Zootaxa 4132 (4): 493–508
http://www.mapress.com/j/zt/

Article

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)

http://doi.org/10.11646/zootaxa.4132.4.2
http://zoobank.org/urn:lsid:zoobank.org:pub:58F09A17-A963-4702-AC53-D495FE654B82

Phylogenetic analysis of the genus *Lensia* (Cnidaria, Hydrozoa, Siphonophora), based on the species morphology

ERIC Y. NISHIYAMA[1*], GUILHERME C. RIBEIRO[2] & OTTO M. P. OLIVEIRA[1]

[1]*Universidade Federal do ABC, Centro de Ciências Naturais e Humanas. Rua São Paulo, Jardim Antares, 09606-070, São Bernardo do Campo, SP, Brazil*

[2]*Universidade Federal do ABC, Centro de Ciências Naturais e Humanas, Rua Santa Adélia, 166, Bangu, 09210-170, Santo André, SP, Brazil*

[3]*Corresponding author. E-mail: ericyukihiro@gmail.com*

Abstract

Siphonophores are poorly studied despite their abundance and ecological importance in marine ecosystems. The genus *Lensia* Totton, 1932 contains the highest number of species within Siphonophora, but systematic studies of these organisms are scarce in the literature. This study presents a phylogenetic analysis for fifteen species of *Lensia* based on morphological data. The material for this study was obtained during two oceanographic campaigns made along the southeastern Brazilian coast. A total of twenty two characters of the anterior nectophore morphology were scored. The shortest trees were searched using parsimony (under different weighting regimes). All analyses provided the same topology: (*M. kochi* (*D. dispar* + *D. bojani*) (*L. leloupi* (*L. havock* (*L. conoidea* (*L. subtilis; L. meteori*; *L. hardy*; *L. fowleri*; (*L. subtiloides* (*L. hotspur*; *L. cossack*; *L. campanella*)); (*L. multicristata* (*L. hunter* (*L. lelouveteau* + *L. grimaldii*))). The monophyly of the genus *Lensia* is supported by the hydroecium measuring up to 1/4 the height of the nectosac.

Key words: Diphyidae; nectophores; phylogeny; siphonophores

Introduction

Siphonophores are divided into three groups, based on the presence or absence of two structures (pneumatophore and nectophores). The Cystonectae are characterized by the presence of pneumatophore and absence of nectophores, the Physonectae are characterized by the presence of both structures, and the Calycophorae are characterized by the presence of nectophores and absence of pneumatophore (Totton 1965; Pugh 1999).

The genus *Lensia* Totton, 1932, belonging to the Calycophorae, is the richest in number of species within Siphonophora. Species of this group have a complex life cycle, composed of a succession of several different life stages that share few similarities in morphology or size (Grossmann *et al*. 2014).

The most well-known life stage is called polygastric stage, usually composed of two large zooids (anterior and posterior nectophores), and a siphosomal stem containing many cormidia. The cormidia, composed of gonads, gastrozooids, tentacles and bracts, are released from the posterior end of the polygastric stage in several species, to form free-living sexual units, called eudoxids. In calycophoran siphonophores, identification is commonly associated with the morphology of the anterior nectophore of the polygastric stage (Grossmann *et al*. 2014).

The genus *Lensia* belongs to the family Diphyidae, which presents a polygastric stage, generally including two serially arranged nectophores (Kirkpatrick & Pugh 1984). The nectophores are asexual medusoid structures, which contain a muscular structure known as nectosac, and an opening known as ostium. Strong contractions of the nectosac force water out of the nectophore, through the ostium, and propel the colony forwards (Mackie 1964).

The anterior nectophore presents a somatocyst, while the posterior nectophore lacks this structure. The somatocyst usually contains oil droplets which may help controlling the flotation of the animal. The nectosac occupies most of the nectophore length, allowing a fast and active swimming (Kirkpatrick & Pugh 1984). Additionally, the siphosomal stem can be totally withdrawn into an external hollowed-out structure known as hydroecium (Mapstone 2014), which is usually reduced in the anterior nectophore (Pugh 1999) (Figure 1).

Accepted by B. Bentlage: 7 Jun. 2016; published: 1 Jul. 2016

The genus *Lensia* includes calycophorans whose anterior nectophores of the polygastric stage are pentagonal, and that may contain up to 15 or more longitudinal ridges. The somatocyst is usually small. The hydroecium is shallow and closed by short and divided mouth plates. The radial canals do not possess comissures. The nectosac reaches almost the top of the nectophore (Alvariño & Wojtan 1984).

Little is known about the phylogenetic relationships within siphonophores (e.g. families and genera) (e.g. Dunn *et al*. 2005; Dunn & Wagner 2006), and very little progress has been made until now for the genus *Lensia*, the group's most diverse genus with 26 species currently considered valid (e.g. Grossmann *et al*. 2014). In this context, the present study aimed to fill a significant portion of this gap. A phylogenetic analysis of 15 species of the genus *Lensia* was performed based on a detailed morphological study of the anterior nectophore of specimens obtained from two oceanographic campaigns made throughout the southeastern Brazilian coast.

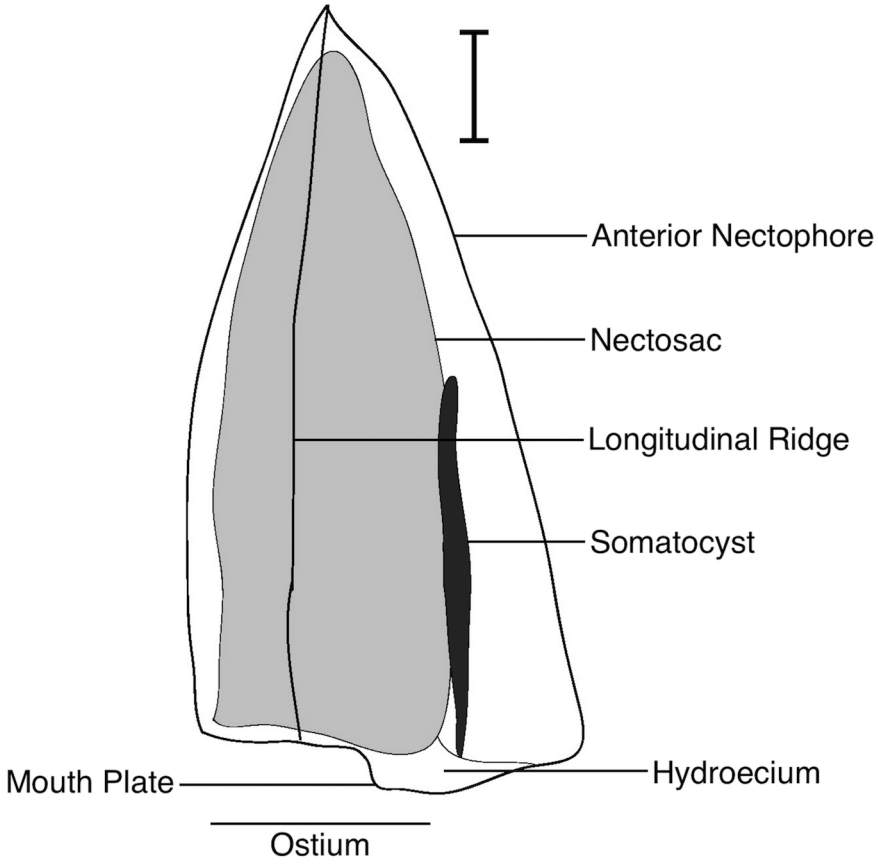
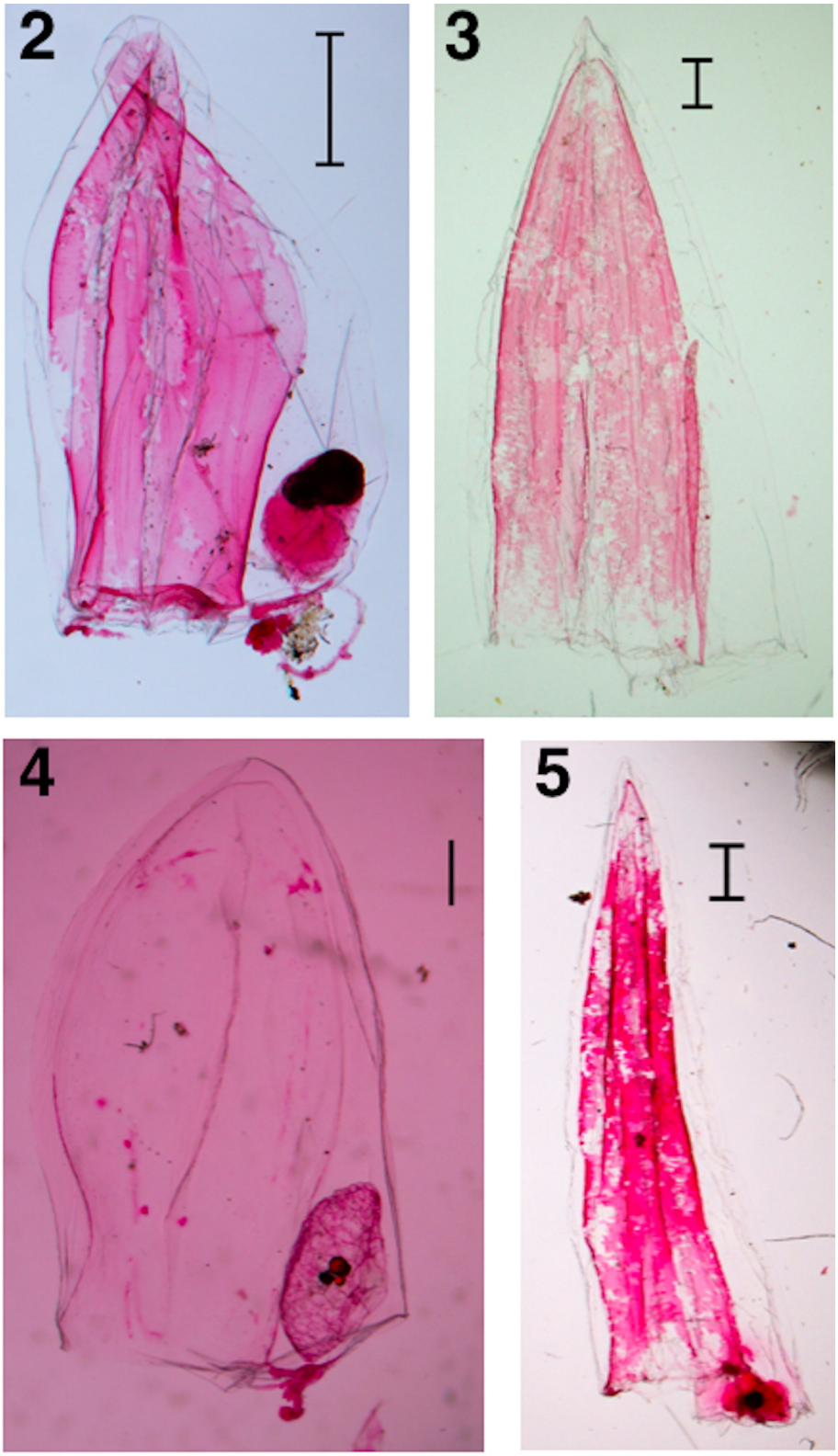


FIGURE 1. Scheme showing the structures of the anterior nectophore of *Lensia conoidea* (scale: 0.5 mm).

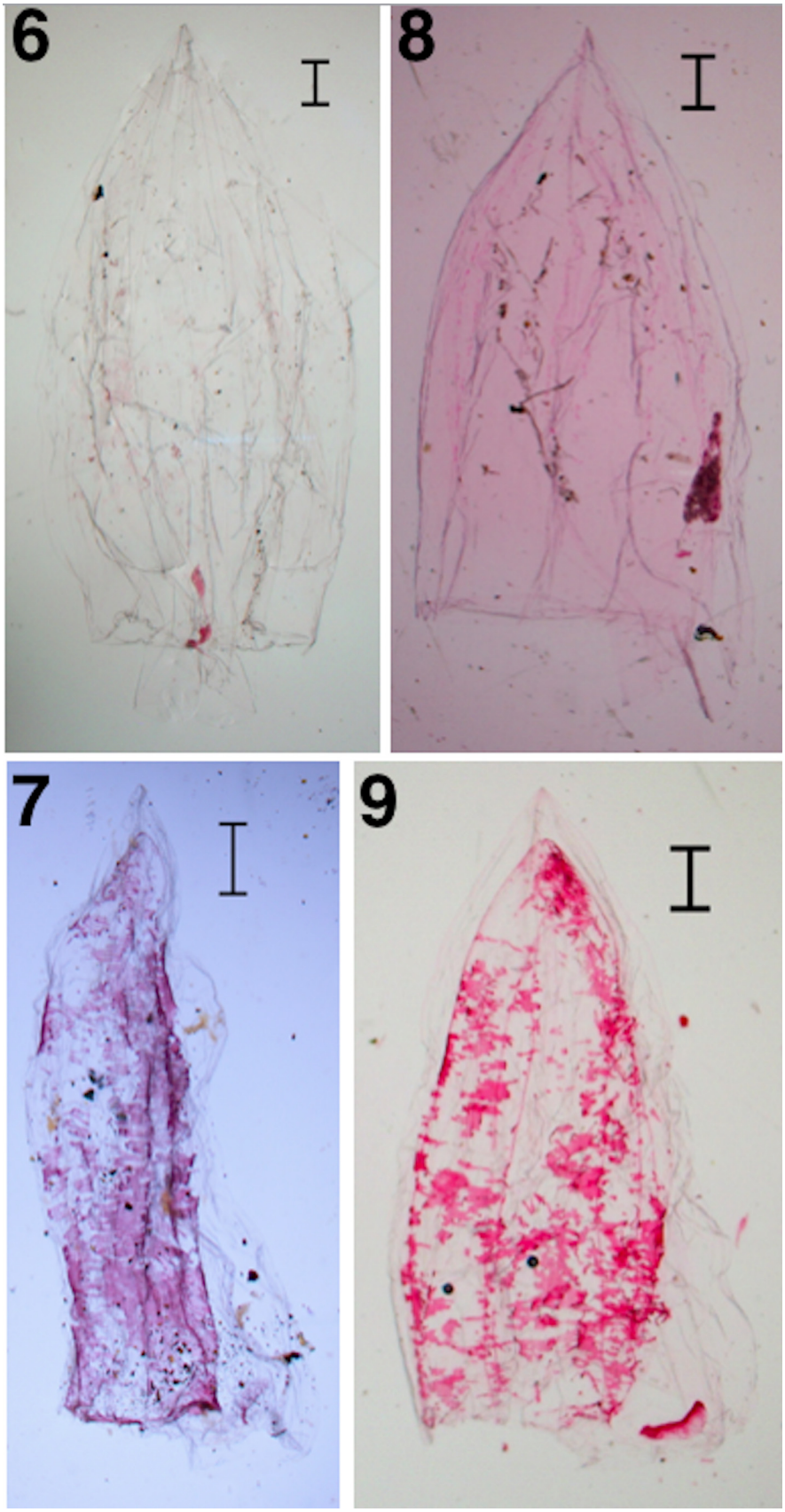
Material and methods

The material for this study was collected during two oceanographic campaigns made along the southeastern Brazilian coast (FINEP III and Habitats Project). The FINEP III campaign was conducted between Cold Cape, Rio de Janeiro (23°S) and the south region of Santa Marta Grande Cape, Santa Catarina (29°S), in May 1976. The Habitats Project campaign was conducted in the Campos Basin, Rio de Janeiro (located between 24°S and 20.5°S), from February 2009 to February 2010.

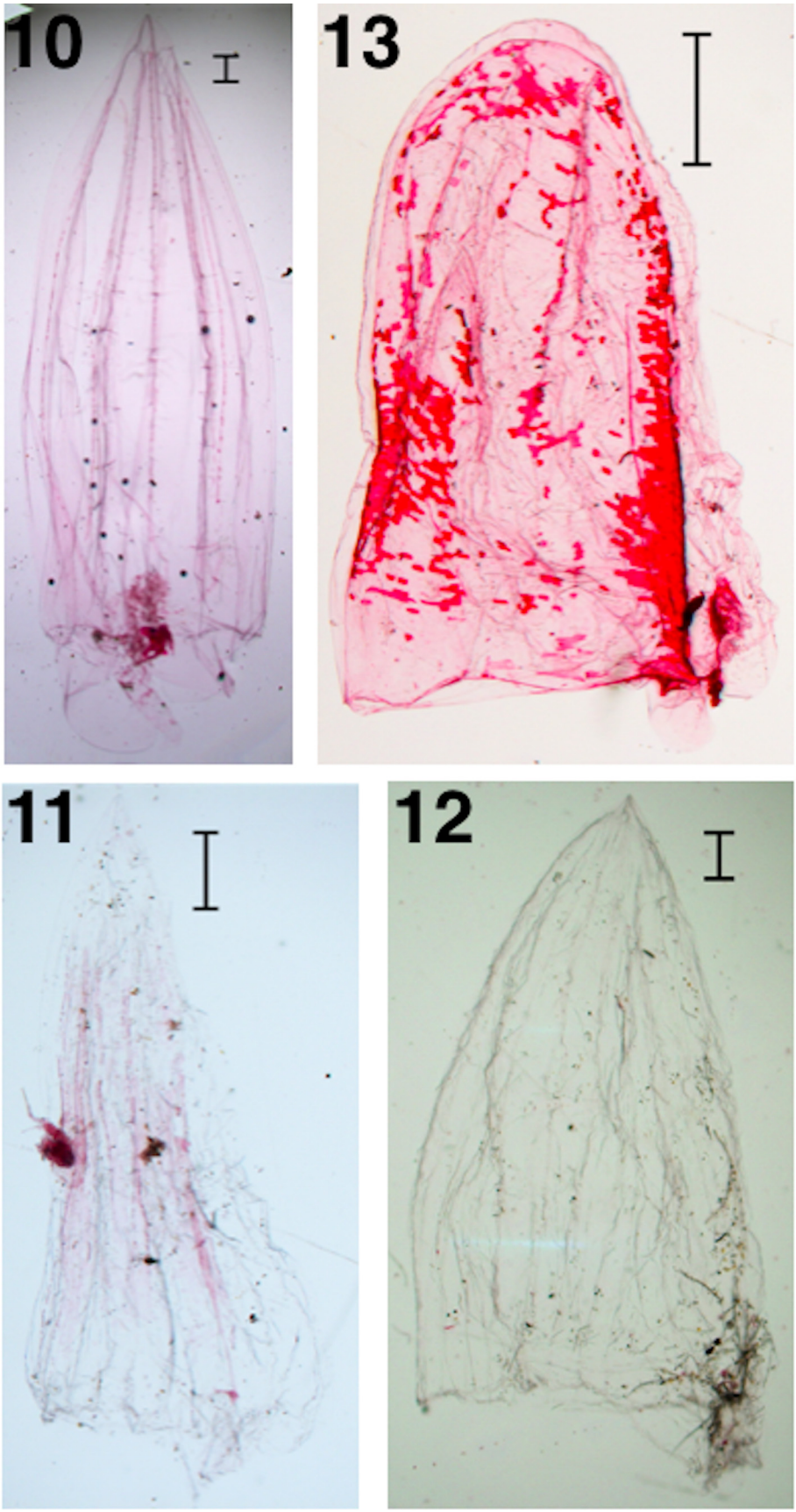
Specimens were preserved in 4% formaldehyde solution and dyed with fuchsine. The anterior nectophores of the analyzed specimens were placed on a hollowed-out microscope slide, and were photographed using a stereomicroscope with digital camera attached (Figures 2 to 19). Specimens were identified using identification keys and descriptions (e.g. Totton 1965; Kirkpatrick & Pugh 1984; Pagès & Gili 1992; Pugh 1999). Drawings of the best preserved specimens were made using the Adobe Illustrator software (Figures 20 to 37). Most specimens were damaged, making it difficult to observe some structures. Thus, we attempted to reconstruct the species morphology based on the observation of more than one single specimen, when available. Additional information about the examined material is given in Appendix 1.



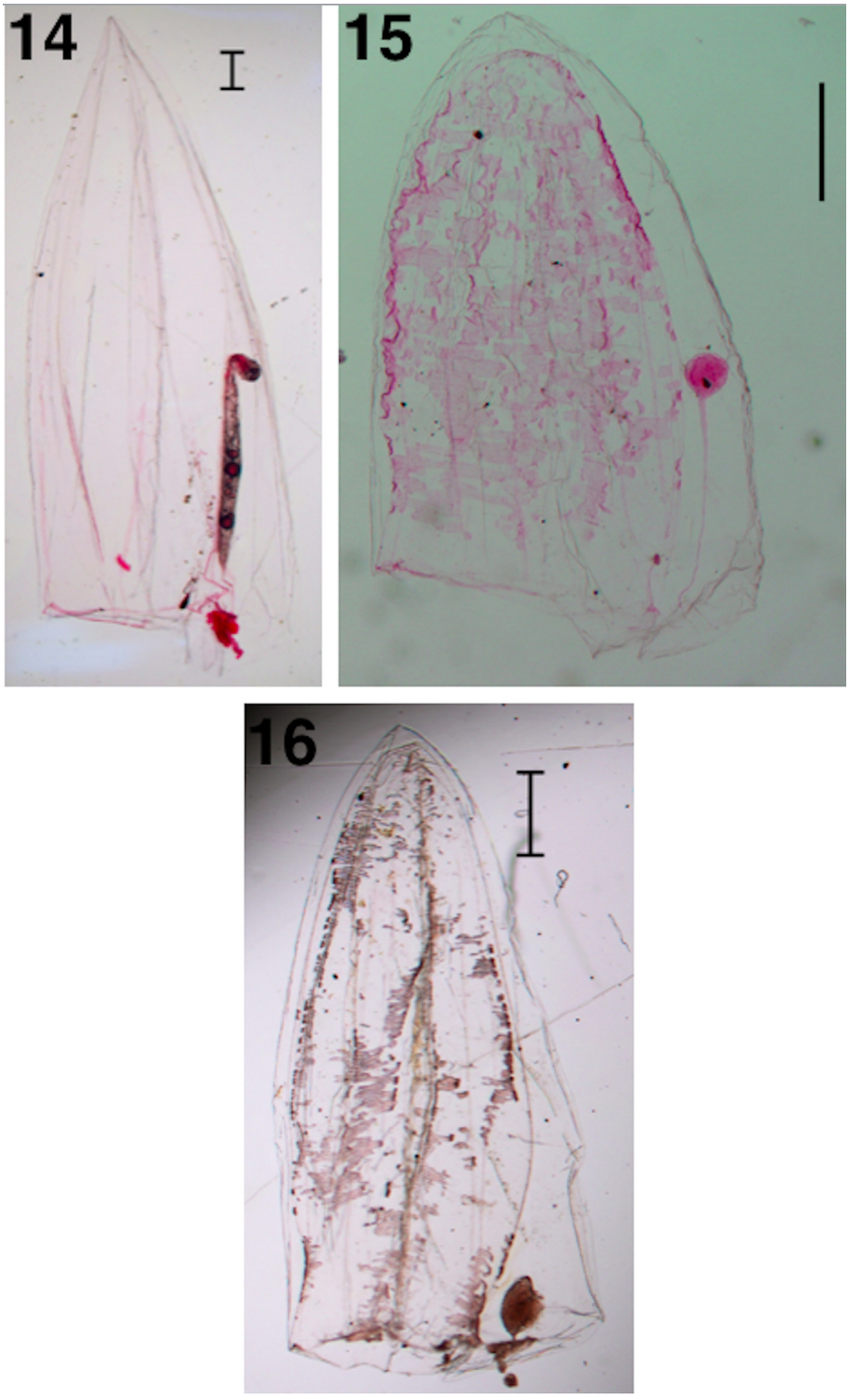
FIGURES 2–5. 2—Lateral view of the anterior nectophore of *Lensia campanella* (scale: 0.5 mm); 3—Lateral view of the anterior nectophore of *Lensia conoidea* (scale: 0.5 mm); 4—Lateral view of the anterior nectophore of *Lensia cossack* (scale: 0.5 mm); 5—Lateral view of the anterior nectophore of *Lensia fowleri* (scale: 0.5 mm).



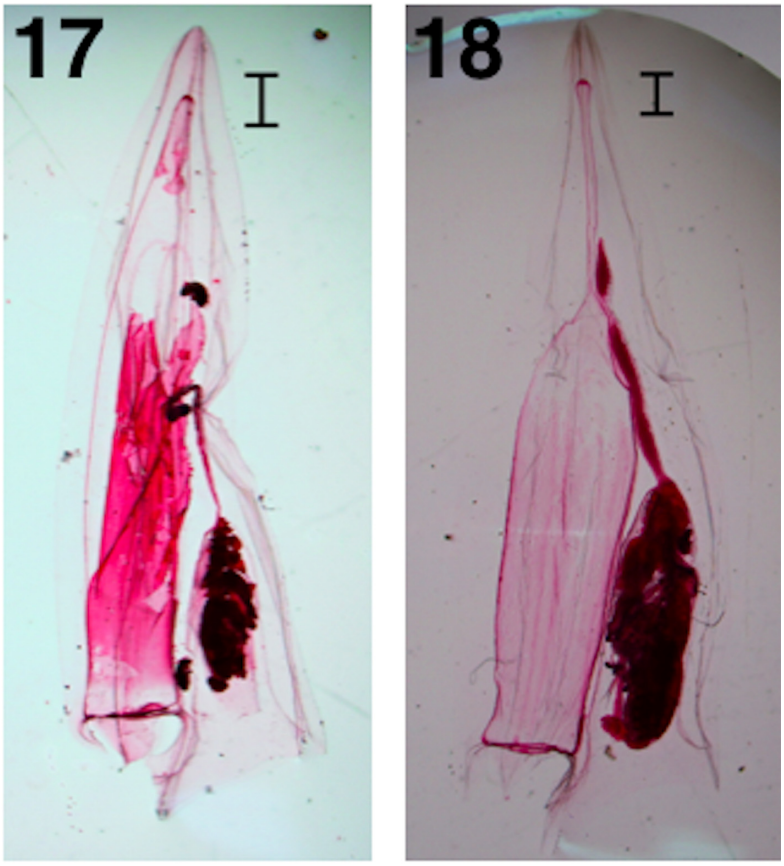
FIGURES 6–9. 6—Dorsal view of the anterior nectophore of *Lensia grimaldii* (scale: 0.5 mm); 7—Lateral view of the anterior nectophore of *Lensia hardy* (scale: 0.5 mm); 8—Lateral view of the anterior nectophore of *Lensia havock* (scale: 0.5 mm); 9—Lateral view of the anterior nectophore of *Lensia hotspur* (scale: 0.5 mm).

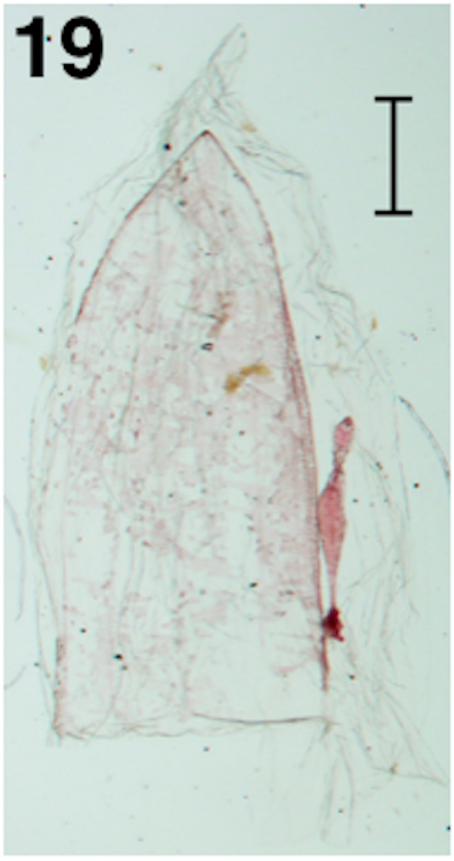


FIGURES 10–13. 10—Ventral view of the anterior nectophore of *Lensia hunter* (scale: 0.5 mm); 11—Lateral view of the anterior nectophore of *Lensia leloupi* (scale: 0.5 mm); 12—Lateral view of the anterior nectophore of *Lensia lelouveteau* (scale: 0.5 mm); 13—Lateral view of the anterior nectophore of *Lensia meteori* (scale: 0.5 mm).



FIGURES 14–16. 14—Lateral view of the anterior nectophore of *Lensia multicristata* (scale: 0.5 mm); 15—Lateral view of the anterior nectophore of *Lensia subtilis* (scale: 0.5 mm); 16—Lateral view of the anterior nectophore of *Lensia subtiloides* (scale: 0.5 mm).





FIGURES 17–19. 17—Lateral view of the anterior nectophore of *Diphyes bojani* (scale: 0.5 mm); 18—Lateral view of the anterior nectophore of *Diphyes dispar* (scale: 0.5 mm); 19—Lateral view of the anterior nectophore of *Muggiaea kochi* (scale: 0.5 mm).

Twenty two characters of the anterior nectophore morphology were scored for 15 species of *Lensia* from southeastern Brazilian waters, plus two species of *Diphyes* and one species of *Muggiaea*, both belonging to the family Diphyidae, as outgroups. The inapplicable, incomparable and unobserved characters were treated as missing data. The resulting data matrix is shown in Table 1.

The parsimony analysis was performed with the TNT software (Goloboff *et al*. 2003), using heuristic searches with tree bisection reconnection (TBR) branch swapping, random stepwise addition sequence and 10.000 replicates holding up to 1000 trees per replication. The characters were polarized *a posteriori* through rooting, following the outgroup method revised by Nixon & Carpenter (1993). In all cases, characters were treated as unordered and the shortest trees were searched using implied weighting, attributing different weights to characters together with tree construction (Goloboff 1993). Such analysis was done combining different values of the weighting constant k (from 2 to 6), evaluated independently through several different parsimony analyses. Additionally, in order to evaluate the relative supports of clades, a bootstrap analysis was performed using the standard option (sample with replacement) provided in TNT. Boostrap values were calculated with 1000 replicates using Traditional Search, and collapsing clades with values below 1. Output results are described as frequency differences.

Results and discussion

Character descriptions:

1. Longitudinal ridges: (0) Not evident; (1) Evident

Set of lines that run through the anterior nectophore, usually from its apex to its base (Figure 1). Also called lateral ridges. In some species, the ridges are not evident (Figure 31).

2. Number of longitudinal ridges: (0) 7 or less; (1) More than 7

Species of the genus *Lensia* present a variable number of longitudinal ridges, usually 5 or 7. However some species present more than 7 longitudinal ridges (Pugh 1999).

3. Incomplete ridges (i.r.): (0) Absent; (1) Present

Ridges that do not extend over the entire height of the anterior nectophore, not reaching the apex or the base of the same (Figure 29).

4. Number of crests per ridge: (0) Ridges with one crest; (1) Ridges with multiple crests

One-crested ridges are similar to simple lines and multiple-crested ridges are similar to lines that split into bifurcations or trifurcations.

5. Ventro-lateral ridges (v.l.r.): (0) Absent; (1) Present

Lateral ridges located at the ventral side (containing the somatocyst and mouth plate) of the anterior nectophore. They are present in seven-ridged species (Figure 26).

6. Extension of ventro-lateral ridges: (0) Do not reach mouth plate; (1) Reach mouth plate

Ventro-lateral ridges may reach the mouth plate (Figure 26).

7. Base of lateral ridges (b.l.r.): (0) Curved dorsally; (1) Continuous basally

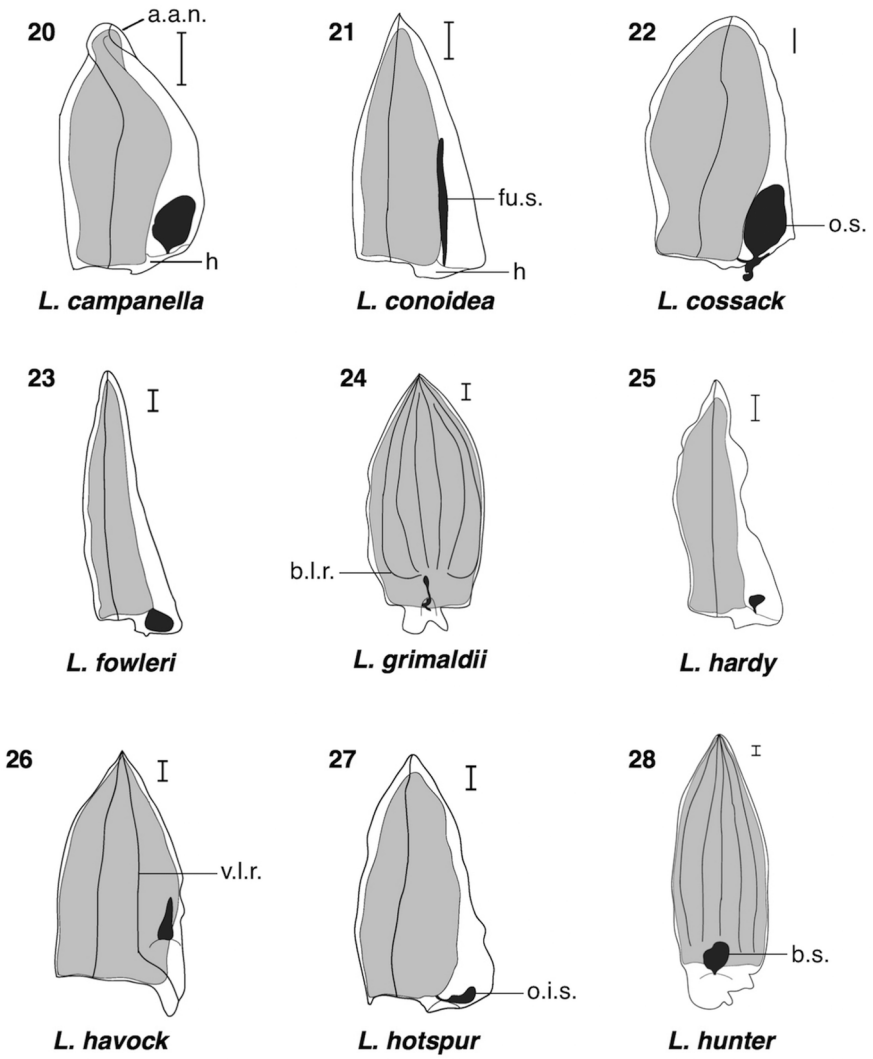
Most species present straight ridges, complete or incomplete, but, in some species, the base may be curved towards the dorsal region of the anterior nectophore (Figure 24).

8. Velar ridge (v.r.): (0) Absent; (1) Present

Transversal ridge present in some species, which is parallel to the ostium (Figure 30).

9. Apex of anterior nectophore (a.a.n.): (0) Not twisted; (1) Twisted

The apex of the anterior nectophore may be twisted, which is characteristic of preserved specimens of *Lensia campanella* (Pugh 1999) (Figure 20).



FIGURES 20–28. Scheme showing the anterior nectophore morphology of the analyzed species of *Lensia* (scales: 0,5 mm). Legend: a.a.n.: apex of anterior nectophore; b.l.r.: base of lateral ridge; b.s.: bilobed somatocyst; fu.s.: fusiform somatocyst; h: hydroecium; o.i.s.: obliquely inclined somatocyst; v.l.r.: ventro-lateral ridge.

10. Apex of nectosac: (0) Not narrow; (1) Narrow

The apex of the nectosac is usually similar in shape to the apex of the anterior nectophore. However, in *Diphyes dispar*, an outgroup used in this study, the apex of the nectosac is narrow (Figure 36).

11. Shape of somatocyst: (0) Spherical (s.s.); (1) Ovoid (o.s.); (2) Bilobed (b.s.); (3) Fusiform (fu.s.); (4) Filiform (fi.s.); (5) Club-shaped (c.s.); (6) Kidney-shaped (k.s.)

The somatocyst shape is variable. In the analyzed species, it can be spherical (Figure 33), ovoid (Figure 22), bilobed (Figure 28), fusiform (Figure 21), filiform (Figure 32), club-shaped (Figure 34) or kidney-shaped (Figure 30).

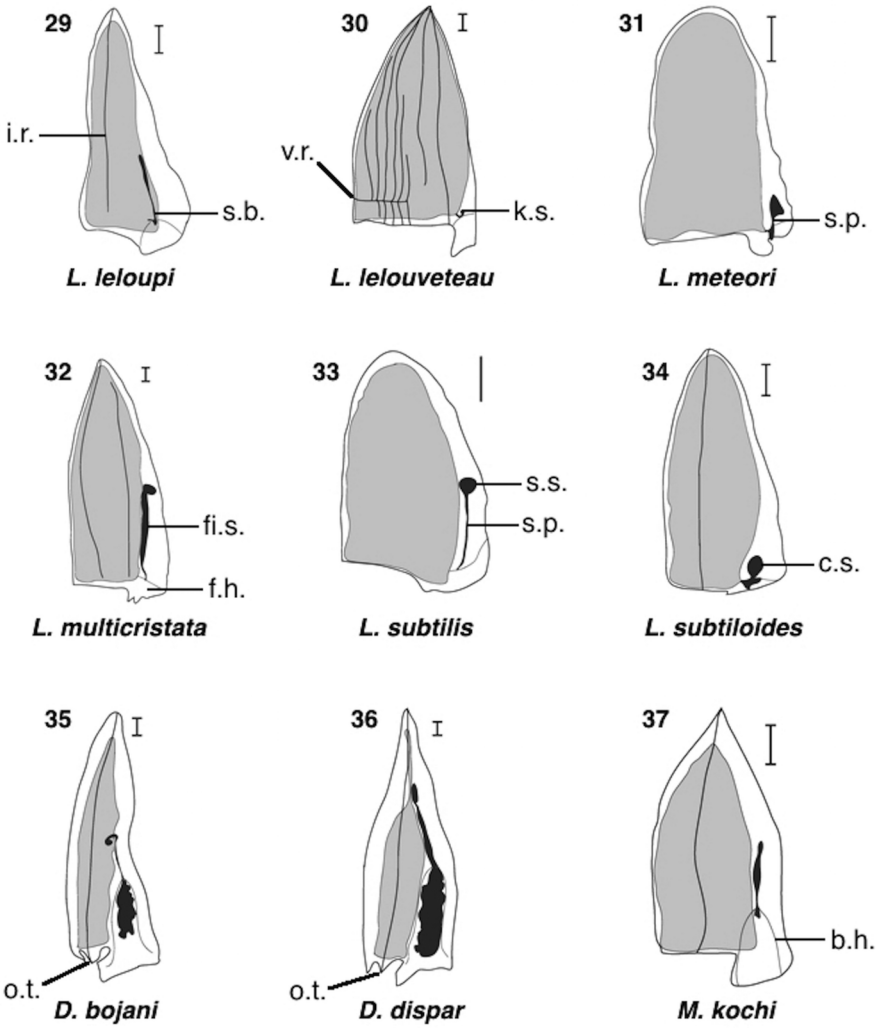
12. Obliquely inclined somatocyst (o.i.s.): (0) Absent; (1) Present

Somatocyst inclined in an approximately oblique angle, in relation to the anterior nectophore (Figure 27).

13. Somatocyst peduncle (s.p.): (0) Absent; (1) Present

Somatocyst with a peduncle at its base (Figure 31).

14. Size of somatocyst peduncle: (0) Up to 1/2 the length of the somatocyst; (1) More than 1/2 the length of the somatocyst



FIGURES 29–37. Scheme showing the anterior nectophore morphology of the analyzed species of *Lensia*, plus the outgroups (scales: 0,5 mm). Legend: b.h.: bell-shaped hydroecium; c.s.: club-shaped somatocyst; f.h.: flattened hydroecium; fi.s.: filiform somatocyst; i.r.: incomplete ridge; k.s.: kidney-shaped somatocyst; o.t.: ostial tooth; s.b.: somatocyst base; s.p.: somatocyst peduncle; s.s.: spherical somatocyst; v.r.: velar ridge.

The peduncle size is variable. In some species, it measures up to half the length of the somatocyst (Figure 31) while in others, it measures more than half the length of the somatocyst (Figure 33).

15. Somatocyst length: (0) Up to 1/3 the length of the nectophore; (1) More than 1/3 the length of the nectophore

The length of the somatocyst is variable. In some species, it measures up to 1/3 the length of the nectophore (Figure 22) while in others, it measures more than 1/3 the length of the nectophore (Figure 32).

16. Location of somatocyst: (0) Below or at same level of ostium; (1) Above level of ostium

The somatocyst may be located below or at the same level of the ostium (Figure 23), or located partially or completely above the level of the ostium (Figure 26).

17. Location of somatocyst base (s.b.): (0) Below or at same level of ostium; (1) Above level of ostium

The base of the somatocyst may be located below or at the same level of the ostium (Figure 25), or located above the level of the ostium (Figure 29).

18. Ostial teeth (o.t.): (0) Absent; (1) Present

The ostial teeth are pointed projections arising from the base of the anterior nectophore. They are only found in the outgroups (*Diphyes bojani* and *Diphyes dispar*) (Figures 35 and 36).

19. Hydroecium: (0) Not open ventrally; (1) Open ventrally

In some species, the hydroecium may present a small opening on its ventral side.

20. Hydroecium shape: (0) Flattened (f.h.); (1) Bell-shaped (b.h.)

The hydroecium in most species of *Lensia* is flattened (Figure 32). However, it may be bell-shaped (Figure 37).

21. Extension of hydroecium: (0) Below or at same level of ostium; (1) Above level of ostium

The hydroecium may be located below or at the same level of the ostium (Figure 21), or may extend above the level of the ostium (Figure 20).

22. Hydroecium length: (0) Up to 1/4 the height of the nectosac; (1) More than 1/4 the height of the nectosac

The hydroecium in *Lensia* species is usually small, measuring up to 1/4 the height of the nectosac in the analyzed specimens (Figure 21). In other genera, it may be deep, measuring more than 1/4 the height of the nectosac (Figure 35).

TABLE 1. Data matrix of the phylogenetic analysis. Inapplicable, incomparable and unobserved character states are coded as "-".

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
L. campanella	1	0	0	0	0	-	-	0	1	0	1	1	1	0	0	1	0	0	0	0	1	0
L. conoidea	1	0	0	0	0	-	1	0	0	0	3	0	0	-	1	1	0	0	0	0	0	0
L. cossack	1	0	0	0	0	-	1	0	0	0	1	1	1	0	0	1	0	0	0	0	0	0
L. fowleri	1	0	0	0	0	-	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
L. grimaldii	1	1	1	1	0	-	0	1	0	0	5	0	1	0	0	1	0	0	1	0	1	0
L. hardy	1	0	0	0	0	-	1	0	0	0	0	0	1	0	0	1	0	0	0	0	0	0
L. havock	1	0	0	0	1	1	1	0	0	0	3	0	0	-	0	1	1	0	1	0	1	0
L. hotspur	1	0	0	0	0	-	1	0	0	0	1	1	1	0	0	1	0	0	0	0	0	0
L. hunter	1	0	1	0	1	1	1	0	0	0	2	0	1	0	0	1	0	0	1	0	0	0
L. leloupi	1	0	1	0	0	-	1	0	0	0	4	0	0	-	0	1	1	0	0	1	1	0
L. lelouveteau	1	1	1	1	1	1	1	1	0	0	6	0	1	0	0	1	0	0	1	0	1	0
L. meteori	0	0	-	-	-	-	-	-	0	0	0	0	1	0	0	1	0	0	0	0	1	0
L. multicristata	1	0	1	0	1	0	1	0	0	0	4	0	1	0	1	1	0	0	0	0	0	0
L. subtilis	0	0	-	-	-	-	-	-	0	0	0	0	1	1	0	1	0	0	0	0	1	0
L. subtiloides	1	0	0	0	0	-	1	0	0	0	5	1	1	0	0	1	0	0	0	0	0	0
D. bojani	1	0	0	0	0	-	1	0	0	0	3	0	0	-	0	1	1	1	0	1	1	1
D. dispar	1	0	0	0	0	-	1	0	0	1	3	0	0	-	0	1	1	1	0	1	1	1
M. kochi	1	0	0	0	0	-	1	0	0	0	4	0	1	1	1	1	1	0	0	1	1	1

Phylogeny. All analyses of the data matrix with different weighting constants (k varying from 2 to 6) yielded four most parsimonious cladograms, with 39 steps. The fittest trees were those obtained under k=2. The strict consensus of the cladograms obtained in each analysis showed the same topology and the same character distribution (Figure 38).

Clade A. The monophyly of the clade including all species of *Lensia* analyzed is supported by the hydroecium measuring up to 1/4 the height of the nectosac (character 22, state 0). The species *L. leloupi* is the sister group to all other species of *Lensia* included in the present study, and is supported by the presence of incomplete ridges (character 3, state 1), which is a homoplastic character.

Clade B. This clade is supported by the flattened hydroecium (character 20, state 0). The species *L. havock* is supported by the presence of ventro-lateral ridges (character 5, state 1) and ventrally open hydroecium (character 19, state 1). These characters are homoplastic, appearing independently in other species.

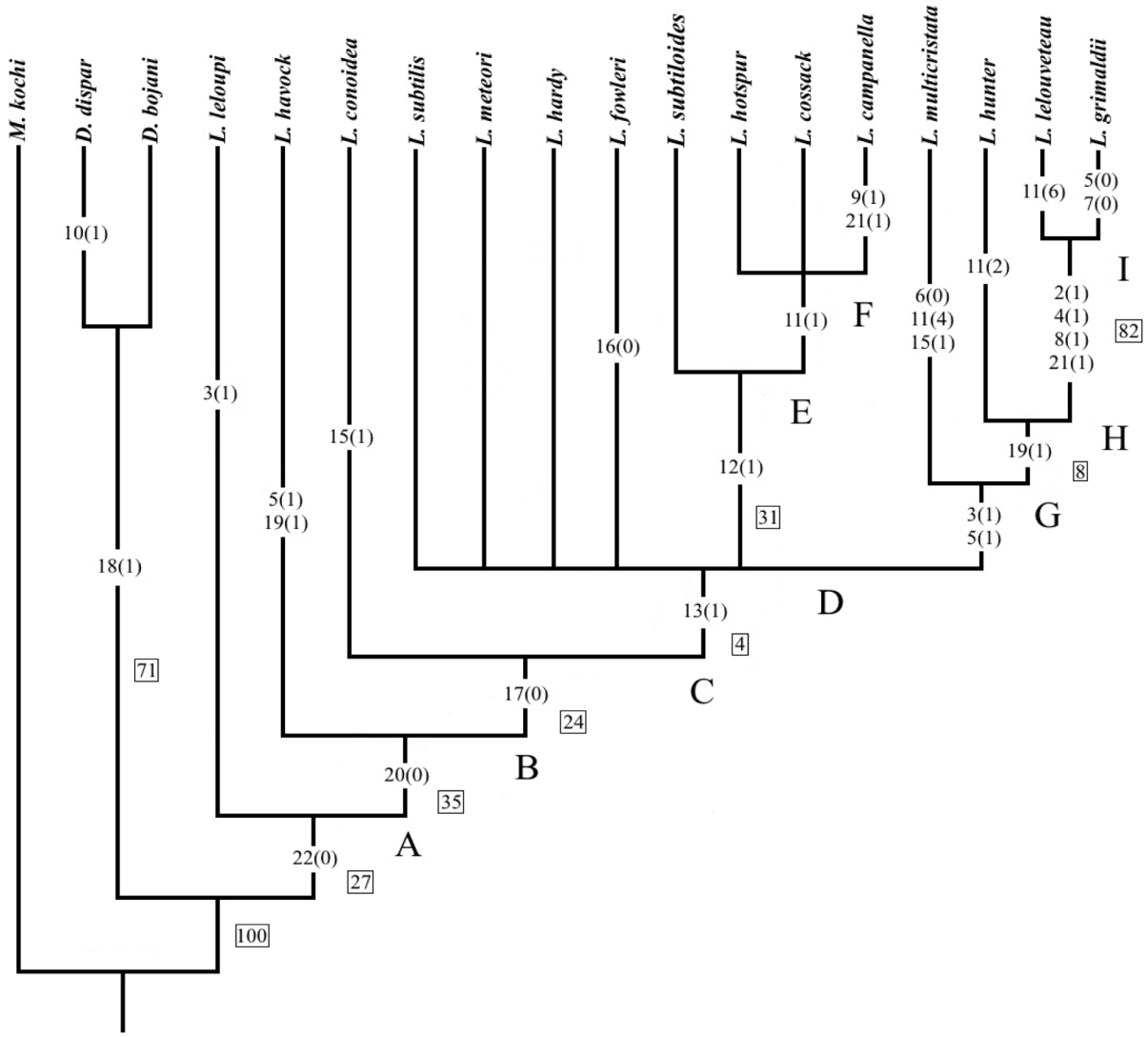


FIGURE 38. Phylogenetic relationships of *Lensia* species from southeastern Brazilian waters, obtained from analysis of the data matrix in Table 1 (k varying from 2 to 6). Each clade discussed in text is identified by a letter (A to I). Numbers refer to the characters and their states (in parenthesis). Numbers inside squares refer to bootstrap values.

Clade C. The somatocyst base located above the level of the ostium is probably plesiomorphic for *Lensia*. The somatocyst base located below or at the same level of the ostium (character 17, state 0) is a synapomorphy supporting this clade. The species *L. conoidea* is supported by the somatocyst measuring more than 1/3 the height of the nectophore (character 15, state 1), which is homoplastic, as it appears independently in *L. multicristata*.

Clade D. The monophyly of this clade is supported by the presence of somatocyst peduncle (character 13, state 1). The somatocyst located below the ostium level (character 16, state 0) is an autapomorphy of *L. fowleri*. The relationships within this clade are not clear as it contains several polytomies.

Clade E. This clade is supported by the presence of obliquely inclined somatocyst (character 12, state 1). The species *L. subtiloides* is sister group to *L. campanella*, *L. cossack* and *L. hotspur*.

Clade F. This clade is well supported by the ovoid somatocyst (character 11, state 1). The relationships between *L. hotspur*, *L. cossack* and *L. campanella* are not well established and form a polytomy. The twisted apex of the anterior nectophore (character 9, state 1) is autapomorphic for *L. campanella*, while the hydroecium extending above the level of the ostium (character 21, state 1) is homoplastic.

Clade G. The presence of incomplete longitudinal ridges (character 3, state 1) and the presence of ventro-lateral ridges (character 5, state 1) are synapomorphies of this clade. The ventro-lateral ridges not reaching the

mouth plate (character 6, state 0) is autapomorphic for *L. multicristata*, while the filiform somatocyst (character 11, state 4) and the somatocyst measuring more than 1/3 the height of the nectophore (character 15, state 1) are homoplastic.

Clade H. The monophyly of this clade is supported by the ventrally open hydroecium (character 19, state 1), although it appears independently in some species. The bilobed somatocyst (character 11, state 2) is an autapomorphy of *L. hunter*.

Clade I. This clade is well supported by the nectophore with more than seven ridges (character 2, state 1), presence of ridges with multiple crests (character 4, state 1), and presence of velar ridge (character 8, state 1). It is also supported by the hydroecium extending above the level of the ostium (character 21, state 1), which appears independently in several species. The kidney-shaped somatocyst (character 11, state 6) is an autapomorphy of *L. lelouveteau*. The base of lateral ridges curved dorsally (character 7, state 0) is autapomorphic for *L. grimaldii*.

Conclusion

Despite the advances we have reached in this study concerning the phylogenetic relationships and the morphological evolution within the genus *Lensia*, the results are still biased by a limited taxon and character sampling. Only 58% of the total of *Lensia* species were included, and the comparative morphological study was restricted to the anterior nectophore. In addition, for some species, there was only one specimen available, which was damaged or poorly preserved, hampering the estimation of new characters that could improve the resolution of the phylogenetic analysis. The study of the entire polygastric stage (including anterior and posterior nectophores, bracts and eudoxids) could be helpful in improving resolution, as more information would be added.

The majority of studies about siphonophore phylogeny are focused on supraspecific taxon relationships. The consensus tree obtained by Dunn *et al*. (2005) was based on data obtained from two genes: the nuclear gene 18S and mitochondrial gene 16S. Their analysis included only one species of *Lensia*, and did not provide information about the genus' internal relationships.

The results of the present study are not congruent with those of the recent investigation conducted by Grossmann *et al*. (2014), who used molecular data to estimate a phylogeny of 12 species of *Lensia* from different areas (mainly Japan). Only 7 of the 15 species included in our study were also included in the analysis of Grossmann *et al*. (2014). Their analysis resulted in two distinct clades of *Lensia* (one containing the five-ridged species and the other containing species with indistinct ridges or more than five ridges). Our result shows that species of this genus form a clade based on the hydroecium length. The only noticeable congruence in both cladograms is that the species *L. campanella* and *L. cossack* are closely related to each other. The monophyly of *Lensia* is supported, in the present study, by the hydroecium measuring up to 1/4 the height of the nectosac. However, considering that our outgroup sampling is reduced, it would be important to test this hypothesis in the future including a larger sample of Diphyidae genera. Therefore, further investigations are necessary to improve the knowledge of the group's phylogeny.

Acknowledgements

This research was sponsored by a Master's fellowship to E.Y.N. provided by CAPES and UFABC. Laboratory facilities used for this study were provided by Fapesp (Grant # 2010/00557-7 to G.C.R).

References

Alvariño, A. & Wojtan, J.M. (1984) Three new species of *Lensia*, and description of the eudoxia stages of *Lensia reticulata* and *Lensia lelouveteau* (Calycophorae: Siphonophorae). *Proceedings of the Biological Society of Washington*, 97 (1), 49–59.

Dunn, C.W. & Wagner, P.G. (2006) The evolution of colony-level development in the Siphonophora (Cnidaria: Hydrozoa). *Development Genes and Evolution*, 216, 743–754. http://dx.doi.org/10.1007/s00427-006-0101-8

Dunn, C.W., Pugh, P.R. & Haddock, S.H.D. (2005) Molecular phylogenetics of the Siphonophora (Cnidaria), with implications

for the evolution of functional specialization. *Systematic Biology*, 54, 916–935.
http://dx.doi.org/10.1080/10635150500354837
Goloboff, P.A. (1993) Estimating character weights during tree search. *Cladistics*, 9, 83–91.
http://dx.doi.org/10.1111/j.1096-0031.1993.tb00209.x
Goloboff, P.A., Farris, J.S. & Nixon, K.C. (2003) TNT: Tree analysis using new technology. Distributed by the authors. Available from: www.zmuc.dk/public/phylogeny (accessed 14 June 2016)
Grossmann, M.M., Collins, A.G. & Lindsay, D.J. (2014) Description of the eudoxid stages of *Lensia havock* and *Lensia leloupi* (Cnidaria: Siphonophora: Calycophorae), with a review of all known *Lensia* eudoxid bracts. *Systematics and Biodiversity*, 12 (2), 163–180.
http://dx.doi.org/10.1080/14772000.2014.902867
Kirkpatrick, P.A. & Pugh, P.R. (1984) Siphonophores and Velellids. *Synopses of the British Fauna New Series*, 29, 1–154.
Mackie, G.O. (1964) Analysis of locomotion in a siphonophore colony. *Proceedings of the Royal Society B: Biological Sciences*, 159, 366–391.
http://dx.doi.org/10.1098/rspb.1964.0008
Mapstone, G.M. (2014) Global diversity and review of the Siphonophorae. *PLOS One*, 9 (2), 1–37.
http://dx.doi.org/10.1371/journal.pone.0087737
Nixon, K.C. & Carpenter, J.M. (1993) On outgroups. *Cladistics*, 9, 413–426.
http://dx.doi.org/10.1111/j.1096-0031.1993.tb00234.x
Pagès, F. & Gili, J.M. (1992) Siphonophores (Cnidaria, Hydrozoa) of the Benguela Current (southeastern Atlantic). *Scientia Marina*, 56, 65–112.
Pugh, P.R. (1999) Siphonophorae. *In*: Boltvoskoy, D. (Ed.), *Zooplankton. Vol. 1*. Backhuys Publishers, Leiden, pp. 467–513.
Totton, A.K. (1932) Siphonophora. *Scientific Reports of the Great Barrier Reef Expedition*, 4 (10), 317–374.
Totton, A.K. (1965) *A Synopsis of the Siphonophora*. Trustees of the British Museum (Natural History), London, 230 pp.

APPENDIX 1. Details on voucher specimens analyzed in the study.

Genus *Diphyes* Cuvier, 1817

***Diphyes bojani* (Eschscholtz, 1825)**

Examined material. Unknown date, Habitats 12 campaign, station I12, Tropical Water, 1 m depth, collected in Campos Basin, 200 μm net, one specimen preserved in 4% formaldehyde.

***Diphyes dispar* Chamisso & Eysenhardt, 1821**

Examined material. Unknown date, Habitats 14 campaign, station H5, Tropical Water, 1 m depth, collected in Campos Basin, 200 μm net, one specimen preserved in 4% formaldehyde.

Genus *Muggiaea* Busch, 1851

***Muggiaea kochi* (Will, 1844)**

Examined material. 05/15/76, collected at 4:27 a.m., 128 m depth, between Cold Cape and Santa Marta Grande Cape (25°30.5'S–45°26.8'W), FINEP III campaign, station 1758, three specimens preserved in 4% formaldehyde.

Genus *Lensia* Totton, 1932

***Lensia campanella* (Moser, 1917)**

Examined material. 05/09/76, collected at 5:45 a.m., 2721 m depth, between Cold Cape and Santa Marta Grande Cape (24°24.8'S–41°07.5'W), FINEP III campaign, station 1701, one specimen preserved in 4% formaldehyde;

05/10/76, collected at 1:00 a.m., 1120 m depth, between Cold Cape and Santa Marta Grande Cape (23°55.5'S–41°43.0'W), FINEP III campaign, station 1708, three specimens preserved in 4% formaldehyde;

05/19/76, collected at 5:00 a.m., 64 m depth, between Cold Cape and Santa Marta Grande Cape (25°13.6'S–46°42.2'W), FINEP III campaign, station 1770, two specimens preserved in 4% formaldehyde.

***Lensia conoidea* (Keferstein & Ehlers, 1860)**

Examined material. 05/20/76, collected at 7:15 a.m., 185 m depth, between Cold Cape and Santa Marta Grande Cape (25°55.2'S–46°00.0'W), FINEP III campaign, station 1780, one specimen preserved in 4% formaldehyde;

03/25/09, Habitats 14 campaign, station F10, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 200 μm net, four specimens preserved in 4% formaldehyde.

***Lensia cossack* Totton, 1941**

Examined material. 05/09/76, collected at 5:45 a.m., 2721 m depth, between Cold Cape and Santa Marta Grande Cape (24°24.8'S–41°07.5'W), FINEP III campaign, station 1701, one specimen preserved in 4% formaldehyde;

05/10/76, collected at 1:00 a.m., 1120 m depth, between Cold Cape and Santa Marta Grande Cape (23°55.5'S–41°43.0'W), FINEP III campaign, station 1708, two specimens preserved in 4% formaldehyde;

05/20/76, collected at 7:15 a.m., 185 m depth, between Cold Cape and Santa Marta Grande Cape (25°55.2'S–46°00.0'W), FINEP III campaign, station 1780, four specimens preserved in 4% formaldehyde;

03/23/09, Habitats 14 campaign, station D10, Tropical Water, 1 m depth, collected in Campos Basin, 120 μm net, one specimen preserved in 4% formaldehyde.

***Lensia fowleri* (Bigelow, 1911)**

Examined material. 05/12/76, collected at 6:21 a.m., unknown depth, between Cold Cape and Santa Marta Grande Cape (24°16.3'S - 43°49.1'W), FINEP III campaign, station 1729, one specimen preserved in 4% formaldehyde.

***Lensia grimaldii* Leloup, 1933**

Examined material. 08/27/09, Habitats 18 campaign, station D8, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 200 μm net, one specimen preserved in 4% formaldehyde.

***Lensia hardy* Totton, 1941**

Examined material. 05/24/76, collected at 8:00 a.m., 178 m depth, between Cold Cape and Santa Marta Grande Cape (28°13.0'S–47°26.5'W), FINEP III campaign, station 1819, one specimen preserved in 4% formaldehyde.

***Lensia havock* Totton, 1941**

Examined material. 03/05/09, Habitats 12 campaign, station A12, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 200 μm net, one specimen preserved in 4% formaldehyde;

08/20/09, Habitats campaign (unknown campaign number), station A8, Antarctic Intermediate Water, 800 m depth, collected in Campos Basin, 120 μm net, two specimens preserved in 4% formaldehyde.

***Lensia hotspur* Totton, 1941**

Examined material. 03/18/09, Habitats 12 campaign, station A8, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 200 μm net, two specimens preserved in 4% formaldehyde.

***Lensia hunter* Totton, 1941**

Examined material. 03/05/09, Habitats 12 campaign, station A12, Antarctic Intermediate Water, 800 m depth, collected in Campos Basin, 120 μm net, one specimen preserved in 4% formaldehyde;

08/03/09, Habitats 18 campaign, station C12, Antarctic Intermediate Water, 800 m depth, collected in Campos Basin, 120 μm net, one specimen preserved in 4% formaldehyde.

***Lensia leloupi* Totton, 1954**

Examined material. 08/20/09, Habitats 18 campaign, station A8, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 120 µm net, one specimen preserved in 4% formaldehyde.

***Lensia lelouveteau* Totton, 1941**

Examined material. 08/09/09, Habitats campaign (unknown campaign number), station C12, Antarctic Intermediate Water, 800 m depth, collected in Campos Basin, 120 µm net, one specimen preserved in 4% formaldehyde.

***Lensia meteori* (Leloup, 1934)**

Examined material. 05/10/76, collected at 1:00 a.m., 1120 m depth, between Cold Cape and Santa Marta Grande Cape (23°55.5'S–41°43.0'W), FINEP III campaign, station 1708, three specimens preserved in 4% formaldehyde;

03/22/09, Habitats 14 campaign, station D8, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 120 µm net, four specimens preserved in 4% formaldehyde;

***Lensia multicristata* (Moser, 1925)**

Examined material. 05/09/76, collected at 5:45 a.m., 2721 m depth, between Cold Cape and Santa Marta Grande Cape (24°24.8'S–41°07.5'W), FINEP III campaign, station 1701, one specimen preserved in 4% formaldehyde;

03/25/09, Habitats 14 campaign, station F10, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 120 µm net, one specimen preserved in 4% formaldehyde.

***Lensia subtilis* (Chun, 1886)**

Examined material. 05/09/76, collected at 5:45 a.m., 2721 m depth, between Cold Cape and Santa Marta Grande Cape (24°24.8'S–41°07.5'W), FINEP III campaign, station 1701, one specimen preserved in 4% formaldehyde;

05/10/76, collected at 1:00 a.m., 1120 m depth, between Cold Cape and Santa Marta Grande Cape (23°55.5'S–41°43.0'W), FINEP III campaign, station 1708, one specimen preserved in 4% formaldehyde;

05/24/76, collected at 12:20 a.m., 1170 m depth, between Cold Cape and Santa Marta Grande Cape (28°29.0'S–46°46.0'W), FINEP III campaign, station 1821, one specimen preserved in 4% formaldehyde;

03/17/09, Habitats 12 campaign, station A10, Upper Circumpolar Water, 1200 m depth, collected in Campos Basin, 120 µm net, one specimen preserved in 4% formaldehyde.

***Lensia subtiloides* (Lens & van Riemsdijk, 1908)**

Examined material. 05/12/76, collected at 6:21 a.m., 240 m depth, between Cold Cape and Santa Marta Grande Cape (24°16.3'S–43°49.1'W), FINEP III campaign, station 1729, two specimens preserved in 4% formaldehyde;

05/13/76, collected at 2:10 a.m., 186 m depth, between Cold Cape and Santa Marta Grande Cape (24°19.3'S–44°17.0'W), FINEP III campaign, station 1737, one specimen preserved in 4% formaldehyde;

05/15/76, collected at 4:27 a.m., 128 m depth, between Cold Cape and Santa Marta Grande Cape (25°30.5'S–45°26.8'W), FINEP III campaign, station 1758, one specimen preserved in 4% formaldehyde;

05/20/76, collected at 7:15 a.m., 185 m depth, between Cold Cape and Santa Marta Grande Cape (25°55.2'S–46°00.0'W), FINEP III campaign, station 1780, two specimens preserved in 4% formaldehyde;

05/21/76, collected at 12:50 p.m., 218 m depth, between Cold Cape and Santa Marta Grande Cape (26°24.5'S–46°33.3'W), FINEP III campaign, station 1791, two specimens preserved in 4% formaldehyde;

05/23/76, collected at 13:30 p.m., 230 m depth, between Cold Cape and Santa Marta Grande Cape (27°33.9'S–47°17.1'W), FINEP III campaign, station 1811, two specimens preserved in 4% formaldehyde;

05/24/76, collected at 12:20 a.m., 1170 m depth, between Cold Cape and Santa Marta Grande Cape (28°29.0'S–46°46.0'W), FINEP III campaign, station 1821, one specimen preserved in 4% formaldehyde;

05/25/76, collected at 4:00 a.m., 640 m depth, between Cold Cape and Santa Marta Grande Cape (28°41.7'S–47°16.0'W), FINEP III campaign, station 1827, two specimens preserved in 4% formaldehyde;

05/25/76, collected at 9:00 a.m., 128 m depth, between Cold Cape and Santa Marta Grande Cape (28°23.0'S–47°55.0'W), FINEP III campaign, station 1829, two specimens preserved in 4% formaldehyde;

08/09/09, Habitats 18 campaign, station C12, South Atlantic Central Water, 250 m depth, collected in Campos Basin, 120 µm net, one specimen preserved in 4% formaldehyde.