Contribution to taxonomy and distribution of the genus *Vestalenula* ROSSETTI & MARTENS, 1998 (Crustacea, Ostracoda, Darwinulidae), with the description of two new species

by Giampaolo ROSSETTI & Koen MARTENS

### **Abstract**

Two new species, Vestalenula flexuosa spec.nov. from Namibia and Vestalenula marmonieri spec.nov. from New Caledonia, are described. Two further species (from a spring near the Dead Sea in Israel and from Lake Sibaya in South Africa) in the same genus, most likely new, are illustrated but are left in open nomenclature, as only one specimen for each of the taxa is available. Some further comments are offered on newly investigated type material of V. marlieri (KISS) and of V. daps (HARDING). The speciose genus Vestalenula has a world-wide distribution, but all species have restricted distributions. In spite of the fact that the ancient asexual darwinulids are known to evolve slowly, it is here nevertheless assumed that most speciation in the genus has occurred relatively recently.

**Key words**: Ostracods, *Vestalenula flexuosa* spec.nov., *Vestalenula marmonieri* spec.nov., morphology, taxonomy, ancient asexuals, parthenogenesis, biodiversity.

# Résumé

Deux nouvelles espèces sont décrites en provenance de Namibie, Vestalenula flexuosa, et de Nouvelle-Calédonie, Vestalenula marmonieri. Deux espèces additionnelles (dans une source près de la mer Morte en Israël et dans le lac Sibaya en Afrique du sud) appartenant au même genre, et très probablement nouvelles, sont également illustrées mais leur nomenclature est laissée ouverte, étant donné qu'un seul spécimen est disponible pour chaque taxon. Quelques informations supplémentaires sont fournies sur V. marlieri (KISS) et V. daps (HARDING) par un nouveau examen des types. Le genre Vestalenula a une distribution mondiale mais toutes les espèces ont une distribution restreinte. Bien que les darwinulides asexuées anciennes soient connues pour évoluer lentement, il est néanmoins supposé ici que l'essentiel de la spéciation s'est produit relativement récemment dans ce genre.

Mots clefs: ostracodes, *Vestalenula flexuosa* spec.nov., *Vestalenula marmonieri* spec.nov., morphologie, taxonomie, anciens asexués, parthénogenèse, biodiversité.

### Introduction

The extant members of the infraorder Darwinulocopina (see MARTENS 1992) all belong to one family, the Darwinulidae Brady & Norman, 1889. Rossetti & MARTENS (1998) recognized five genera within this family: the validity of the nominate genus Darwinula BRADY & ROBERTSON, 1885 and of the genus Microdarwinula Danielopol, 1968 was confirmed, while three new genera were described: Alicenula Rossetti & Martens, 1998; Penthesilenula Rossetti & Martens, 1998 and Vestalenula ROSSETTI & MARTENS, 1998. This revision summarized both old and new findings, which were scattered in the literature, for example contributions by DANIELOPOL (1968, 1970, 1980), MARTENS & ROSSETTI (1997), MARTENS et al. (1997), Rossetti et al. (1996, 1998a,b), Rossetti & MARTENS (1996), but taxonomic relationships within this group still remain to some extent unclear. Therefore, rather then being the endpoint of a research project, the revision of Rossetti & Martens (1998) will stimulate further detailed analysis of the morphology, the taxonomy, the history and present and past distribution of this group of ancient asexuals. The present contribution presents new data on two established species within the genus Vestalenula, describes two new species (from New Caledonia and Namibia) and two possible new species (from Israel and Lake Sibaya, South Africa) in the same genus. As from the latter two countries only one female each is known, the latter species are left in open nomenclature.

# Abbreviations used in text and figures

# Valves

Cp carapace

H height

L length

LV left valve

RV right valve

Ms muscle scar(s)

dv dorsal view

vv ventral view

lv lateral view

iv internal view

Limbs and soft parts

**A1** Antennula A2 Antenna Md Mandibula P-Abd Post-abdomen Fii Caudal ramus

hook-like process on A2 h exopodite on A2 exo

ventral aesthetasc clump A2 Acspecific setae on limbs  $\alpha$ , b, c,  $s_1$ ,  $s_2$ , t, w, x, y, z

aesthetascs  $y_{1-3}$ 

Nomenclature proposed by DANIELOPOL (1970), and adapted by Rossetti & Martens (1996, 1998) and ROSSETTI et al. (1996), is used in the description of chaetotaxy of soft parts.

### **Taxonomic Descriptions**

Ostracoda LATREILLE, 1806 Class Subclass Podocopa G.W. Müller, 1894 Order Podocopida SARS, 1866 Suborder Podocopina SARS, 1866 Infraorder Darwinulocopina SOHN, 1988 Darwinuloidea Brady & Norman, 1889 Superfamily

Family Darwinulidae Brady & Norman, 1889

Genus Vestalenula Rossetti & Martens, 1998

# Remarks:

- 1. The diagnosis of this genus remains as in ROSSETTI & MARTENS (1998). Descriptions of the species listed below are all abbreviated, as they conform to the typical structure of species of this genus as extensively described in MARTENS & ROSSETTI (1997) and ROSSETTI & MARTENS (1998).
- 2. All species described hereafter belong in the boteaigroup of the genus, as the postero-ventral keels on the RV and the internal antero-ventral teeth in the LV are short, not elongated.
- 3. As Danielopol (1980) already described species A and B, belonging to this genus, in open nomenclature, we name our species in open nomenclature in the present paper Vestalenula spec. C (Israel) and spec. D (South Africa).
- 4. As for all darwinulids, male individuals were consistently absent from all populations investigated.

# Vestalenula flexuosa spec.nov. (Figs 1A-H)

### TYPE LOCALITY

Agabeb fountain, a series of pools in a river, over c 500 m, on the road from Sesfontein to Opuwo, Kaokoland, Namibia. All specimens originate from the lowermost pool (c 2 x 2 m, 1 m deep), sample NAM/87/12 collected on 01.10.1987 by K. MARTENS & B. CURTIS. Approximate coordinates: S 18°51' E 14°00'. Accompanying ostracod fauna: Apatelecypris schultzei, Cypretta seurati, Plesiocypridopsis spec., Cyprilla mastigophora and Strandesia spec.

#### TYPE MATERIAL

About 25 carapaces without soft parts.

Holotype: a dried Cp in a micro-pal slide lodged in the ostracod collection of the Royal Belgian Institute of Natural Sciences (Brussels, Belgium) (no. OC2213). Paratypes: one dried Cp on a micro-pal slide SMN 51696 and 6 carapaces in alcohol are kept in the National Museum of Namibia (Windhoek, Namibia SMN 51690-51695); 6 carapaces in alcohol as well as four figured specimens on micropal slides are stored in the Ostracod Collection of the Royal Belgian Institute of Natural Sciences (Brussels, Belgium), nos. OC2214-2218.

### DERIVATION OF NAME

The species is named after the shape of the carapace in dorsal and ventral view (from the Latin flexuosus, meaning 'sinuous').

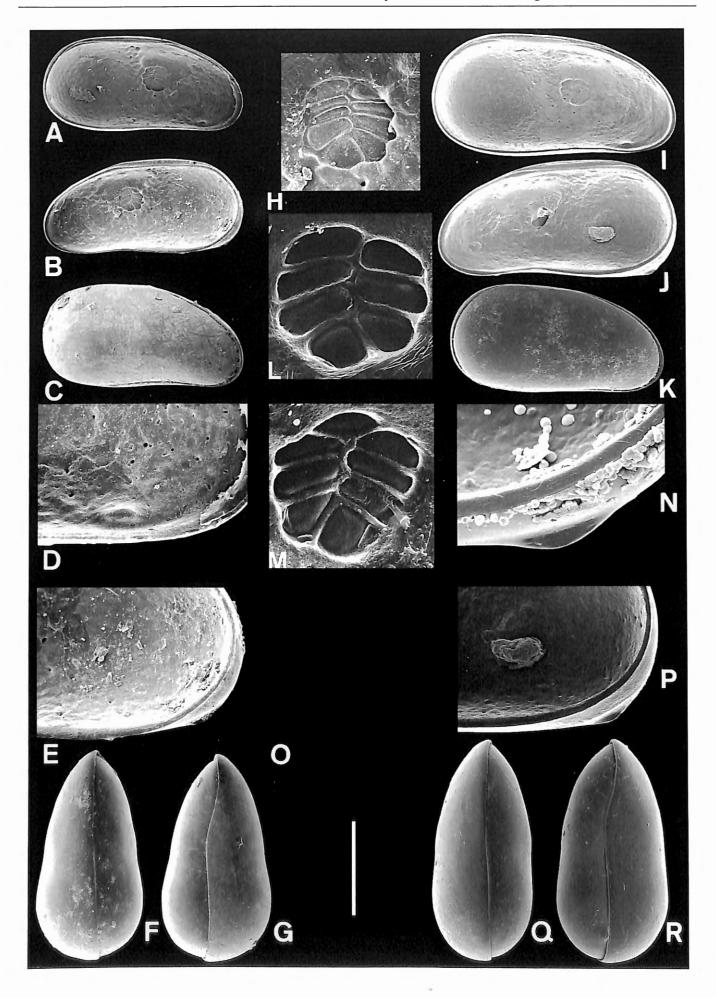
# ABBREVIATED DESCRIPTION

Carapace (Fig. 1C) and valves (Figs. 1A,B) in lateral view relatively short and high, with caudal margin broad, nearly straight and anterior margin asymmetrically produced towards the ventral side; straight part of dorsal side short (<1/3) and parallel to ventral side; ventral margin sinuous. Antero-ventral internal tooth in LV (Fig. 1D) rather small, no posterior tooth. Postero-ventral external keel on RV (Fig. 1E) weakly developed and positioned well towards the caudal side. 8-9 Ms (Fig. 1H), with similar pattern in both valves. In dorsal (Fig. 1F) and ventral (Fig. 1G) views, posterior half of body very swollen and anterior half pointed, central part of carapace constricted. Soft part morphology unknown.

Fig. 1 - A-H: Vestalenula flexuosa spec.nov. (Kaokoland, Namibia). I-R: Vestalenula molopoensis (MARTENS & ROSSETTI, 1997) (Molopo, North West Province, R. South Africa).

V. flexuosa spec. nov.: A. LV, iv (OC2218). B. RV, iv (idem). C. Cp, right lv (OC2215). D. LV, iv, detail antero-ventral corner (OC2218). E. RV, iv, detail postero-ventral corner (idem). F. Cp, dv (OC2216). G. Cp, vv (OC2217). H. RV, iv, detail central Ms (OC2218).

V. molopoensis: I. LV, iv (OC2101). J. RV, iv (idem). K. Cp, right lv (OC2228). L. RV, iv, detail central Ms (OC2098). M. LV, iv, detail central Ms (idem). N. RV, iv, detail postero-ventral corner (OC2227). O. Cp, vv, detail postero-ventral corner (OC2229). P. RV, iv, detail postero-ventral corner (OC2101). Q. Cp, dv (specimen lost). R. Cp, vv (OC2229). Scale bar = 220  $\mu$ m for A-C, F, G; 57  $\mu$ m for D, H; 100  $\mu$ m for E; 188  $\mu$ m for I, J; 217  $\mu$ m for K, Q, R; 34  $\mu$ m for L, M; 37 µm for N; 54 µm for O; 90 µm for P.



### **MEASUREMENTS**

L (LV) = 469-484  $\mu m$  (n = 4); H (LV) = 227-242  $\mu m$  (n = 2).

### **DIFFERENTIAL DIAGNOSIS**

The closest congener of Vestalenula flexuosa spec. nov. is doubtlessly V. molopoensis (ROSSETTI & MARTENS, 1997), described from dolomitic springs in the northern part of South Africa. In order to allow a good differential diagnosis, some new illustrations from the latter species (from Marico River) are given (Figs. 1I-R). The new species differs from all other species in the genus by its special shape in dorsal and ventral view, as its congeners have far less asymmetry between anterior and posterior sides in these views. The new species can further be distinguished from V. molopoensis by the carapace shape in lateral view, especially by the length and position of dorsal margin, which is longer and more sloping in V. molopoensis, by the form and position of the keel on the RV, which is more pronounced in the latter species and by the fact that V. molopoensis generally has a slightly different pattern and less scars in the central Ms (7-8).

### **REMARKS**

The present species is here formally described, in spite of the fact that no soft parts were available. We have decided to do this because a long series of good type specimens was available, showing that the distinguishing features are not due to individual variability and because the genus Vestalenula in general has very conservative soft parts, and species in this genus are generally identified on carapace and valve features. The only soft part features that could possibly be useful in determining the position of V. flexuosa spec.nov. in the genus are: presence or absence and shape of the P-Abd (a feature already difficult to see in the best of dissections), shape and size of caudal rami. Exceptionally, the chaetotaxy of other limbs offers diagnostic features: the A1 and A2 in the case of V. pagliolii, and the Md-palp in the case of V. inconspicua and V. cuneata. However, the latter are exceptions. We are thus confident that the present species can be identified on the characters presented here.

# Vestalenula marmonieri spec.nov. (Figs 2A-I, 3A-D)

### TYPE LOCALITY

A tributary of the River Diahot, close to the village of Ouegoa, north of the New Caledonia Island. River 3 m wide and 0.25 m deep. Sample taken interstitially; sediment mostly made of coarse gravel (mean diameter 2.6 cm) and little sand (14%). Water temperature 29 to 32 °C. Coordinates: S 27°20' E 164°25'. All material was collected on 03.01.1999 by P. MARMONIER (France).

### TYPE MATERIAL

About fifteen specimens were retrieved from the sample. Holotype: a dissected female, with soft parts in glycerine in a sealed slide and with valves stored dry, lodged in the Ostracod Collection of the Royal Belgian Institute of Natural Sciences (Brussels, Belgium), no. OC2219. Paratypes: 5 figured specimens, 2 dissected and 3 carapaces kept dry in micro-pal slides, all stored in the Ostracod Collection of the Royal Belgian Institute of

Natural Sciences (Brussels, Belgium), nos. OC2220-2224.

### DERIVATION OF NAME

We take pleasure in naming this species after Dr PIERRE MARMONIER (Université de Savoie, France) who kindly provided his darwinulid material collected in New Caledonia.

# ABBREVIATED DESCRIPTION

Carapace (Fig. 2C) and valves (Figs. 2A,B) relatively elongated in lateral view, with straight part of dorsal margin long, about half the length, and sloping towards the front, not parallel to the straight ventral margin. Antero-ventral tooth in LV (Fig. 2G) large; postero-ventral keel on RV (Fig. 2F) small, elongate (but not as in the danielopoli-group of this genus) and positioned well to the front. Ms (Figs. 2H,I) large, with 10 scars. Carapace in dorsal (Fig. 2D) and ventral (Fig. 2E) views with anterior part rather bluntly pointed, posterior part more widely rounded but less so than in the preceding species.

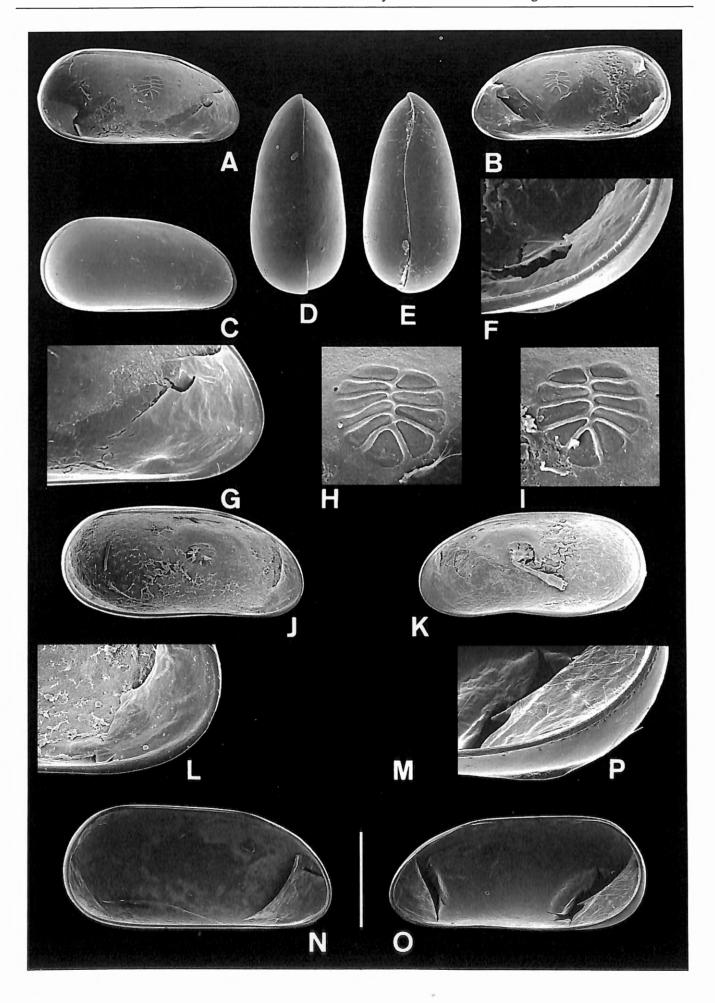
Fig. 2 - A-I: Vestalenula marmonieri spec.nov. (tributary of River Diahot, New Caledonia). J-M: Vestalenula spec. C (Israel). N-P: Vestalenula spec. D (Lake Sibaya, KwaZulu-Natal, R. South Africa).

V. marmonieri spec.nov.: A. LV, iv (OC2223). B. RV, iv (idem). C. Cp, right lv (OC2222). D. Cp, dv (OC2221). E. Cp, vv (OC2220). F. RV, iv, detail postero-ventral corner (OC2223). G. LV, iv, detail antero-ventral corner (idem). H. RV, iv, detail central Ms (idem). I. LV, iv, detail central Ms (idem).

V. spec. C: J. LV, iv (OC2225). K. RV, iv (idem). L. LV, iv, detail antero-ventral corner (idem). M. RV, iv, detail postero-ventral corner (idem).

V spec. D: N. LV, iv (OC2226). O. RV, iv (idem). P. RV, iv, detail postero-ventral corner (idem).

Scale bar = 240  $\mu$ m for A-E; 68  $\mu$ m for F; 96  $\mu$ m for G; 59  $\mu$ m for H, I; 180  $\mu$ m for J, K, N, O; 65  $\mu$ m for L; 73  $\mu$ m for M; 70  $\mu$ m for P.



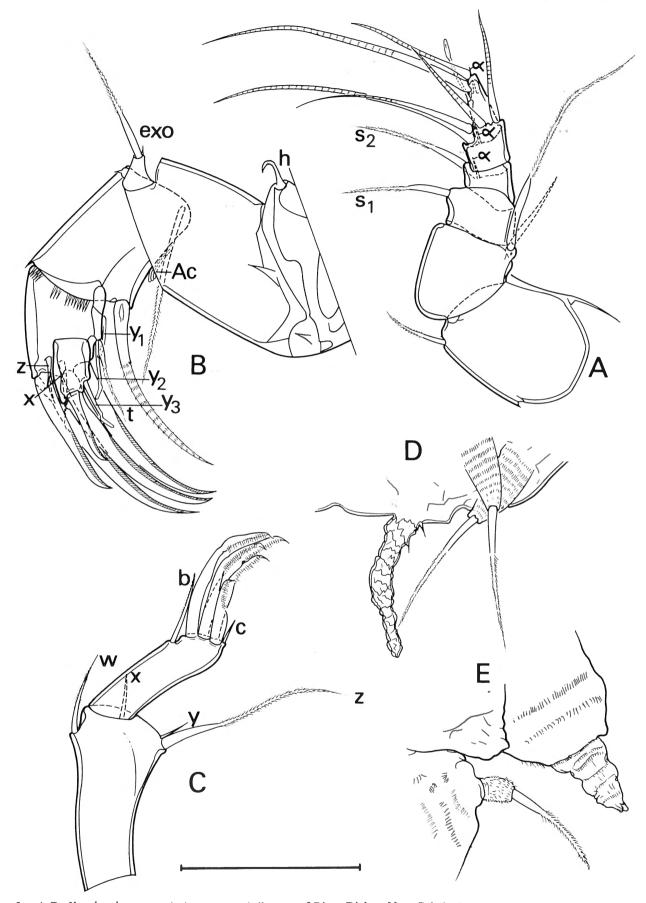


Fig. 3 – A-D: Vestalenula marmonieri spec. nov. (tributary of River Diahot, New Caledonia). E: Vestalenula molopoensis (MARTENS & ROSSETTI, 1997) (Molopo, North West Province, R. South Africa).

A. A1 (OC2224). B. A2 (idem). C. Md-palp (idem). D. Fu and P-abd (OC2223). E. Fu and P-abd (OC2100). Scale bar = 50 µm.

Appendages as typical of the genus. A1 (Fig. 3A) with first endopodal segment without ventro-apical seta. A2 (Fig. 3B) with first endopodal segment with 1 ventro-apical seta. Md-palp (Fig. 3C) with seta 'z' on penultimate segment longer than final segment. Caudal rami (Fig. 3D) large, with conical bases and long setae. P-Abd (Fig. 3D) elongate, with almost parallel sides, about 5x as long as its greatest width.

### **MEASUREMENTS**

L (LV) = 504-516  $\mu$ m (n = 4); H (LV) = 244-248  $\mu$ m (n = 2).

### **DIFFERENTIAL DIAGNOSIS**

Also this new species is closest to *V. molopoensis*, from which it can be distinguished primarily by the shape of the caudal rami and, especially, of the P-Abd (see illustration of these structures in *V. molopoensis* Fig. 3E), as well as in the shape of the valves in lateral view (Figs. 1I-K). Following the diagnostic key in ROSSETTI & MARTENS (1998), *V. marmonieri* spec.nov. can be distinguished from the *danielopoli*-group by the shape of the postero-ventral keel on the RV and the antero-ventral tooth in the LV, from *V. daps, V. inconspicua* and *V. cuneata* by the presence of a P-Abd, from *V. pagliolii* by the chaetotaxy of the A1 and A2, from *V. lundi* and *V. marlieri* by the size and the shape of the valves in lateral view and from *V. flexuosa* spec.nov. by the shape of the carapace in dorsal and ventral view.

# Vestalenula spec. C (Figs 2J-M)

### MATERIAL INVESTIGATED

One adult female collected in 1991 by Dr Y. SPIRA (Jerusalem) in a spring near the Dead Sea, Israel. Coordinates and date of collection unknown. The dissected specimen is stored in the Ostracod Collection of the Royal Belgian Institute of Natural Sciences (Brussels, Belgium), no. OC2225, with soft parts (mostly damaged) kept in glycerine in a sealed slide and valves kept dry in a micro-pal slide.

# ABBREVIATED DESCRIPTION

Carapace and valves (Fig. 2J,K) elongate, with dorsal margin straight over more than half the length, parallel to the sinuous ventral margin. Caudal margin evenly rounded. Antero-ventral tooth in LV (Fig. 2L) prominent, postero-ventral keel on RV (Fig. 2M) small, short and positioned well towards the front.

### **MEASUREMENTS**

L (LV) = 462 
$$\mu$$
m (n = 1); H (LV) = 216  $\mu$ m (n = 1).

### REMARKS

As only one specimen is available, and soft parts are in bad condition, no formal description of this species is given here. It is very difficult to assess the affinity of this specimen, as the shape of the Cp in dorsal view remains unknown. Its elongated shape in lateral view (L/H = 2.1) would put it near *V. boteai*, but the keel on the RV has a different shape (see illustration in MARTENS et al. 1997). The elongated aspect in any case suggests an interstitial mode of life, but new material is necessary before taxonomic position and ecology can be established with confidence.

# Vestalenula spec. D (Figs 2N-P, 4A-D)

### MATERIAL INVESTIGATED

One adult female collected by K. MARTENS, M. HAMER & M. COKE in Lake Sibaya, KwaZulu-Natal, R. South Africa (16.10.1994). Coordinates: S 27°25'13" E 32°41'53". The dissected specimen is stored in the Ostracod Collection of the Royal Belgian Institute of Natural Sciences (Brussels, Belgium), no. OC2226, with soft parts kept in glycerine in a sealed slide and valves kept dry in a micro-pal slide.

# ABBREVIATED DESCRIPTION

Cp and valves (Fig. 2N,O) elongated, dorsal and ventral margin straight and parallel to each other, straight part of dorsal margin >1/2 of total length. Antero-ventral tooth in LV rather small, postero-ventral keel in RV (Fig. 2P) elongate (although not as in the *danielopoli*-lineage) and positioned well to the front.

Appendages as typical of the genus. A1 (Fig. 4A) with first endopodal segment without ventro-apical seta. A2 (Fig. 4B) with first endopodal segment with 1 ventro-apical seta. Md-palp (Fig. 4C) with seta 'z' on penultimate segment longer than final segment. Caudal rami (Fig. 4D) large, with elongated conical bases. P-Abd (Fig. 4D) present, but with unclear morphology.

### **MEASUREMENTS**

$$L = 510 \mu m (n = 1); H = 237 \mu m (n = 1).$$

### **REMARKS**

As only one specimen is available, no formal description of this species is given here either. Both the shape (dorsal and ventral margin straight and parallel) and the elongated aspect puts this specimen close to *V. lundi* from Sri Lanka. Most soft parts of the present specimen agree to the

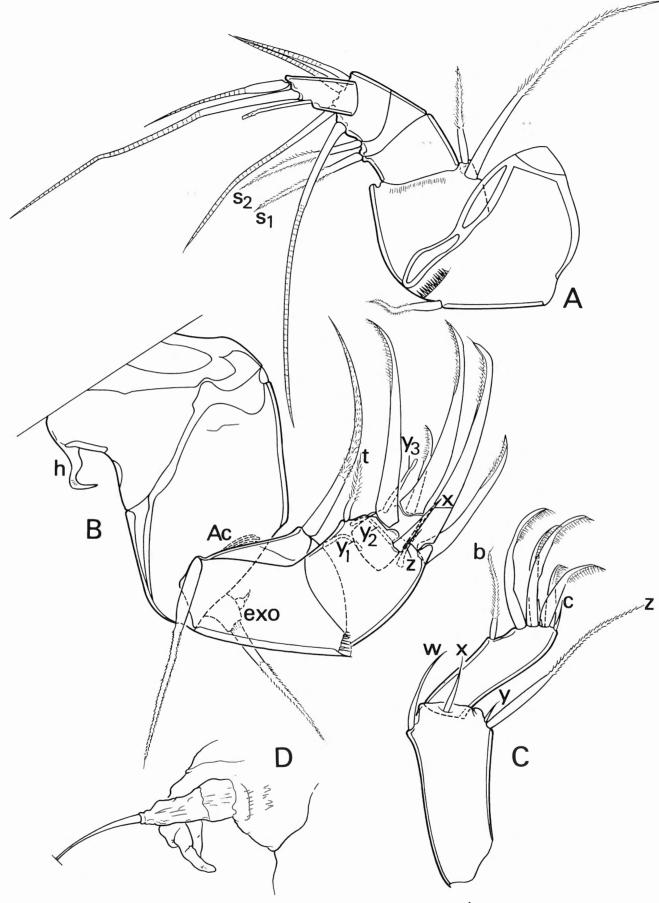


Fig. 4 – *Vestalenula* spec. D (Lake Sibaya, KwaZulu-Natal, R. South Africa). A. A1 (OC2226). B. A2 (idem). C. Md-palp (idem). D. Fu and P-abd (idem). Scale bar =  $50~\mu m$ .

morphology of V lundi, except for the shape of caudal rami and the P-Abd. However, the latter was difficult to observe in the South African specimen. The present specimen is also larger (L (LV) = 400  $\mu$ m in V lundi), but its presumed lacustrine mode of life would be consistent with this. New material from Lake Sibaya is necessary to establish the taxonomic position of this species, but a second visit to the same locality in 1995 did not yield further specimens. Either the species is very rare, has a patchy distribution or lives in a special microhabitat.

## Vestalenula marlieri (KISS, 1959)

### TYPE MATERIAL

As *Darwinula marlieri*, described from the Luhanga-Uvira stream, at the northern extremity of Lake Tanganyika, Zaire. Coll. 22.06.1958 by G. MARLIER. Material presently curated in the Royal Museum for Central Africa (Tervuren, Belgium).

No. 50330: dissected soft parts (re-illustrated by Rossetti & Martens, 1998, Fig. 10).

No. 50328: one Cp of an immature darwinulid (newly investigated material).

### **REMARKS**

As the newly investigated material of this species consisted of an immature specimen only, no additional information on valve morphology, and more in particular the presence or absence of a keel in the RV and internal teeth on the LV, is therefore available.

## Vestalenula daps (HARDING, 1962)

# TYPE MATERIAL

Holotype: a specimen in a mounted slide. Zoological Museum (Copenhagen, Denmark), no. CRU-2651 (re-illustrated by ROSSETTI & MARTENS, 1998, Fig. 7). Paratypes: a dissected specimen with soft parts in sealed slide and seven specimens stored in alcohol, the latter with decalcified valves. From the stomach of the fish *Eleotris fusca*, Lake Te-Nggano, Rennel Island, Solomon Islands. Coll. 23.10.1951 by T. Wolff. Zoological Museum (Copenhagen, Denmark).

Newly investigated types: several carapaces stored in alcohol. British Museum (London, UK), no. 1962.12.10.3.

# REMARKS

The newly investigated type specimens had decalcified carapaces, not suitable for SEM preparations. Nevertheless, investigations with light microscope confirmed the presence of a postero-ventral keel on the RV, as expected for the genus *Vestalenula*. This morphological character was neither reported in the original description of *Darwinula daps* by HARDING (1962) nor detectable in the holotype material.

### Discussion

The present paper provides new localities of the genus Vestalenula (New Caledonia, Israel, South Africa and Namibia), thus confirming the global distribution of this genus already shown by Rossetti & Martens (1998: Fig. 30). The total number of species presently recognized in this genus, including the four species left in open nomenclature (2 by DANIELOPOL 1980, 2 in the present paper), now amounts to 15, each with a restricted geographical distribution (Table 1). None of these species were thus far recorded outside the geographical area from which they were originally described. This is exceptional, as several species in other darwinulid genera (for example Darwinula stevensoni, Penthesilenula brasiliensis. Microdarwinula zimmeri) have intercontinental distributions. Therefore, whereas the global generic distribution indicates that Vestalenula might be an old group, the relatively high specific diversity (relative to other recent darwinulids) and the high degree of endemicity indicate that this genus might have experienced a relatively high degree of speciation. As we are dealing with ancient asexuals, the time scales used here are relevant and the global distribution of Vestalenula might at first glance even be correlated to a Pangean distribution of the ancestor, with a slow divergence of the different species in Post-Palaeozoic times. Given the impeded rate of

Table 1: Recent species presently recognised in the genus *Vestalenula* and their occurrence.

Germany

danielopoli-lineage

V. danielopoli (MARTENS et al. 1997)

V. spec. A (in DANIELOPOL 1980)	Cuba
V. spec. B (in DANIELOPOL 1980)	Tunisia
<i>boteai-</i> lineage	
Vestalenula boteai (DANIELOPOL, 1970)	Romania, France
Vestalenula cuneata (KLIE, 1939)	Kenya
Vestalenula daps (HARDING, 1962)	Solomon Islands
Vestalenula flexuosa spec.nov.	Namibia
Vestalenula inconspicua (KLIE, 1935)	Ivory Coast
Vestalenula lundi (NEALE & VICTOR, 1978)	Sri Lanka
Vestalenula marlieri (KISS, 1959)	D.R. Congo
Vestalenula marmonieri spec.nov.	New Caledonia
Vestalenula molopoensis	
(Martens & Rossetti, 1997)	R. South Africa
Vestalenula pagliolii	
(Pinto & Kotzian, 1961)	Brasil
Vestalenula spec. C (this paper)	Israel
Vestalenula spec. D (this paper)	R. South Africa

evolution in ancient asexuals (SCHÖN et al. 1998, MARTENS 1998), it cannot entirely be excluded that cladogenesis in this group indeed requires such time scales. However, as there are no biological or ecological indications to assume that dispersal mechanisms in Vestalenula are less effective than in other darwinulids, these speciation events most likely took place relatively recent, for example in the Tertiary or even later. Molecular analyses of these taxa, combined with a re-interpretation of the fossil record will provide more realistic and more rigid time frames in which the evolution of these ancient asexuals can be interpreted. Such analyses are underway.

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Koen MARTENS
Royal Belgian Institute of Natural Sciences
Freshwater Biology
Vautierstraat 29
B-1000 Brussels, Belgium
(to whom reprint requests should be sent)

Giampaolo ROSSETTI
Department of Environmental Sciences
University of Parma
Parco Area delle Scienze, 33A
I-43100 Parma, Italy