medial region.



**Fig. 3.** *Laqueusplana bocki* gen. et sp. nov. **A.** Transverse histological sections of the male reproductive organ. **B.** Transverse reconstruction of the male reproductive system.

Male and female genital pores well separated, opening at beginning of last third of body. Ovaries well developed, lying dorsally between intestinal branches. Testes ventral.

MALE REPRODUCTIVE SYSTEM. Male copulatory organ consists of an interpolated prostatic vesicle, a true seminal vesicle and a penis papilla with a long, slender stylet, and is located almost immediately behind pharyngeal cavity (Fig. 2C). Testes located ventrally, sometimes observed between intestinal branches. Vasa deferentia run ventrally along both sides of posterior body region, swelling in spermiducal vesicles before entering seminal vesicle proximally. Seminal and prostatic vesicles forward directed (Fig. 2A). Seminal vesicle with thick muscular walls, connected to prostatic vesicle by short and narrow ejaculatory duct (Fig. 2D). Ejaculatory duct projects proximally into prostatic vesicle, crosses it and opens near distal end. Prostatic vesicle oval-shaped, enlarged and covered with strong muscular layers. Prostatic vesicle internally subdivided into at least eight parallel tubular chambers extending from proximal end, giving characteristic citrus-like appearance (Figs 2D, 3A). Extravesicular glands can be found lining proximal end of prostatic vesicle (Fig. 2A). Long coiled stylet emerges from distal end of prostatic vesicle, forms characteristic spiral-like loop between prostatic vesicle and pharynx, turns backwards and opens dorso-ventrally into male atrium (Figs 2A–D, 3). Small conical-shaped penis papilla housed within ciliated atrium (Fig. 2B).

Female Reproductive system. Female system consists of external and internal vaginas, oviducts, uteri and Lang's vesicle (Fig. 2C). Muscular external vagina, or vagina bulbosa, lined with cuboidal epithelium with fringe-like extensions. Vagina bulbosa narrows distally, then extends dorsally or anteriorly towards male copulatory system before reaching internal vagina. Internal vagina turns posteriorly and runs dorsally to posterior end. Oviducts lead separately into common receptacle at proximal end of internal vagina. Numerous shell and cement glands open in region surrounding both vaginas. After receiving oviducts, vagina continues into enlarged Lang's vesicle that extends posteriorly, sometimes reaching posterior end of animal (Fig. 2E).

#### Differentional diagnosis

Due to the presence of a prostatic vesicle filled with tubular chambers (*atomata*-type), separated gonopores and a vagina bulbosa, the genus *Laqueusplana* gen. nov. belongs to the family Pleioplanidae. However, the presence of a long stylet with a spiral loop and a of forward directed male copulatory system justifies the erection of a new genus within this family.

Species of the genera *Izmira* Bulnes, 2010, *Melloplana* Faubel, 1983 and *Persica* Maghsoudlou *et al.*, 2015 share some characteristics of the Pleioplanidae family, mainly the presence of an *atomata*-type prostatic vesicle. However, these genera can be differentiated from the genus *Laqueusplana* gen. nov. et by the following: *Izmira* lacks cerebral eyes and Lang's vesicle and has a backwards directed male copulatory system and a penis rod; *Melloplana* is characterized by the presence of an unarmed, muscular penis papilla and *Persica* presents distinct characteristics such as the presence of nuchal tentacles, a male copulatory organ wrapped in a muscular bulb and an elongated stylet.

Laqueusplana gen. nov. and Pleioplana Faubel, 1983 are morphologically most closely related to each other, which is reflected in a number of shared characteristics. Externally, both have an elongated body with pale ground colour, two clusters of both tentacular and cerebral eyes and lack tentacles. They also possess a ruffled pharynx that occupies the middle third of the body and a copulatory system that begins posterior to the pharynx. Laqueusplana gen. nov., unlike Pleioplana, has the seminal and prostatic vesicles directed forward and a longer, coiled stylet situated between the pharynx and prostatic vesicle.

The female copulatory system of the genus *Laqueusplana* gen. nov. consists of a well-developed vagina bulbosa and an enlarged Lang's vesicle, which sometimes reaches the posterior end of the animal. In

general, the female system in this genus is larger than those of other Pleioplanidae genera. *Pleioplana* and *Melloplana* show a similar female system as *Laqueusplana* gen. nov., although smaller in size, while *Izmira* and *Persica* present a shorter female track and a complete absence of Lang's vesicle.

#### Biology

Live specimens have rapid scrolling movements and, when stressed, are able to swim by rotating the body, orienting the ventral body surface towards the water surface, and shaking the body while waving its edges.

#### Remarks

During the study and analysis of *Laqueusplana bocki* gen. et sp. nov. and similar species we discovered that in the description of *Notoplana atomata* of Bock (1913: 202), this author mentioned that some individuals in his material showed a long stylet forming a loop. However, in his reconstruction of *Notoplana atomata* (Bock 1913: 201, fig. 39) this author did not draw a stylet with a loop or a prostatic vesicle directed forward, but he photographed a specimen (Bock 1913: table XX, fig. 3) with these characteristics. This fact means that Bock took note of the outstanding features of the new genus *Laqueusplana* gen. nov., but probably due to the scarcity of material did not give it any importance and considered the differences as exceptions or artefacts of fixation that did not allow the establishment of new species. Nevertheless, it is clear that of *Laqueusplana* gen. nov. was present in the study of Bock but it wasn't described; therefore, we dedicate the new species to this great specialist of Polycladida.

#### **Distribution**

Laqueusplana bocki gen. et sp. nov. has only been found along the north-western Atlantic coast of Galicia, Spain. Notably, within the family Pleioplanidae (4 genera and 13 species), *Pleioplana atomata* (O.F. Müller, 1776) and *Laqueusplana bocki* gen. et sp. nov. are the only representatives of the family found along European Atlantic coasts. Nonetheless, further research must be done to determine the entire distribution range of the new species.

*Laqueusplana megala* (Marcus, 1952) comb. nov. Figs 1, 4, 6B

Notoplana megala Marcus, 1952: 85, figs 154-158.

Pleioplana megala – Faubel 1983: 117.

## **Examined material**

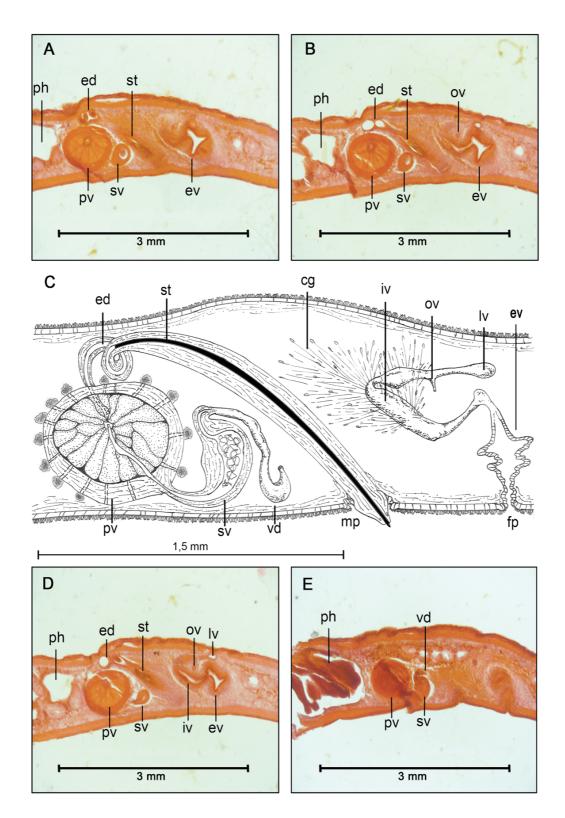
One mature, sagittally sectioned specimen from the invertebrate collection of the Swedish Museum of Natural History in Stockholm (Sweden), Cat Nr: SMNH 109094 to 109096 (3 slides).

## Type locality

BRAZIL: Island of São Sebastião.

## Description

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Without mottled background. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present (Fig. 1D). Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters (Fig. 1E). Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral



**Fig. 4.** *Laqueusplana megala* (Marcus, 1952) comb. nov. **A–B**. Histological sections of the female reproductive system (anterior end to the left). **C**. Sagittal reconstruction of the reproductive system. **D–E**. Histological sections of the male reproductive system (anterior end to the left).

pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM (Fig. 4A–E). Single male pore. Male copulatory apparatus not enclosed within muscular bulb, oriented forward and located in last body third. Ejaculatory duct with spiral-shaped stylet (forming a loop between prostatic vesicle and pharynx). Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system (Fig. 4A–D). Vagina elongated, tube-like, looping towards male complex. Bulbous Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### **Biology**

Specimens found on algae from the upper littoral zone.

#### Distribution

Ilha de São Sebastião, Ubatuba (São Paulo State), Brazil.

**Pleioplana** Faubel, 1983 Fig. 5A–F

Pleioplana Faubel 1983: 117, fig. 37A.

#### Type species

Pleioplana atomata (O.F. Müller, 1776) (individuals with straight stylets).

#### **Diagnosis**

Pleioplanidae with tentacular and cerebral eye clusters; without tentacles. Male copulatory apparatus directed backwards. Seminal vesicle and/or spermiducal bulbs present. Penis duct continuous with a short (thick or thin) penis stylet. Female apparatus usually with vagina bulbosa; Lang's vesicle present.

#### Valid species of the genus

Pleioplana atomata (O.F. Müller, 1776).

Pleioplana mortenseni (Bock, 1913).

Pleioplana delicata (Yeri & Kaburaki, 1918).

Pleioplana californica (Plehn, 1898).

Pleioplana bosphorensis Bulnes, Kalkan & Karhan, 2009.

Pleioplana okusi Bulnes, Kalkan & Karhan, 2009.

**Pleioplana atomata** (O.F. Müller, 1776) Fig. 5A

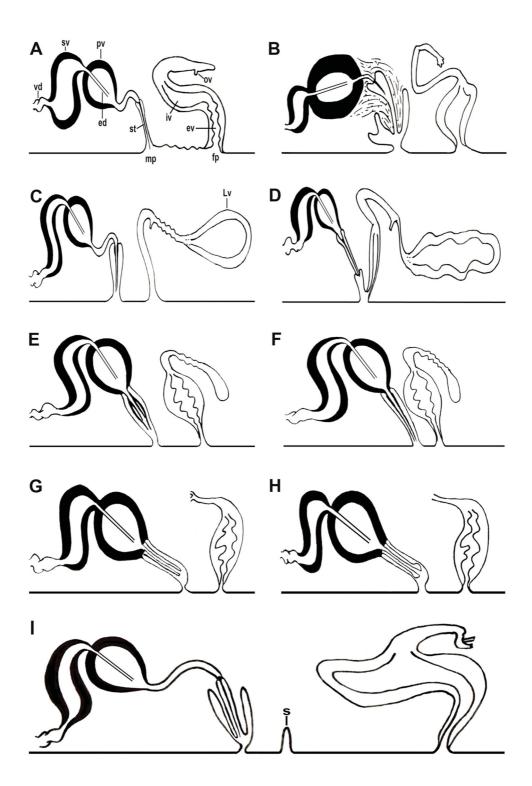
*Planaria atomata* O.F. Müller, 1776: 37, pl. XXXII, figs 3–4. *Planaria punctata* O.F. Müller, 1776: 223.

Leptoplana fallax Diesing, 1840: 198–199.

Leptoplana droebachensis Oersted, 1845: 415.

Polycelis variabilis Girard, 1851: 251.

*Planaria maculata* Dalyell, 1853: 104–106, pl. IV, figs 27–32.



**Fig. 5.** Schematic representations comparing the copulatory apparatus. **A.** *Pleioplana automata* (O.F. Müller, 1776). **B.** *P. mortenseni* (Bock, 1913). **C.** *P. delicata* (Yeri & Kaburaki, 1918). **D.** *P. californica* (Plehn, 1898). **E.** *P. bosphorensis* Bulnes, Kalkan, & Karhan, 2009. **F.** *P. okusi* Bulnes, Kalkan, & Karhan, 2009. **G.** *Izmira cinari* Bulnes, 2010. **H.** *I. turkeyi* Bulnes, 2010. **I.** *Persica qeshmensis* Maghsoudlou *et al.* 2015.

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Leptoplana ellipsoides Girard, 1854: 27, table 2, fig. 16.
Leptoplana virilis Verrill, 1893: 478, table 43, figs 1, 1a.
Leptoplana angusta Verrill, 1893: 485, table 40, fig. 8, table 44, figs 2, 2a, 3.
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*Leptoplana atomata* – Oersted 1843: 569; 1844: 49, 79, pl. II, fig. 24. — Leuckart 1847: 149; 1859: 183. — Diesing 1850: 197; 1862: 532. — Maitland 1851: 187–188. — Stimpson 1857: 21. — Johnston 1869: 7. — MacIntosh 1874: 150. — Möbius 1875: 154.

*Notoplana atomata* – Bock 1913: 195–202, pl. IV, fig. 8, pl. X, figs 3, 6, 8–9.

*Planaria atomata* – Gmelin 1789: 3091. — Fleming 1823: 297. — Delle Chiaje 1841: 179. — Blainville 1826: 217. — Forbes & Goodsir 1839: 353. — Johnston 1845: 436.

Pleioplana atomata – Faubel 1983: 117.

*Leptoplana droebachensis* – Diesing 1862: 526. — Jensen 1878: 76, pl. VII, figs 10–14. — Lang 1884: 494–495.

Notoplana fallax – Bock 1913: 204–205.

Polycelis fallax – Quatrefages 1845: 135, pl. 3, fig. 10, pl. 7, fig. 1, pl. 8, figs 1, 7.

*Leptoplana fallax* – Stimpson 1857: 31. — Diesing 1862: 533. — Lang 1884: 492. — Vaillant 1890: 654. — Hallez 1893: 166. — Laidlaw 1903: 308.

Leptoplana variabilis – Diesing 1862: 542.

*Leptoplana ellipsoides* – Stimpson 1857: 27. — Diesing 1862: 533. — Verrill 1874: 43, 45, 132; 1893: 483, pl. 40, figs 5–6, pl. 42, figs 4, 4 a–b; 1895: 533. — Lang 1884: 512. — Girard 1893: 200, pl. 6, fig. 61. — Whiteaves 1901: 63.

Notoplana virilis - Bock 1913: 208.

Leptoplana angusta – Pearse 1938: 76–77, fig. 26.

#### Material examined

One mature, sagittally sectioned specimen deposited in the invertebrate collection of the Museo Nacional de Ciencias Naturales de Madrid (Spain). Cat Nr: MNCN 4.01/763 to MNCN 4.01/834 (72 slides).

#### **Type locality**

**NORWAY** 

#### **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Heterogeneous mottled background with maculae or patches. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located at end of first body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in the posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Enlarged Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### **Biology**

Commonly found along coastal shores, mainly under stones.

#### **Distribution**

Along the northern Atlantic.

**Pleioplana mortenseni** (Bock, 1913) Fig. 5B

Notoplana mortenseni Bock, 1913: 192-195, pl. V, fig. 7, pl. VI, fig. 10.

Pleioplana mortenseni - Faubel 1983: 117.

## Type locality

THAILAND: Gulf of Thailand.

#### **Description**

Body shape oval. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with spots and dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle without projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs absent.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Enlarged Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

## Distribution

Gulf of Thailand.

*Pleioplana delicata* (Yeri & Kaburaki, 1918) Fig. 5C

Notoplana delicata Yeri & Kaburaki, 1918: 13–15, figs 11–13, pl. II, fig. 14.

Pleioplana delicata – Faubel 1983: 117.

## Type locality

JAPAN: Misaki.

#### **Description**

Body shape elongated. Cryptic colour. Translucent dorsal appearance. Longitudinal median line highlighted by concentration of spots. Heterogeneous mottled background with spots and dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Enlarged Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

## **Biology**

Found along the coast during the low tide.

#### **Distribution**

Shirahama (Prov. Awa), Japan.

**Pleioplana californica** (Plehn, 1897) Fig. 5D

*Leptoplana californica* Plehn, 1897: 93, table 5, figs 1–2. *Stylochoplana plehni* Bock, 1913: 80.

Leptoplana californica – Plehn 1899: 451, fig. B. *Pleioplana californica* – Faubel 1983: 117. *Stylochoplana plehni* – Hyman 1953: 305.

## Type locality

USA: Monterey Bay, Monterey, California.

#### **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with spots and dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Male and female gonopores located in a common pore.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal

glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at the proximal end of the prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like, looping towards the male complex. Enlarged Lang's vesicle. Paired uterus. One pair of uterine vesicles. Tube-like cement chamber. Endolecithal eggs.

## **Biology**

Found during low tide between rocks and sand.

#### **Distribution**

California, USA.

#### Note

*Pleioplana inquieta* Heath & McGregor, 1912, considered synonymous with *P. californica* by Hyman (1953). According to Hyman, *P. inquieta* not only shows the same morphological features as *P. californica*, but also shares the same habitat and distribution, namely Monterey Bay, California. Therefore, in this review, *P. inquieta* is considered a synonym of *P. californica*.

**Pleioplana bosphorensis** Bulnes, Kalkan & Karhan, 2009 Fig. 5E

Pleioplana bosphorensis Bulnes, Kalkan & Karhan, 2009: 5–6, figs 1–2.

## Type locality

TURKEY: Altinkum, Bosphorus Strait (Bosphorus).

## **Description**

Body shape oval. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with spots and dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within a muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present. External (or false) seminal vesicle absent.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Bulbous Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### **Biology**

Found on a hard bottomed mussel bed of *Mytilus galloprovincialis*.

#### **Distribution**

Bosporus Strait, Turkey.

**Pleioplana okusi** Bulnes, Kalkan & Karhan, 2009 Fig. 5F

Pleioplana okusi Bulnes, Kalkan & Karhan, 2009: 8–9, fig. 3.

#### Type locality

TURKEY: Altinkum, Bosphorus Strait (Bosporus).

#### **Description**

Body shape oval. Cryptic colour. Translucent dorsal appearance. Longitudinal median line not outstanding. Heterogeneous mottled background with spots and dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in last body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs absent.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Bulbous Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

## **Biology**

Found on a hard bottomed mussel bed of *Mytilus galloprovincialis*.

#### **Distribution**

Bosporus Strait, Turkey.

*Izmira* Bulnes, 2010 Fig. 5G–H

Izmira Bulnes, 2010: 13-14.

#### Type species

Izmira cinari Bulnes, 2010.

#### **Diagnosis** (after Bulnes 2010)

Pleioplanidae, oval to elongated, without tentacles. Tentacular eyes present; cerebral, frontal and marginal eyes absent. Male apparatus oriented backwards with a true seminal vesicle, interpolated true citrus-type prostatic vesicle, without stylet and with a penis rod. Female apparatus with vagina bulbosa; Lang's vesicle absent.

## Valid species:

*Izmira cinari* Bulnes, 2010. *Izmira turkeyi* Bulnes, 2010.

*Izmira cinari* Bulnes, 2010 Fig. 5G

Izmira cinari Bulnes, 2010: 14, 16, figs 5-6.

#### **Type locality**

TURKEY: Izmir Bay, Aegean Sea.

## **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Longitudinal median line highlighted by concentration of spots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes absent. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores located very close to each other (in same body third).

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in second body third. Ejaculatory duct unarmed, without stylet, but with penis rod. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like and oriented backwards. Lang's vesicle absent. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

## **Biology**

Found in sublittoral mussel beds of *Mytilus galloprovincialis*.

#### Distribution

Bosporus, Aegean Sea, Turkey.

*Izmira turkeyi* Bulnes, 2010 Fig. 5H

Izmira turkeyi Bulnes, 2010: 17, 20, figs 7-8.

## Type locality

TURKEY: Alsancak Harbour, Izmir Bay, Aegean Sea.

#### **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with dots. Smooth dorsal surface. Nuchal tentacles

absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes absent. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores located very close to each other (in same body third).

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in second body third. Ejaculatory duct unarmed, without stylet, but with penis rod. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like and oriented backwards. Lang's vesicle absent. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### Biology

Found in sublittoral mussel beds of *Mytilus galloprovincialis*.

#### **Distribution**

Aegean Sea, Turkey.

**Persica** Maghsoudlou, Bulnes & Rahimian, 2015 Fig. 5I

Persica Maghsoudlou, Bulnes & Rahimian, 2015: 5.

#### Type and only species

Persica qeshmensis Maghsoudlou, Bulnes & Rahimian, 2015.

#### **Diagnosis**

Pleioplanidae with tentacular and cerebral eye clusters; with small nuchal tentacles. Male copulatory apparatus directed backwards. Seminal vesicle and/or spermiducal bulbs present. Penis duct continuous with a long penis stylet. Female apparatus usually with vagina bulbosa; Lang's vesicle absent.

**Persica qeshmensis** Maghsoudlou, Bulnes & Rahimian, 2015 Fig. 5I

Persica qeshmensis Maghsoudlou, Bulnes & Rahimian, 2015: 10, 12, figs 2–6, 7a–c.

#### **Type locality**

IRAN: Persian Gulf, Qeshm Island.

#### **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with dots. Smooth dorsal surface. Nuchal tentacles present, small. Tentacular knobs absent. Marginal eyes absent. Tentacular eyes at base of or within tentacles. Dorsal and ventral frontal eyes absent. Cerebral eyes in two clusters. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third

and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

Male REPRODUCTIVE SYSTEM. Single male pore. Male apparatus enclosed within muscular bulb, oriented backwards and located in second body third. Ejaculatory duct with straight stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs present.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Lang's vesicle absent. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### Biology

Found in the intertidal zone along the Iranian coast of the Persian Gulf.

#### **Distribution**

Iran. Persian Gulf.

*Melloplana* Faubel, 1983 Fig. 6C–D

Melloplana Faubel, 1983: 117, fig. 37b.

#### Type species

Melloplana ferruginea (Schmarda, 1859).

#### **Diagnosis** (after Faubel 1983)

Pleioplanidae with tentacular and cerebral eye-spots; tentacles lacking. Male copulatory apparatus oriented backwards with seminal vesicle, without stylet and with penis papilla. Female apparatus usually with vagina bulbosa. Lang's vesicle present.

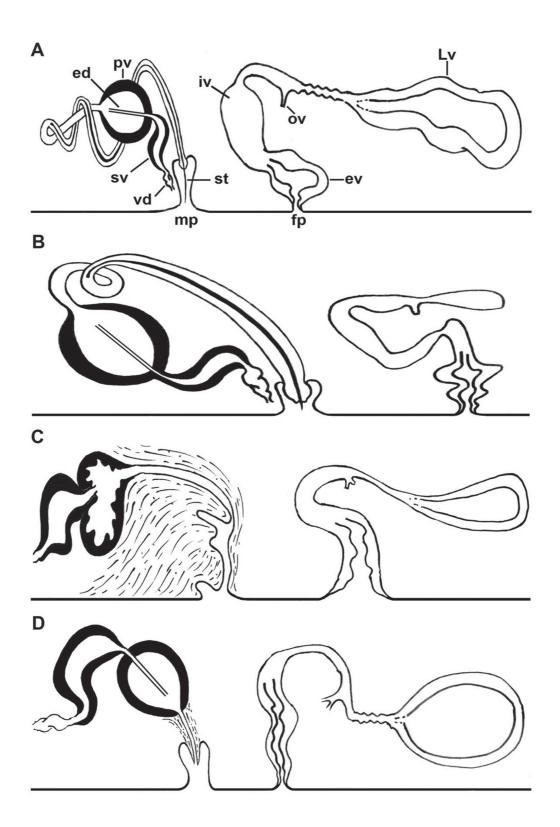
#### Valid species:

*Melloplana ferruginea* (Schmarda, 1859). *Melloplana japonica* (Kato, 1937).

## *Melloplana ferruginea* (Schmarda, 1859) Fig. 6C

Polycelis ferruginea Schmarda, 1859: 22, pl. IV, fig. 48. Discocelis binoculata Verrill, 1901: 43–44, pl. 5, figs 3–4. Notoplana bahamensis Bock, 1913: 208–210, pl. VI, figs 2–3. Notoplana caribbeana Hyman, 1939: 2–4, figs 3–8.

Notoplana binoculata – Hyman 1939: 8–10, figs 13–14. Leptoplana ferruginea – Diesing 1862: 530. Notoplana ferruginea – Stummer-Traunsfels 1933: 3486, 3521–3522, fig. 60. Melloplana ferruginea – Faubel 1983: 117.



**Fig. 6.** Schematic representations comparing the copulatory apparatus. **A.** *Laqueusplana bocki* gen. et sp. nov. **B.** *L. megala* (Marcus, 1952) comb. nov. **C.** *Melloplana ferruginea* (Schmarda, 1859). **D.** *M. japonica* (Kato, 1937).

## **Type locality**

BERMUDA: Long Bird Island.

#### **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Homogeneous mottled background with dots. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two groups. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in second body third. Ejaculatory duct unarmed, without stylet. Prostatic vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle without projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs absent.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Enlarged Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

## **Biology**

Found under stones and on algae, e.g., Halimeda.

#### Distribution

Bermuda.

*Melloplana japonica* (Kato, 1937). Fig. 6D

*Notoplana japonica* Kato, 1937: 215–216, figs 5–6, pl. XIV, figs 6–7.

Melloplana japonica – Faubel 1983: 117.

#### Type locality

JAPAN: Kanayama (Yuzaki, Yunosaki).

## **Description**

Body shape elongated. Cryptic colour. Opaque dorsal appearance. Longitudinal median line not outstanding. Without mottled background. Smooth dorsal surface. Nuchal tentacles absent. Tentacular knobs present. Marginal eyes absent. Tentacles absent; instead tentacular eyes present in rounded and compact clusters. Dorsal and ventral frontal eyes absent. Cerebral eyes in two groups. Pharynx located in second or last body third, strongly ruffled and ventrally oriented. Oral pore position in second body third and in posterior region of pharynx. Intestine with numerous branches without anastomosis. Intestinal vesicles absent. Gonopores clearly separated.

MALE REPRODUCTIVE SYSTEM. Single male pore. Male apparatus not enclosed within muscular bulb, oriented backwards and located in second body third. Ejaculatory duct unarmed, without stylet. Prostatic

vesicle interpolated. Connection between ejaculatory duct and prostatic vesicle with projection. Internal glandular lining of prostatic vesicle with tubular chambers. Extravesicular prostatic secretion. Prostatic glands located at proximal end of prostatic vesicle. Seminal vesicle simple or spindle-like. Spermiducal bulbs absent.

Female Reproductive system. Vagina elongated, tube-like, looping towards male complex. Enlarged Lang's vesicle. Paired uterus. Uterine vesicles absent. Tube-like cement chamber. Endolecithal eggs.

#### **Biology**

Specimens were collected from the undersurface of stones deeply embedded in the sand at low tide during spring and summer.

#### Distribution

Japan.

## Phylogenetic analysis

The obtained tree had a length of 55 steps and had the following values: Consistency index (CI) = 0.6000; Homoplasy index (HI) = 0.4000; CI excluding uninformative characters = 0.5417; HI excluding uninformative characters = 0.4583; Retention index (RI) = 0.5510 and Rescaled consistency index (RC) = 0.3306.

The recovered topologies from both trees (MP and NJ) support the genus *Persica* as the sister taxon of all other family members (Fig. 7). In addition, the genera *Melloplana*, *Pleioplana* and *Laqueusplana* gen. nov. group together in a well-supported clade (BP=100), with *Izmira* as the sister group of this clade. Furthermore, based on the NJ tree, *Pleioplana atomata* and the two species belonging to *Laqueusplana* gen. nov. show greater similarity to the *Melloplana* species than to the other *Pleioplana* species.

## **Discussion**

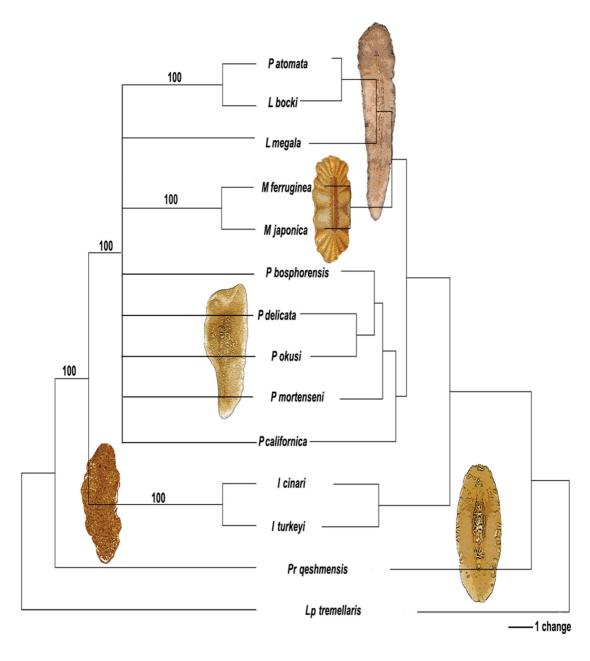
Following a systematic revision of the family Pleioplanidae, a new genus *Laqueusplana* gen. nov. is established and described. *Laqueusplana* gen. nov. is characterized by a very long stylet that forms a spiral-shaped loop and a forward oriented male copulatory organ (likely due to the manner in which the stylet loops). Phylogenetic analyses, used to elucidate the relationship between this new genus and other genera of Pleioplanidae, notably show the early divergence/differentiation of the genus *Persica*. In contrast to other genera of Pleioplanidae, *Persica* has nuchal tentacles. In addition, its reproductive system is distinct, having prostatic glands distributed over the entire surface of the prostatic vesicle, a male copulatory organ wrapped in a muscular bulb and an elongated stylet.

The phylogenetic analyses also recovered the two species of the genus *Izmira* as a monophyletic group, which form the sister group of all the other species of the family, with the exception of *Persica*. The genus *Izmira* is differentiated from the other genera by the absence of cerebral eyes, gonopores located closely together, a backwards-directed vagina and an unarmed male copulatory organ with a penis rod.

Our results support a close evolutionary relationship among *Pleioplana*, *Melloplana* and *Laqueusplana* gen. nov. The species of these genera are characterized by, in *Pleioplana*, the presence of a straight, wide or narrow, stylet, in *Melloplana* by an unarmed male organ without stylet or rod and in *Laqueusplana* gen. nov. by a long, spiral-shaped stylet. Although *Laqueusplana bocki* gen. et sp. nov. and *L. megala* comb. nov. have previously been described as belonging to the genus *Pleioplana*, and specifically in the case of *L. bocki* as *P. atomata*, both *Laqueusplana* gen. nov. species differ from *P. atomata* in the length and shape of the stylet and the orientation of the copulatory organ.

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**Fig. 7.** Maximum parsimony and NJ trees obtained from analyses of Pleioplanidae based on morphological data. The MP tree is shown on the left with bootstrap values indicated at the nodes. The NJ tree is shown on the right.

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