

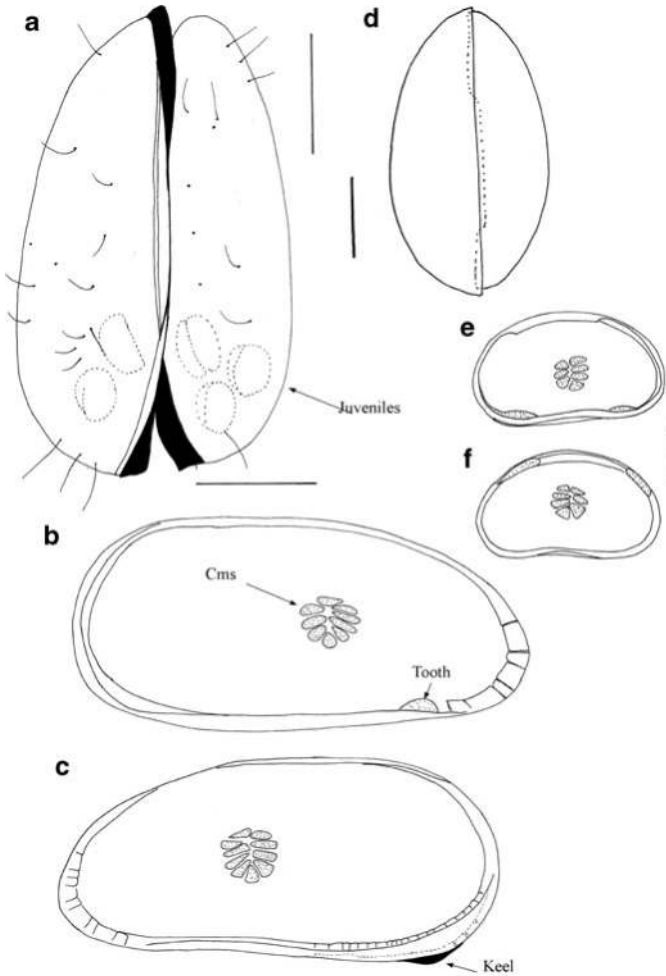
# **Superfamily Darwinuloidea Brady and Norman 1889 (Suborder Darwinolocopina Brady and Norman 1889)**

Diagnosis (after Horne et al. 2002, amended for male characters after Smith et al. 2006): Carapace mostly 0.8 mm long or less, shape elongate, subquadrate, or oval (Figs. 1 and 8a–c). Valves smooth, or rarely finely reticulated). CIL narrow, without selvage or continuous lists. Some groups with narrow ventro-caudal keel (Figs. 1c and 8b) or with internal ventral or caudal teeth in one valve (Fig. 1b). Hinge adont. CMS consisting of tight “rosette” of 5–10 scars (Figs. 1a, b and 8a), arranged radially; the rosette sometimes vertically elongated to give the appearance of a double vertical row. Normal pores simple. Females with brooding chamber posteriorly (Fig. 1a). Seven pair of appendages present. UR absent or reduced to 1–2 short setae (Fig. 7b). A2 sexually dimorphic, in male second segment of protopod subdivided, 2 setae (t2 and t3) transformed. Md vibratory plate small. Mx1 with large vibratory plate bearing many unreflexed and four reflexed rays (Fig. 5). L5 endopod with 3 segments, protopod with vibratory plate (Fig. 6a). In males protopod transformed into prehensile palps. L6, L7 walking legs (Figs. 6b, 8e, h and 7a). Hemipenis laterally compressed and with three lobes. Internally hemipenis with striations in distal third.

The superfamily has six families of which only the family Darwinulidae has living representatives.

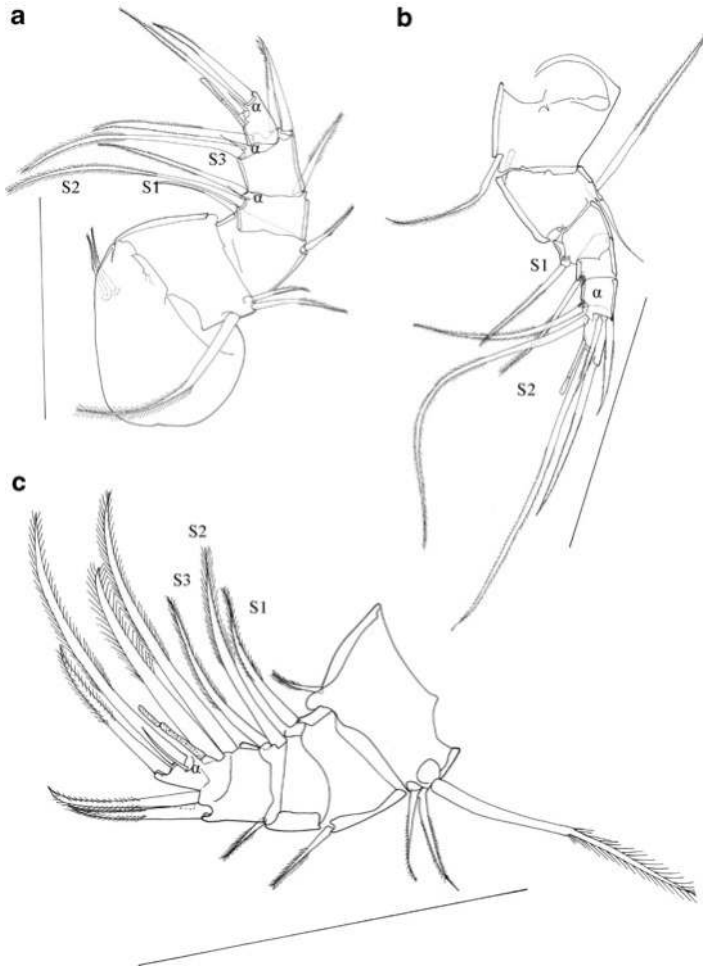
## **1 Family Darwinulidae Brady and Norman 1889 [Syn.: Microdarwinulidae Kashevarova and Neustrueva 1982]**

Diagnosis (after Rossetti and Martens 1998): Small- to medium-size ostracods, elongate, subsquarish, or rounded in lateral view. Hinge adont or with medial groove, anterior and posterior cardinal teeth on RV, cardinal ridge and sockets on LV, and valve margins without selvages or continuous inner lamella. Fused zones very narrow with few and very short MPC. Sometimes ventro-caudal keel on RV, or



**Fig. 1** Line drawings: (a–c) *Vestalenula* sp.; (d–f) *Microdarwinula zimneri* (Menzel 1916): (a) dorsal view of the carapace with brooding chamber and juveniles; (b) LV, inside view; (c) RV, inside view; (d) dorsal view; (e) LV, inside view; (f) RV, inside view. Scales = 0.1 mm. (d–f) modified after Danielopol (1968a, b)

with internal ventral or caudal teeth on LV. A2 without swimming setae (Figs. 3 and 8e, i). P5 with 3-segmented protopod, transformed into clasping organ in male. UR if present reduced to single seta (Fig. 7b). P-abd present or absent (Figs. 7c, 8j). Brooding chamber present. Hemipenis laterally compressed and with three lobes.

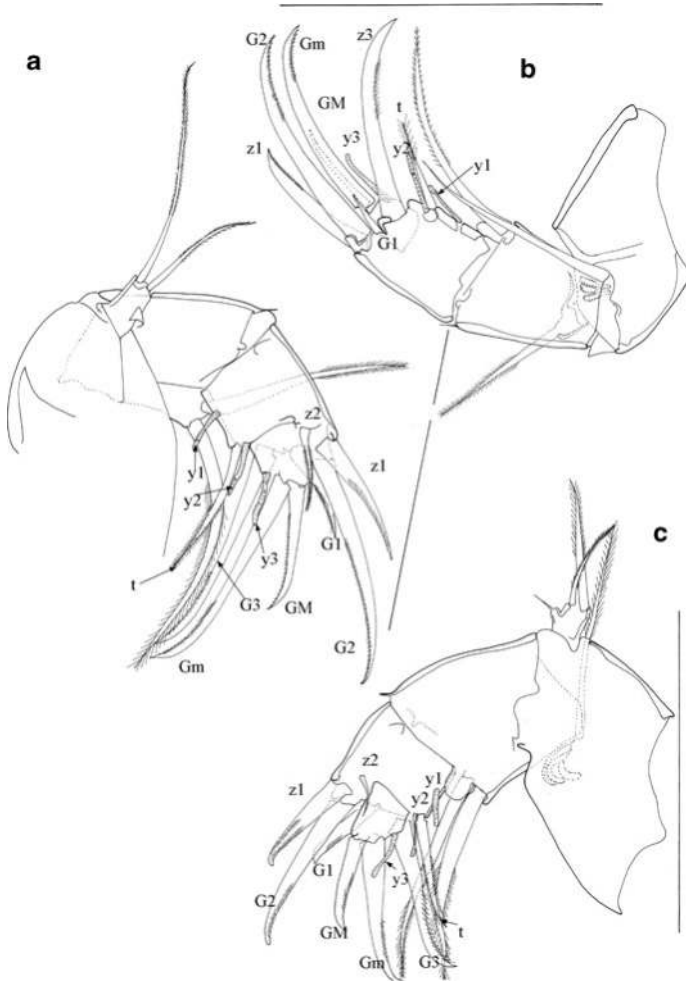


**Fig. 2** Line drawings: (a) *Darwinula stevensoni* (Brady and Robertson 1870); (b) *Vestalenula matildae* Martens and Rossetti, 2002; (c) *Penthesilenula brasiliensis* (Pinto and Kotzian 1961); A1. Scales = 0.1 mm

Internally, hemipenis with striations in distal third. Distally above hemipenis and connected to duct, ovate structure present.

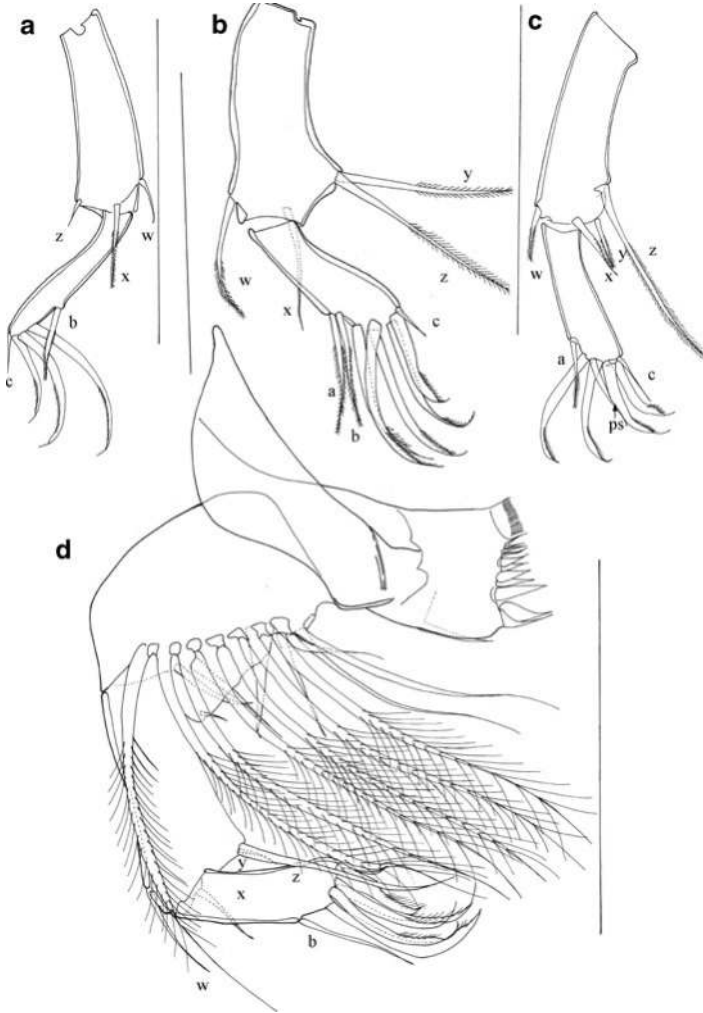
Type genus: *Darwinula* Brady and Robertson 1885

Other genera: *Alicenula* Rossetti and Martens 1998; *Microdarwinula* Danielopol 1968; *Penthesilenula* Rossetti and Martens 1998; *Vestalenula* Rossetti and Martens 1998.



**Fig. 3** Line drawings: (a) *Darwinula stevensoni* (Brady and Robertson 1870); (b) *Vestalenula matildae* Martens and Rossetti 2002; (c) *Penthesilenula brasiliensis* (Pinto and Kotzian 1961): A2. Scales = 0.1 mm

Remarks: There are approximately 30 living species. Of the nine non-marine ostracod lineages survived today Darwinulidae were probably the first having appeared in non-marine habitats over 350 million years ago (Smith and Kamiya 2008). Due to the lack of males in living populations and in fossil assemblages from the Mesozoic onward, it has been claimed that the group has been asexual for over 200 million years (Martens et al. 2003; Van Doninck et al. 2004; Martens et al. 2005). This has been questioned with the discovery of rare males of *Vestalenula cornelia* Smith et al. 2006. Since the males are much smaller than females, it has been postulated that they could be overlooked (perhaps mistaken for juveniles) in samples (Smith et al. 2006). The lack of the Zenker organ and the possession of a



**Fig. 4** Line drawings: (a) *Alicenula serricaudata* (Klie 1935); (b) *Penthesilenula brasiliensis* (Pinto and Kotzian 1961); (c) *Darwinula stevensoni* (Brady and Robertson 1870); (d) *Vestalenula matildae* Martens and Rossetti, 2002: Md. Scales = 0.1 mm

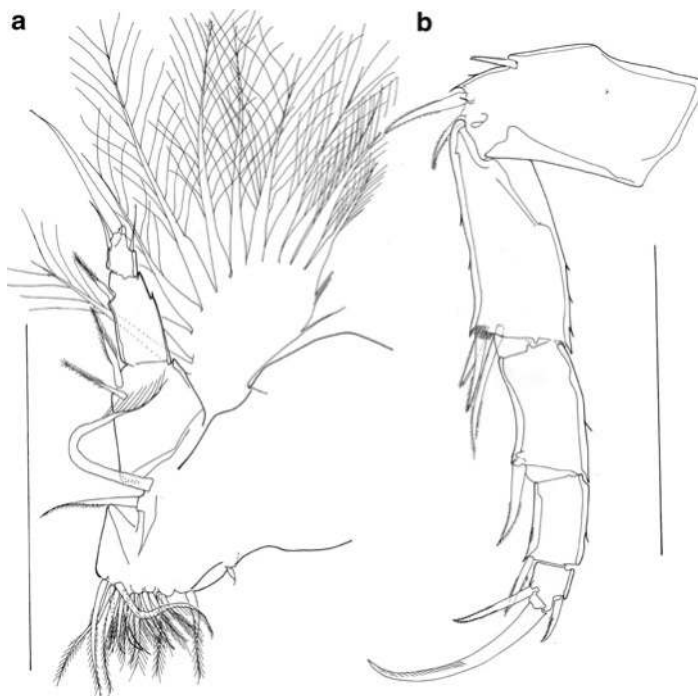
“rake” of setae (for filter feeding) on the mandibular palp, place Darwinulidae close to the Terrestricytheroidea (a superfamily of Cytherocopina), what has also been suggested using cladistic analysis (Horne et al. 2005). However, Cypridoidea and Darwinuloidea share two characters: presence of the vibratory plate on the L5 (which is absent in Terrestricytheroidea), and 2-segmented palp on the same appendage (4-, 5-segmented in Terrestricytheroidea). Nevertheless, the appearance of all other appendages, including the partly muscular hemipenis, places Darwinulidae indeed much closer to Cytheroidea than to Cypridoidea.

**Fig. 5** Line drawings:  
*Vestalenula matildae* Martens  
 and Rossetti 2002: Mxl.  
 Scale = 0.1 mm



**Key to the genera of Darwinulidae (after Rossetti and Martens 1998)**

1. Carapace rounded in lateral view, no externally visible brood pouch (Fig. 1d-f) . . . *Microdarwinula* Danielopol 1968
  - Carapace elongated or subsquarish in lateral view, with brood pouch recognizable by considerably inflated posterior end of carapace (Figs. 1a–c and 8a–c) . . . . . 2
2. RV without postero-ventral keel; LV with or without internal teeth. A1 with two dorsal setae on first segment (Fig. 2c); second segment with one dorso-apical seta and three ventral setae. A2 exopod with two setae and a spine (Fig. 3c) . 3
  - RV with postero-ventral keel (Fig. 1c); LV with antero-ventral internal tooth (Figs. 1b and 8b). A1 with one dorsal seta on first segment; second segment with two ventral setae and dorso-apical seta absent (Fig. 2b). A2 exopod with one seta and a spine (Fig. 3b) . . . . . *Vestalenula* Rossetti and Martens 1998
3. LV without internal teeth. Last segment of Md-palp with less than five claws (3 or 4); penultimate segment with seta y short or absent (Fig. 4a, c) . . . . . 4
  - LV with internal teeth. Last segment of Md-palp with five claws; penultimate segment with y seta long (Fig. 4b) . . . *Penthesilenula* Rossetti and Martens 1998
4. RV overlapping LV. Fourth segment of A1 with two large dorsal setae (Fig. 2a). Md-palp, penultimate segment with seta z long, seta y short; last segment with



**Fig. 6** Line drawings: *Vestalenula matildae* Martens and Rossetti 2002: (a) L5; (b) L6. Scales = 0.1 mm

“poil stevensoni”, setae *a* and *b* present (Fig. 4c)... *Darwinula* Brady and Robertson 1885

- RV overlapping LV, or LV overlapping RV. Md-palp penultimate segment with seta *z* short, seta *y* absent, last segment without “poil stevensoni”, seta *a* absent (Fig. 4a). . . . . *Alicenula* Rossetti and Martens 1998

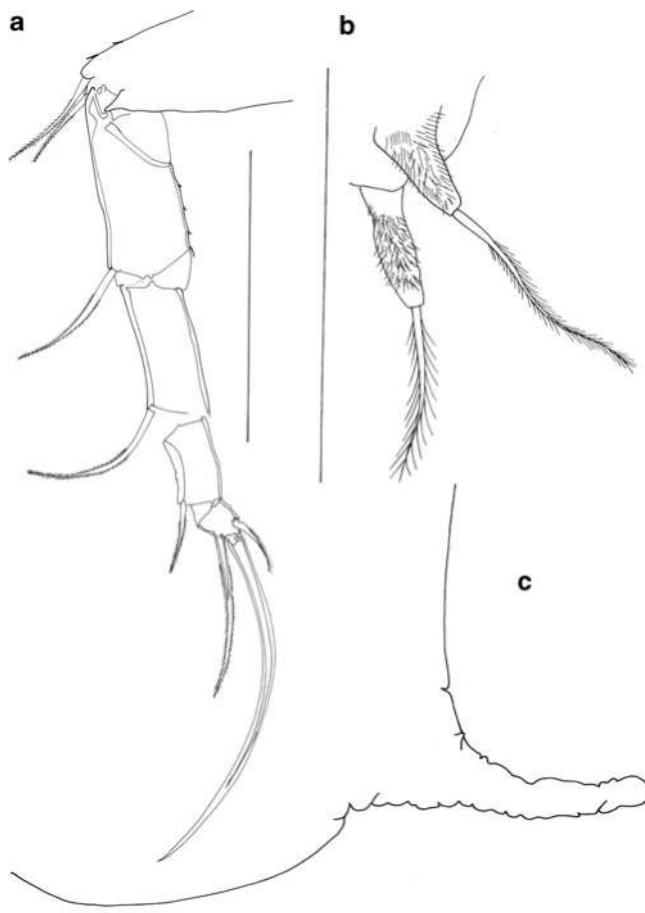
***Alicenula* Rossetti and Martens 1998.**

Diagnosis (after Rossetti and Martens 1998): LV overlapping RV. Elongate darwinulids, without internal teeth on the LV, and without ventral keel on the RV. Dorsal margin evenly sloping. Large brooding chamber present. First segment of A1 with two dorsal setae, second segment with one dorso-apical seta, and three ventral setae. A2 with two long setae and a spine on exopod. Seta *y* on penultimate segment of Md-palp absent. Adults with CR. P-abd mostly large and spinous.

Type species: *A. serricaudata* (Klie 1935a)

Species list with type locality and type material

1. *A. furcabdominis* (Keyser 1975). North River, Everglades National Park, Florida, USA, 80°54'20"W, 25°20'30"N: Holotype (♀), ZMH – K-30 328.



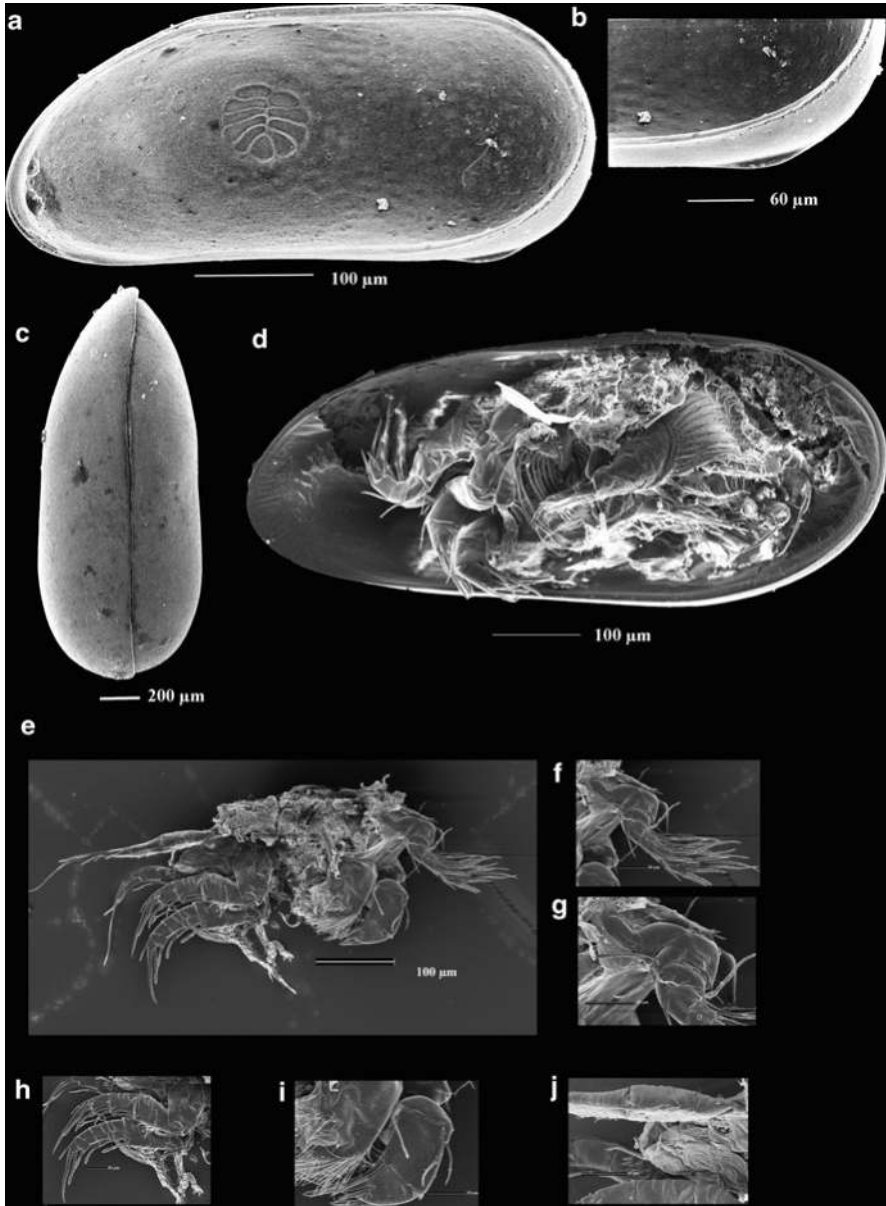
**Fig. 7** Line drawings: *Vestalenula matildae* Martens and Rossetti 2002: (a) L7; (b) UR; (c) P-abd. Scales = 0.1 mm

2. *A. inversa* (Martens and Rossetti 1997). Dolomitic spring at Marico Oog, Molopo Oog area, North West Province (formerly Transvaal), South Africa, 25°47'30''S 26°22'10''E: Holotype (♀), AM – TDW-7A.
3. *A. serricaudata* (Klie 1935a): Different localities in Ivory Coast (Tourni, Danané, Man), 09°34'N 07°31'W: Syntypes, ZMK – CR-292.

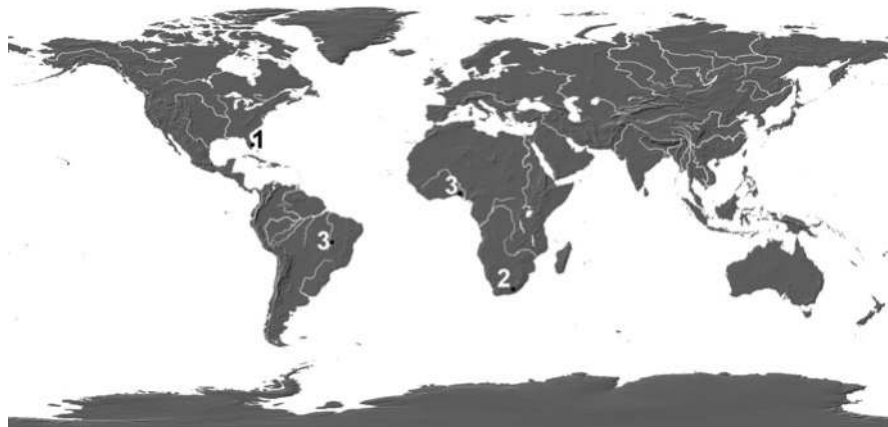
#### Key to the species

1. Last segment of Md-palp with three apical claws, seta *c* present. . . . . 2
- Last segment of Md-palp with four apical claws, and seta *c* absent ...  
*A. furcabdominis* (Keyser 1975)





**Fig. 8** SEM: (a–c) *Vestalenula paglioli* (Pinto and Kotzian 1961), (d) *Darwinula stevensoni* (Brady and Robertson 1870); (e–j) *Penthesilenula brasiliensis* (Pinto and Kotzian 1961): (a) RV, inside view; (b) inside view; (c) dorsal view; (d) body inside the valve; (e) whole body; (f) A1; (g) second segment of A1; (h) L5 and L6; (i) A2; J, P-abd. (a, c) Photos: D. Keyser



**Fig. 9** Distribution of the genus *Alicenula* Rossetti and Martens 1998: Numbers correspond to the species list

2. LV overlapping RV . . . . . *A. serricaudata* (Klie 1935a)  
 – RV overlapping LV . . . . . *A. inversa* (Martens and Rossetti 1997)

Ecology and distribution (Fig. 9)

Species can be found in rivers (*Alicenula serricaudata* and *A. furcabdominis*), mosses (*A. serricaudata*) and springs (*A. inversa*). Species have restricted distribution, and have been found so far only from their type localities with an exception of *A. serricaudata* (Klie 1935a) which has been recorded from the Paraná River in Brazil (Pinto and Kotzian 1961; Higuti et al. 2009a), and Ivory Coast (Klie 1935a).

***Darwinula* Brady and Robertson 1885.**

Diagnosis (after Rossetti and Martens 1998): RV overlapping LV on all sides except at the hinge. Brood chamber present. Shell elongate, greatest H in the posterior quarter, posterior margin more broadly rounded than the anterior one, ventral margin almost straight. First segment of A1 with two dorsal setae and one ventro-apical seta; second segment with one dorsal and three ventral setae; fourth segment with two large dorsal setae. Exopod A2 with unequal setae, one ½ as long as the other. Second segment of Md-palp with four terminal setae. Third segment with four apical claws, two subterminal, and two terminal setae. UR absent in adult specimens, P-abd conical, vermiform.

Type species: *D. stevensoni* Brady and Robertson 1885

Species list with synonyms, type locality, and type material

1. *D. stevensoni* (Brady and Robertson 1885). Somerton Board, Norfolk, England, UK, 52°36'41"N 00°54'50"E: Lectotype (♀ carapace), NEWHM:1.56.08.

Syn.: *Argilloecia aurea* Brady and Robertson 1870; *D. improvisa* Turner 1895; *Argilloecia aurea* Brady and Robertson 1870; *D. protracta* Rome 1953; *D. variabilis* Tagliasacchi-Masala 1968; *D. sphenoides* Rome 1977 (in Rome and De Deckker 1977)

#### Ecology and Distribution

The species can be found in all freshwater water bodies, including interstitial waters and slightly saline environments (up to maximum 15‰) (Meisch 2000). The species is cosmopolitan; it has been found on all continents, except Antarctica.

#### ***Microdarwinula* Danielopol 1968.**

Diagnosis (after Rossetti and Martens 1998): Small (<0.4 mm) animals, with rounded carapace, without externally visible brooding chamber. Carapace with LV/RV overlap. Hinge with prominent but smooth ridge on LV; RV with groove and prominent anterior and posterior cardinal teeth. RV without keel; LV with large antero- and postero-ventral internal teeth. A1 with two dorsal setae on the first segment; one dorsal seta and three ventral setae on the second segment. A2 with two long setae and a spine on exopod; first segment of endopod with two long ventro-apical setae. Setae *y* and *z* on penultimate segment of Md-palp long and subequal; terminal segment with six claws. UR and P-abd absent in adults and also in juveniles.

Type species: *M. zimmeri* (Menzel 1916)

#### Species list with type locality and type material

1. *M. inexpectata* Pinto et al. 2005a. Caetetus Ecological Station, Municipality of Gália, São Paulo State, Brazil, 22°22'42.9"S 49°41'17.2"W: Holotype (♀), MZUSP – 16591.
2. *M. zimmeri* (Menzel 1916). E Usambara, Amani, Tanzania, 04°44'59"S 38°29'59"E: Repository of type material unknown (see Rossetti and Martens 1998). Danielopol (1968a, b) deposited the material he examined of this species into the following institutions: ISER, NHMBU, BM, RIS, and NHMS.

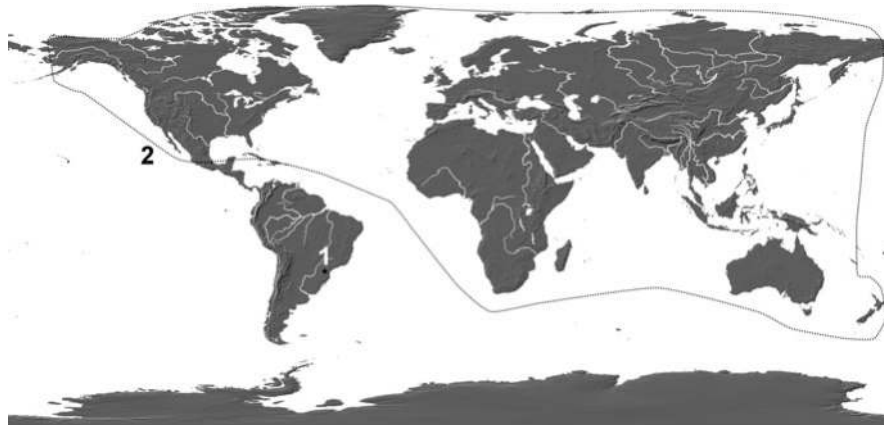
#### **Key to the species**

1. Shape more elongate in lateral view, postero-ventral tooth more ventrally positioned, *yl* aesthetasc absent on the penultimate segment of A2 . . . *M. inexpectata* Pinto et al. 2005a
- Shape more oblong in lateral view, postero-ventral tooth more caudally positioned, *yl* aesthetasc present on the penultimate segment of A2 . . . *M. zimmeri* (Menzel 1916)

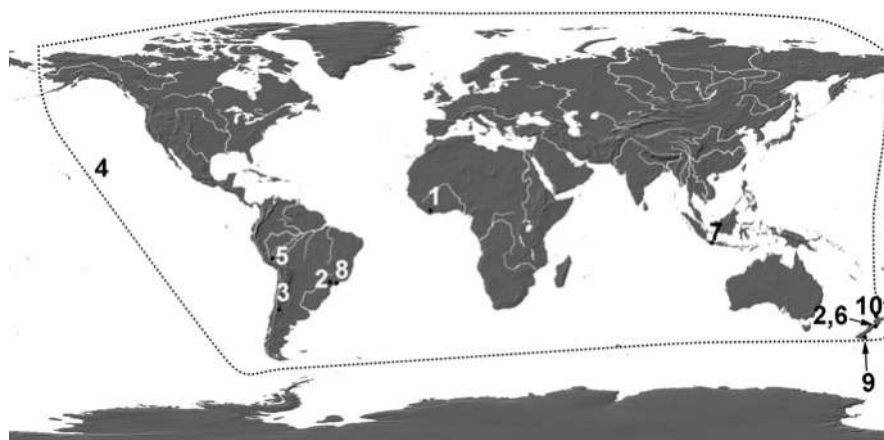
#### Ecology and distribution (Fig. 10)

Species can be found in interstitial waters and semiterrestrial habitats (Pinto et al. 2005a).

*Microdarwinula zimmeri* (Menzel 1916) has a wide distribution, and has been found so far on all continents, except South America and Antarctica (Pinto et al. 2005a). *Microdarwinula inexpectata* Pinto et al. 2005a has been found so far only in South America (Pinto et al. 2005a).



**Fig. 10** Distribution of the genus *Microdarwinula* Danielopol 1968: Numbers correspond to the species list



**Fig. 11** Distribution of the genus *Penthesislenula* Rossetti and Martens 1998: Numbers correspond to the species list

***Penthesislenula* Rossetti and Martens 1998.**

Diagnosis (after Rossetti and Martens 1998): Small (0.4 mm) to relatively large (0.8 mm) animal, subsquarish in lateral view, with dorsal margin straight over at least part of its L, anterior margin narrowly rounder, and posterior margin nearly straight. Hinge adont. Large caudal brooding chamber visible externally. In frontal view, LV and RV almost symmetrical, not oblique. LV generally overlapping RV on all sides except dorsally. LV either with rounded internal teeth (antero-ventral and postero-ventral in *africana*-group) or with one pointed caudal internal tooth (*incae*-group). RV without postero-ventral keel. First segment of A1 with two dorsal setae; second segment with three ventral and one dorsal seta. Exopod of

A2 with two long setae and a spine; first segment of endopod with two ventro-apical setae. Setae y and z on penultimate segment of Md-palp both long, subequal.

Type Species: *P. incae* (Delachaux 1928)

Species list with synonyms, type locality, and type material

1. *P. africana* (Klie 1935a). Moss spring at 21 km on road from Man to Touba, Ivory Coast, 07°24'19"N 07°32'35"W: Holotype (♀), ZMK – CR 291.
2. *P. aotearoa* (Rossetti et al. 1998). Small swamp overflowing into Karori Stream, Kinnoull Station, Wellington, New Zealand, 41°19'S 164°41'E: Holotype (♀), RBINS – O.C. 2143.
3. *P. araucana* (Löffler 1961a). Region of Villarica and Puyehue, Central Chile, 40°43'03"S 72°12'13"W: Repository of the type material unknown.
4. *P. brasiliensis* (Pinto and Kotzian 1961). Riverine pools at km 119.5 on road from Pôrto Alegre to Tramandai, Rio Grande do Sul, Brazil, 30°05'41"S 50°10'28"W: Holotype (♀), UFRGS – MP-0-11.
5. *P. incae* (Delachaux 1928). Lake Huaron, Region of Huancavelica, Department of Jurin, Peru, 13°23'S 72°15'W: Repository of the type material unknown. Rossetti et al. (1996) have examined a couple of specimens from the type locality and deposited the material in RBINS – O.C. 1791–1796.
6. *P. kohanga* (Rossetti et al. 1998). Small swamp overflowing into Karori Stream, Kinnoull Station, Wellington, New Zealand, 41°19'S 174°41'E: Holotype (♀), RBINS – O.C. 2151.
7. *P. malayica* (Menzel 1923). Moss springs along the road from Cibodas to Gede at 2,100 m a.s.l., West Java, Indonesia, 07°05'48"S 107°30'51"E: Repository of the type material unknown.
8. *P. reidae* Pinto et al. 2004. Boracéia Biological Station, Municipality of Salesópolis, São Paulo State, Brazil, 23°40'00.7"S 45°54'08.9"W: Holotype (♀), MZUSP – 15851.
9. *P. reboa* (Chapman 1963). Ross Creek, Dunedin, New Zealand: Holotype (♀), OMD – collection number unknown.
10. *P. sphagna* (Barclay 1968). *Sphagnum* swamp, Ardmore, Auckland district, New Zealand, 37°01'58"S 174°58'35"E.: Holotype (♀), MNZ – collection number unknown.

**Key to the species (after Rossetti and Martens 1998)**

1. LV with internal tooth along posterior margin, no postero-ventral tooth (*incae*-group) . . . . . 2
  - LV with postero-ventral internal tooth, no caudal tooth (*africana*-group) . . . . 6
2. LV overlapping RV . . . . . *P. araucana* (Löffler 1961a)
  - RV overlapping LV . . . . . 3
3. Size >0.7 mm . . . . . 4
  - Size <0.7 mm . . . . . 5

4. L/H ratio <2.2, P-abd absent . . . . . *P. incae* (Delachaux 1928)
  - L/H ratio >2.2, P-abd present . . . . . *P. kohanga* (Rossetti et al. 1998)
5. In lateral view, dorsal margin distinctly slanting forward, anterior end slender . . .
  - P. aotearoa* (Rossetti et al. 1998)
  - In lateral view, dorsal margin slightly curved, anterior end rounded . . . *P. sphagna* (Barclay 1968)
6. L/H ration 2.15–2.29, valves being more elongated . . . *P. reidae* Pinto et al. 2004
  - L/H ration 1.94–2.13, valves being less elongated . . . . . 7
7. Fourth segment of A1 with one large dorsal seta . . . *P. malayca* (Menzel 1923)
  - Fourth segment of A1 with two large dorsal setae . . . . . 8
8. Fourth segment of A1 with ventro-apical seta . . . *P. brasiliensis* (Pinto and Kotzian 1961)
  - Fourth segment of A1 without ventro-apical seta . . . . *P. africana* (Klie 1935a)

#### Remarks

The genus was divided into two species groups: *incae*- and *africana*-group. Some additional differential characters have been pointed out by Pinto et al. (2004), which have been observed on *Penthesilenula brasiliensis* (Pinto and Kotzian 1961), *P. reidae* Pinto et al. (2004), and *P. aotearoa* (Rossetti et al. 1998). The asymmetry between two setae on the exopod of A2 is more pronounced in the *incae*-group, and, the aesthetasc *yl* on A2 is pointed (seta-like) in *africana*-group, and has a rounded tip in *incae*-group. Most of the species in the genus have a very stable morphology of the soft parts. There is only a variability in the L of the carapace, while the shape (especially the L/H ratio) remains stable in all populations of all the species, even two populations of *P. aotearoa*, one from New Zealand (Rossetti et al. 1998) and the other from South America (Pinto et al. 2004). The only exception is *P. brasiliensis*, which shows a great variability in the wide area of its distribution (Fig. 11), and it is possible that it is a junior synonym of *P. africana* (Klie 1935a).

#### Species not included in the key

*Penthesilenula repoa* (Chapman 1963) described from New Zealand is not included in the key to the species of the genus *Penthesilenula* Rossetti and Martens 1998 because of the insufficient description. Rossetti and Martens (1998) even questioned the position of the species in the genus. *Penthesilenula setosa* (Daday 1902) was described from Argentina, but was considered as a junior synonym of *P. incae* (Delachaux 1928) by Rossetti and Martens (1998).

#### Ecology and distribution (Fig. 11)

The species of this genus have been collected from springs, rivers, and from both saturated and nonsaturated terrestrial habitats (Pinto et al. 2004). Most of the species have a restricted distribution, with the exception of *P. brasiliensis* (Pinto and Kotzian 1961), which has been collected from all continents, excluding Antarctica.

Pinto et al. (2004) reported *P. aotearoa* (Rossetti et al. 1998) from South America; the species was previously known only from New Zealand (Rossetti et al. 1998).

***Vestalenula* Rossetti and Martens 1998.**

Diagnosis (after Rossetti and Martens 1998): U Valves elongated or short and subquadrate. RV with postero-ventral external keel, which is short or elongated. LV with long or short internal antero-ventral tooth. Valves with L/R overlap. Dorsal margin straight over part of its L. Brooding chamber visible. First segment of A1 with one dorsal seta; second segment with two ventral setae; fourth segment without ventro-apical seta. Exopod of A2 with one long seta and a spine on exopod. Seta y on penultimate segment of Md-palp short; terminal segment with five apical claws. Adults mostly with UR, P-abd smooth or absent.

Type species: *V. boteai* (Danielopol 1970)

Species list with type locality and type material

1. *V. boteai* (Danielopol 1970). Interstitial of the river Mraconia, 2 km from confluence with river Danube, at 25 km of Orsova, Department Caras-Severin, Romania, 44°42'N 22°20'E: Holotype was not designated; some material is deposited in ISER, NHMBU, BM, and RBINS.
2. *V. botocuda* Pinto et al. 2003. Sítio da Colônia, Mulungu, near Araçoiaba da Serra, São Paulo State, Brazil, 23°32'02"S 47°39'41"W: Holotype (♀), MZUSP – 15041.
3. *V. carveli* Artheau 2007. Hyporheic zone of Font Calda spring, Reynes River, surroundings of village Reynès, Languedoc-Roussillon, France, 42°28'N 02°42'E: Holotype (♀), NHMP – MNHN-Os972.
4. *V. cornelia* Smith et al. 2006. Coastal area at Yudomari, S Yakushima Island, Kagoshima Prefecture, Japan, 30°13'48.7"N 130°28'50.5"E: Holotype (♀), LBM – 1430000876.
5. *V. cuneata* (Klie 1939a). Moss spring at the foot of Kikuyu, near Maji ya Moto, Kenya, 01°15'S 36°40'E: Holotype (♀), ZMK – CR-294.
6. *V. cylindrica* (Straub 1952). Type locality is "Miocene deposits" in Germany. Recent material has been collected from Lake Biwa (Japan) and deposited at LBM – 1430003528.
7. *V. danielopoli* (Martens et al. 1997). In calcareous silt sediment (Holocene, Subatlanticum) at Mùcheln, Geiseltal, 45 km W of Leipzig, Saxony-Anhalt, Germany, 51°18'11"N 11°50'25"E: Holotype, RBINS – O.C. 2085.
8. *V. daps* (Harding 1962). Stomach of the fish *Eleotis fusca*, Lake Te-Nggano, Rennel Island, Solomon Islands, 11°42'S 160°25'E: Holotype (♀), ZMC – CRU-2651.
9. *V. inconspicua* (Klie 1935a). Rocky banks of Bandama River, Ivory Coast, 06°13'N 02°44'E: Holotype (♀), ZMK – CR-293.
10. *V. irajai* Pinto et al. 2003. Tamoios Road, near Paraibuna, São Paulo State, Brazil, 23°31'9.6"S 45°32'21"W: Holotype (♀), MZUSP – 15049.

11. *V. lundi* (Neale and Victor 1978). Sandy river bank in Sabaragamuwa Province, Sri Lanka, 06°49'N 80°22'E: Holotype (♀), BM – 1977.76.
12. *V. marlieri* (Kiss 1959a). Luhanga stream, N part of Lake Tanganyika, Zaire, 03°30'S 29°15'E: Holotype (♀), RMCA – R.G. 50330.
13. *V. marmonieri* Rossetti and Martens 1999. A tributary of Diahot River, close to the village of Ouegoa, Province North, New Caledonia, 27°20'S 164°25'E: Holotype (♀), RBINS – O.C. 2219.
14. *V. matildae* Martens and Rossetti 2002. Government well 027, Ashburton River valley, Pilbara region, WA, Australia, 22°29'S 115°23'E: Holotype (♀), WAM – C28868-GR401.
15. *V. molopoensis* (Martens and Rossetti 1997). Molopo Oog area, North West Province (formerly Transvaal), South Africa, 25°46'40"S, 26°26'00"E: Holotype (♀), AM – TDW-26A.
16. *V. paglioli* (Pinto and Kotzian 1961). Riverine pools at 119.5 km on road from Pôrto Alegre to Tramandai, Rio Grande do Sul, Brazil, 30°01'S 51°13'W: Holotype (♀), UFRGS – MP-0-8.

**Key to the species (modified after Rossetti and Martens 1998)**

1. External keel on the RV elongate ... *V. lundi* Neale and Victor 1978; *V. danielopoli* Martens et al. 1997; *V. matildae* Martens and Rossetti 2002; *V. cornelia* Smith et al. 2006
  - External keel on the RV short ..... 2
2. P-abd present ..... 3
  - P-abd absent ..... 9
3. P-abd with seta ..... *V. botocuda* Pinto et al. 2003
  - P-abd without seta ..... 4
4. Third segment of A1 with one ventro-apical seta ... *V. paglioli* (Pinto and Kotzian 1961); *V. irijai* Pinto et al. 2003
  - Third segment of A1 without any ventro-apical setae ..... 5
5. L/H ratio >2.2. .... 6
  - L/H ratio <2.2. .... 7
6. Dorsal margin in lateral view rounded ..... *V. marlieri* (Kiss 1959a)
  - Dorsal margin in lateral view straight ..... *V. cylindrica* (Straub 1952)
7. Basis of UR carrying seta, very elongated ( $L = 3 \times W$ ) ... *V. boteai* (Danielopol 1970)
  - Basis of UR carrying seta, short ( $L = 1.5 \times W$ ). .... 8
8. P-abd very long and almost cylindrical ... *V. marmonieri* Rossetti and Martens 1999



- P-abd conical and not elongated . . . *V. malopensis* (Martens and Rossetti 1997)
- 9. Size > 0.6 mm . . . . . *V. daps* (Harding 1962)
- Size <0.6 mm . . . . . 10
- 10. Penultimate segment of Md-palp with seta *z* short; terminal segment with seta *c* . . . . . *V. inconspicua* (Klie 1935a)
- Penultimate segment of Md-palp with seta *z* long; terminal segment with seta *c*. . . . . *V. cuneata* (Klie 1939a)

Remarks

When the genus was described, it was divided into two species groups: *boteai*-group and *danielopoli*-group (Rossetti et al. 1998). The groups were distinguished based on the carapace characters: the keel on the RV is short in *boteai*-group, and elongate in *danielopoli*-group. The only representative of the latter group was a subfossil species, *V. danielopoli* Martens et al. 1997, described from German Holocene deposits. Later on, Martens and Rossetti (2002) described *V. matildae* Martens and Rossetti 2002 from Australia, with the same properties of the carapace as *V. danielopoli*. As the Australian species was described after the soft parts as well, it was thought that the presence of seta on the P-abd is also a distinguishing character of the *danielopoli*-group. However, description of *V. botocuda* Pinto et al. 2003, from the semiterrestrial habitats in Brazil (Pinto et al. 2003), has changed the previous taxonomy of the genus and its division into two groups. The Brazilian species has a carapace of the *boteai*-group and the soft parts (the presence of the seta on the P-abd) of the *danielopoli*-group. Most recent papers (Smith et al. 2006; Smith and Janz 2009) still retain the division of the genus into two species group, acknowledging only the carapace character as a distinguishing feature between them. The species belonging to the *danielopoli*-group could not be distinguished in the key as they differ in carapace characters (L, position of the keel, shape of the margins) which are not well defined by their respectful authors. When comparing their species the authors have described these differences as “slightly more”/ “slightly less” or just “more” or “less”; therefore they are very hard to use in the key. When distinguishing between the species, one should be extra cautious, and rely on the original drawings and descriptions (Neale and Victor 1978; Martens et al. 1997; Martens and Rossetti 2002; Smith et al. 2006; and Smith and Janz 2009). In the key to the species of the genus *Vestalenula* Rossetti and Martens 1998, given by Rossetti and Martens (1998), *V. lundi* (Neale and Victor 1978) has been placed in the *boteai*-group, as the authors only had decalcified carapaces and were unable to evaluate the appearance of the external keel on the RV. However, Smith and Janz (2009) have examined material they identified as *V. lundi* from Lake Biwa and have shown that the species actually belongs to the *danielopoli*-group. In the *boteai*-group, it is difficult to distinguish between the following species: *V. paglioli* (Pinto and Kotzian 1961) and *P. irajai* Pinto et al. 2003 both described from Brazil (Pinto and Kotzian 1961; Pinto et al. 2003). According to Pinto et al. (2003) *V. irijai* is more elongated with nearly straight dorsal and ventral margins, which



**Fig. 12** Distribution of the genus *Vestalenula* Rossetti and Martens 1982: Numbers correspond to the species list

furthermore run parallel to each other. Unfortunately, this difference is not clearly evident in the original illustrations of Pinto and Kotzian (1961, p. 47, Figs. 1 and 4) nor in the SEM of this species provided by Rossetti and Martens (1998, p. 100, Fig. 28a, b).

#### Ecology and distribution (Fig. 12)

*Vestalenula matildae* has been found in subterranean waters only (Martens and Rossetti 2002). Some Brazilian species have been described from semiterrestrial habitats (Pinto et al. 2003), while most of the species live in springs, swamps, and lakes (Martens and Rossetti 1997, 1998, 1999; Smith et al. 2006; Smith and Janz 2009). The species have pretty much restricted distribution, being found only in their type locality or the area surrounding it. With an exception of *V. boteai* (Danielopol 1970), *V. cornelia* Smith et al. 2006, *V. cylindrica* (Straub 1952), and *V. danielopoli* (Martens et al. 1997), which are known from the northern hemisphere, species of this genus are distributed in the southern hemisphere. The localities of the species *V. cylindrica*, presented on the map, are records of living representatives of this species (Smith and Janz 2009), while the species was described from Miocene deposits in Germany.