New species of *Temnocephala* Blanchard (Platyhelminthes, Temnocephalida) ectosymbiont on *Aegla serrana* Buckup & Rossi (Crustacea, Anomura) from southern Brazil [1]

José Felipe Ribeiro Amato, Suzana B. Amato & Luiz Carlos Campos Daudt

[1] Contribution number 403 of the Departamento de Zoologia, Universidade Federal do Rio Grande do Sul.

[2] Departamento de Zoologia, Instituto de Biociências, Universidade Federal do Rio Grande do Sul. Porto Alegre, Caixa Postal 15014, 91501-970 Porto Alegre, Rio Grande do Sul, Brasil. E-mail: jfamato@terra.com.br

ABSTRACT. A new species of the genus *Temnocephala* Blanchard, 1849 is described from southern Brazil, ectosymbiont on *Aegla serrana* Buckup & Rossi, 1977, an anomuran crustacean, collected in a creek and a reservoir of the highlands in the State of Rio Grande do Sul. All crustaceans examined were positive for this species of *Temnocephala* and carried eggs in different regions of the ventral side: perioral area, pleural strips, esternal plates, pereiopods and chelipods; to a lesser extent in the dorsal side of the cephalothorax and dorsal side of the uropods; as well as adult and young specimens. The most distinctive characters of the new species are: 1) cyanophilous glands forming an irregular-shaped, grape-like, bunch of approximately 10-15 cells, deeply staining with hematoxylin; 2) shape and size of the cirrus and its introvert section; 3) number, size and distribution of the rhabdite glands and 4) shape and position of the post tentacular, 'excretory' syncytial plates, with the off-centered nephridiopore.

KEY WORDS. Ectocommensals, Neotropical, Rio Grande do Sul, South America, taxonomy.

Fourteen of the 19 recorded species of *Temnocephala* Blanchard, 1849 for the Neotropical region were studied by Damborenea & Cannon (2001). Eight species were originally described from Brazil: *T. brevicornis* Monticelli, 1889 from freshwater turtles; *T. iheringi* Haswell, 1893 from ampullariid mollusks, *T. axenos* Monticelli, 1899 from anomuran crustaceans, *T. microdactyla* Monticelli, 1903, *T. lutzi* Monticelli, 1913, *T. travassosfilhoi* Pereira & Cuocolo, 1941 and *T. lanei* Pereira & Cuocolo, 1941 from trichodactylid crustaceans and *T. kingsleyae* Damborenea, 1994 from a pseudothelphusid crustacean.

The host of *T. axenos*, *Aegla laevis* (Latreille, 1818), must have been misidentified by the original collectors, as it does not occur in Blumenau, State of Santa Catarina, Brazil – the type locality (G. Bond-Buckup, pers. comm.). Presently, the species of *Aegla* known to occur in Blumenau are *A. jarai* Bond-Buckup & Buckup, 1994 and *A. odebrechtii* Müller, 1876. Additionally, the municipality of Blumenau has been divided, over the years, into several new municipalities, what renders it impossible to obtain precise information about the correct type locality determination for the species. Dioni (1967) identified as *T. axenos,* an ectosymbiont of an undetermined species of *Aegla* collected from three different geographical areas in the State of Rio Grande do Sul: Dom Pedro Creek in São Francisco de Paula, Porto Alegre and Jarau, near Quaraí and has proposed *T. bresslaui* González, 1949, found on *Aegla castro* Schmitt, 1942 from Curitiba, State of Paraná, as a junior synonym of *T. axenos*, because both species were morphologically similar. The existing illustrations of *T. axenos*, including the various drawings of the cirrus and the collected specimens from different hosts and localities in Rio Grande do Sul have shown that different morphologies might be compatible with different, undescribed species. A complete study of *T. axenos* with the application of the various techniques is under way.

The State of Rio Grande do Sul has a rich fauna of aeglids (21 species described to the present) (Buckup & Bond-Buckup 1999). *Aegla serrana* Buckup & Rossi, 1977, one of them, so far known only from this state, has not had any of its ectosymbiont community members (protists or helminths) recorded and/or described.

The present report describes a new species of *Temnocephala* from *A. serrana*, as well as documents, photographically (Figs 1 and 2), for the first time the: 1) areas of egg deposition on the host, shape of the eggs, morphology of the young, body pigmentation and the red, fugacious eye pigmentation in live specimens; 2) cirrus structure as revealed by de Faure's (deF) mounting medium preparations, Nomarski´s interference and phase contrast microscopy and histology; 3) paired, dorso-lateral, post tentacular 'excretory' syncytial plates as revealed by silver nitrate (SN) staining and scanning electron microscopy (SEM).

MATERIAL AND METHODS

Live temnocephalans were obtained from samples of *A. serrana* collected from a creek and a small reservoir in the Utopia II Farm, 5 km East of State Road RS-020, locality of Tainhas, Municipality of Cambará do Sul, State of Rio Grande do Sul (29º15'10"S, 50º15'45"W), southern Brazil. Some hosts were donated by Drs G. Bond-Buckup and L. Buckup, Laboratório de Crustáceos, Universidade Federal do Rio Grande do Sul (UFRGS), Porto Alegre, Rio Grande do Sul, already in ethanol 70% (amount of water calculated by percentage), while others was collected with dip nets and transported live to the Laboratório de Helmintologia; some helminths from live hosts were fixed in cold A.F.A. (70º GL ethanol – 93 parts; formalin 37% – five parts, glacial acetic acid – two parts), under slight cover slip pressure, stained in Delafield hematoxylin (HUMASON 1972), cleared in cedar oil and mounted in Canada balsam.

The description of the new species follows the Delta System format (DALLWITZ & PAINE 1986). The morphology of the post tentacular, 'excretory' syncytial plates was studied by fixing live specimens with hot (60ºC) silver nitrate (SN) (JOFFE *et al.* 1995, ROMEIS 1968). For SEM, specimens were flooded with hot (90ºC) formalin (HF), washed several times with distilled water, dehydrated in a graded ethanol/acetone series and critical point dried; coated with gold and examined with a Jeol, JSM-5800, scanning electron microscope. Cirrus morphology was studied after micro dissection and mounting each cirrus in deF. Groups of eggs removed by scrapping the crustaceans' exoskeleton were dehydrated, cleared in cedar oil and mounted in Canada balsam.

For histology, the specimens previously killed with ethanol 70% by the carcinologists were fixed in A.F.A., pre-stained in Delafield hematoxylin, sectioned with 6 µm, stained with Harris hematoxylin (HUMASON 1972) and eosin, and mounted in Canada balsam. Photomicrographs were taken with a Zeiss Axiolab microscope using phase contrast (or just the phase contrast condenser) or with a Leica DMR Hc microscope and Nomarski's interference contrast filters. The photographic images and line drawings were scanned and prepared using Adobe's Photoshop®. Measurements are in micrometers (µm) unless otherwise indicated; ranges are followed (between parentheses) by the mean, the number of specimens measured for a given character (when different than 10) and the standard deviation values. Measurements of cirrus length were made along the outside of the shaft wall but inside the introvert swelling; measurements of total cirrus length were made along its longest side and include the introvert; measurements of width of the introvert base include the thickness of the shaft walls but exclude the introvert swelling. The terminology to describe the reproductive structures follows CANNON (1993), CANNON & SEWELL (1995) and SEWELL & CANNON (1998). Drawings were made with a drawing tube on a Leitz Dialux 20-EB microscope.

The holotype and some paratypes, as well as slides containing individual cirri mounted in deF mounting medium were deposited in the Coleção Helmintológica do Instituto Oswaldo Cruz (CHIOC), Rio de Janeiro, Rio de Janeiro, Brazil.

RESULTS

Temnocephala cyanoglandula sp. nov.

Figs 3-30

Description (based on: 1573 specimens collected; 13 whole-mounted specimens – 12 adults and 1 juvenile; 7 cirri mounted in deF; 1 whole-mounted specimen fixed in silver nitrate, 2 specimens mounted on stubs for SEM, 1 specimen included in paraffin for sectioning; 10 specimens measured).

External characteristics

Body length, without tentacles, 1.85-3.15mm (2.61mm, 444); 1.73-2.32 mm (2.18 mm, 174); oval, wider at level of pharynx and narrower at level of posterior testes and anterior border of adhesive disk (Figs 5 and 10); body pigmentation present (Fig. 7), showing a well defined pattern appearing as faint orange in live specimens; adhesive disk ventral, pedunculate (Figs 5 and 14 AD, P), 549-659 (606, n = 4, 45) in diameter; disk peduncle 200, measured in the specimen of figure 14, observed with SEM. Eye spots round, with red pigment in live specimens (Fig. 7); pigment dissolving if specimens are fixed in ethanol, but not in formalin.

Epidermal mosaic (demonstrated through staining with silver nitrate) with two dorso-lateral, post tentacular, elliptical, 'excretory' syncytial plates (Figs 8 and 9), with sinuous contour (varying among individuals) (Figs 9 and 13), extending from base of first and fifth tentacles, respectively; left plate 467 long, 375 wide, right plate 476 long, 366 wide; length of 'excretory' syncytial plates/total body length without tentacles relationship 17.8-18.2%. Excretory pore (nephridiopore) inside of each 'excretory' syncytial plate, at approximately equatorial level, but displaced toward the inner side of the plate (Figs 8, 9 and 13 N). Secretion bodies with a varied or polyhedral shape, present in tegument of the adhesive disk (Fig. 30).

Alimentary system

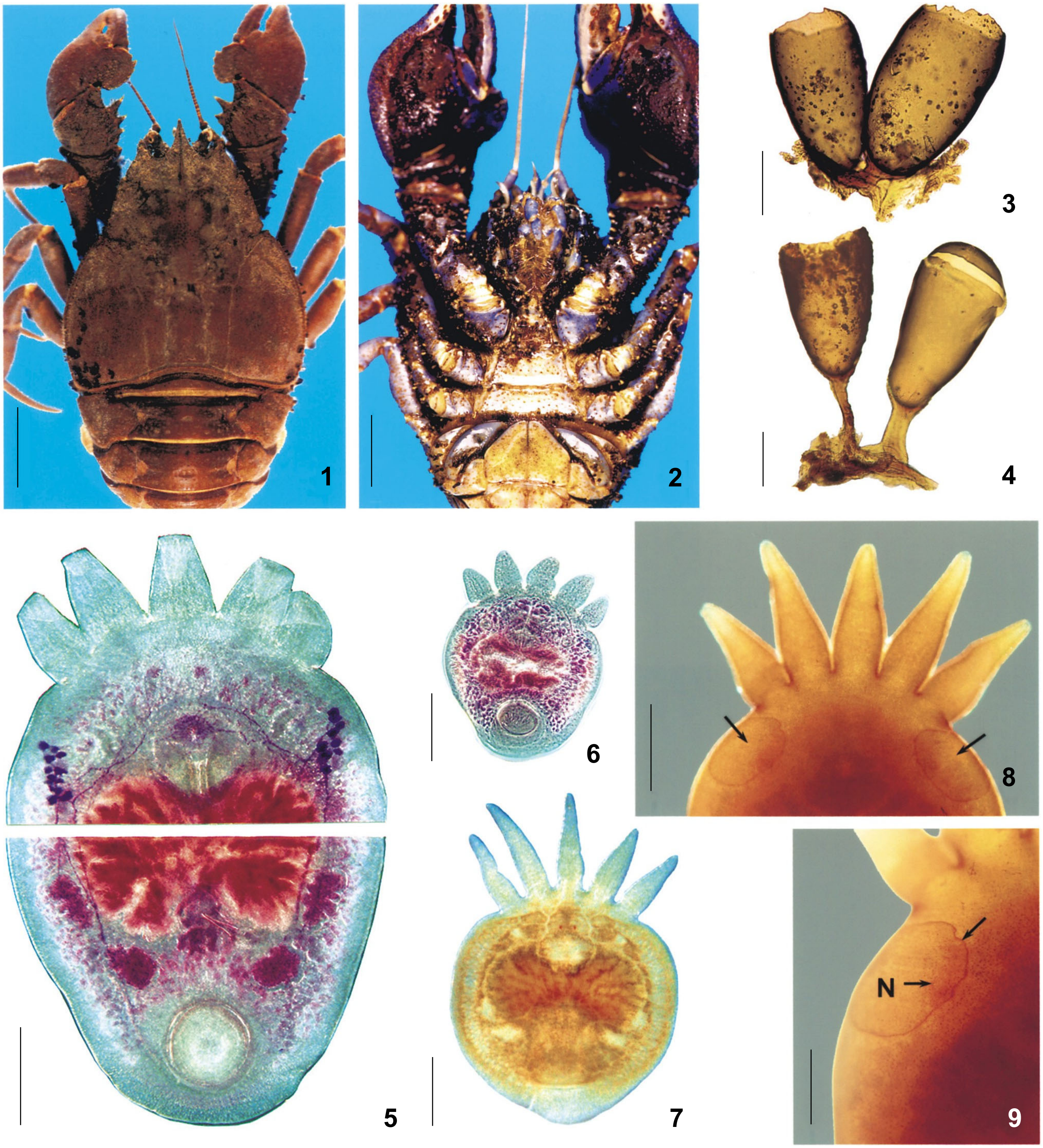
Mouth surrounded by muscular pharynx (Figs 5, 10 and 24), between first and second thirds of body; pharynx wider than long (Figs 5, 10, 24 and 25 PH), 194-344 (257, 43.37) long, 305-444 (366, 44.36) wide; esophageal glands surrounding it at base (Fig. 25), staining deep-purple; intestinal sac wider than long, not surpassing the cyanophilous glands (CGDs) on both sides of body, with conspicuous septations in young, ill-defined septations in adults.

Excretory system

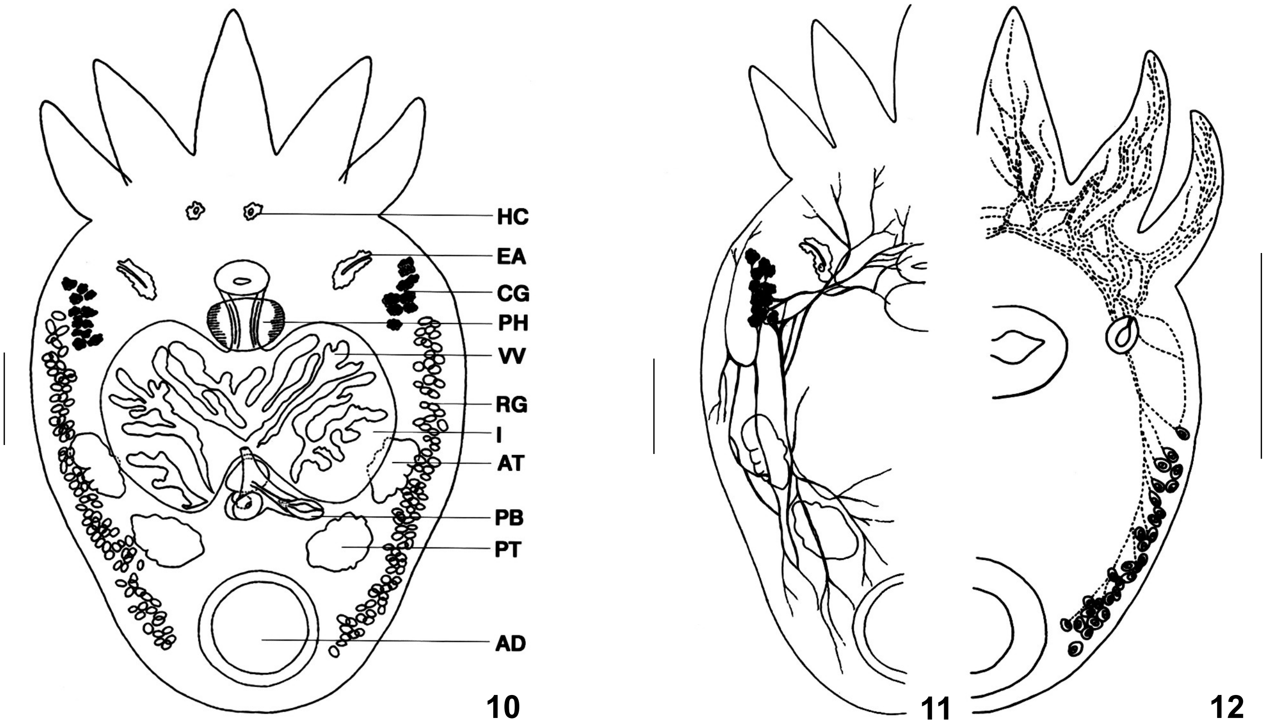
Excretory ampullae at level of mouth (Fig. 27 EA, N), generally directed outwards, many times with the shape of a boomerang. The excretory pore is located at equatorial level, but displaced to the dorsal side of the plate.

Glands

Rhabdite producing glands (Figs 5, 10, 12 and 26), numerous, small, spheroid, 64-91 (73, 9.36), in lateral fields, spreading from level of pharynx to mid-level of adhesive disk, leaving a space to body margin (Figs 5, 10 and 26), appearing with a white glow under phase contrast (Fig. 5); rhabdite tracts clearly visible, uniting just before entering tentacles (Fig. 29). Cyanophilous glands unusually conspicuous and typical in this species (Figs 5, 10, 11 and 26 CG), forming two irregular-shaped, grape-like bunches of approximately 10-15 cells, 7-14 (10, 2.40), staining deep-purple with hematoxylin, each bunch in either side of body, at level of pharynx; ducts of cyanophilous glands staining light purple (Figs 5 and 26 CGD), forming a network with main ducts going anteriad in the direction of the mouth and posteriad in the direction of the adhesive disk, making loops between intestinal sac and body margin. Two Haswell´s cells, showing affinity with hematoxylin, in front of the eyes



Figures 1-9. (1-2) *Aegla serrana*, dorsal and ventral views showing eggs of temnocephalans, bars = 10 mm; (3-4) eggs: (3) sessile eggs; (4) pedunculated eggs, bars = 200 µm; (5-9) *Temnocephala cyanoglandula* **sp. nov.**: (5) holotype, bar = 500 µm; (6) young specimen, showing the circular mass of vitellaria over the intestinal sac, bar = 250 µm; (7) live, adult specimen, showing red pigment of eyes and pattern of orange body pigment, bar = 800 µm; (8) syncytial 'excretory' plates seen in specimen stained with silver nitrate, with off-centered nephridiopores (arrows), bar = 500 µm; (9) syncytial 'excretory' plates seen in specimen stained with silver nitrate, bar = 125 µm. (N) Nephridiopore, arrow indicating sinuous perimeter of syncytial plate.



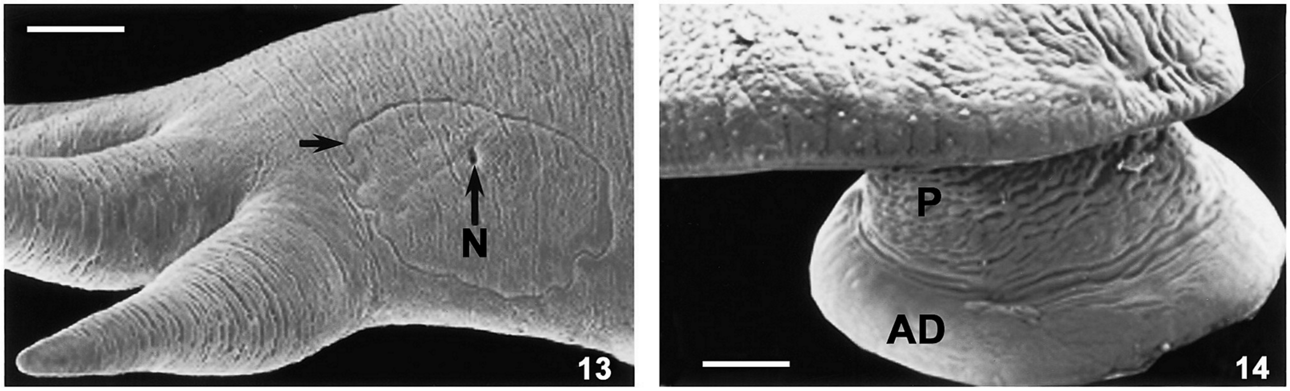
Figures 10-12. *Temnocephala cyanoglandula* **sp. nov.** (10) Holotype, diagrammatic; (11) Cyanophilous glands and the main ducts; (12) Rhabdite glands extending to midlevel of the adhesive disk and its ducts entering the tentacles. (AD) adhesive disk), (AT) anterior testis, (CG) cyanophilous glands, (EA) excretory ampullae, (HC) Haswell´s cells, (I) intestinal sac, (PH) pharynx, (PB) prostatic bulb, (PT) posterior testis, (RG) rhabdite glands, (VV) vitellaria. Bars = 500 µm.

and brain transverse band (Figs 5, 10 and 24 HC); left cell in pair 82-128 (104, 16) across, right cell 82-146 (116, 19) across. Esophageal glands stained reddish-purple, forming collar (Fig. 25) between pharynx and intestinal sac. Ootype glands inconspicuous. Disc glands between adhesive disc and genital complex. Shell glands light brown, between testes of the same side (Fig. 28 SG).

Reproductive system

Female. Gonopore between middle and posterior thirds of body; components of female reproductive system characteristically incospicuous and difficult to observe in this species; ovary difficult to observe and measure; seminal receptacles not observed; vitellaria dendritic in adult specimens, never exceeding limits of intestinal sac, dorsally and ventrally (Figs 5, 7, 10 and 26); in young specimens (Fig. 6) beginning development with the appearance of a circular mass, with an "open" central area, without lateral projections to margins of the body, which are occupied by rhabdite producing glands, cyanophilous glands and anterior testes, leaving a relatively wide space to the lateral margins of body (Figs 5, 10 and 26); vagina and genital atrium incospicuous; sphincter well developed, opening in front of cirrus introvert; vesicula resorbens 153-209 (190, n = 3, 32) long, 148-218 (191, n = 3, 37) wide, indenting intestinal sac and vitellaria posteriorly. Eggs deposited without a preferred location on host, but always on the external surface of hosts (Figs 1 and 2), mainly on ventral side; sessile eggs (Fig. 3), 550-641 (611, n = 5) long, and pedunculate eggs (Fig. 4), 797-934 (782, n = 5) long; two eggs measured 733 and 779 long, respectively.

Male. Testes four, lobated, anterior pair with deeper lobations than posterior pair, slightly oblique; anterior pair lateral to intestinal sac, posterior pair always immediately posterior to intestine; right anterior testis 256-421 (356, 55) long; 146-302 (231, 47) wide; right posterior testis 183-329 (282, 54) long, 266-412 (368, 44) wide; left anterior testis 202-384 (308, 71) long; 229-330 (287, 30) wide; left posterior testis 165-421 (277, 74) long; 275-476 (400, 53) wide; both deferent vessels uniting to form small seminal vesicle, before entering small, oval, prostatic bulb, 146-201 (182, 20) long, 62-110 (88, 17) wide; genital capsule present, observed in frontal section (Fig. 22); cirrus when viewed from top, apparently straight, (Figs 10, 15, 17 and 19-21), slightly curved in lateral view, 238-266 (256, n = 6, 11) long, shaft 207-238 (224, n = 6, 11) wide, introvert with fine spines (Figs 15-18) 28-37 (31, n = 6, 3) long, shaft base 57-68 (64, 45) wide; introvert never observed more extroverted than is shown in figures 15-21. In transversal section (Fig. 23), there are 29 rows of spines, which are as teeth of a comb, with the "handle" positioned backwards (Fig. 18). Proximal limit of introvert marked with very small teeth-like protuberances (Figs 15-18). Ratio between total length of cirrus and maximum width of shaft base 2.89.



Figures 13-14. *Temnocephala cyanoglandula* **sp. nov.**, SEM. (13) Antero-lateral area showing leftmost tentacle and left dorso-lateral 'excretory' syncytial plate (N) nephridiopore, arrow showing sinuous perimeter, bar = 100 µm; (14) posterior portion of body showing adhesive disk (AD) and peduncle (P), bar = 50 µm.

Type host. *Aegla serrana* Buckup & Rossi, 1977; specimens deposited in the Coleção de Crustáceos do Departamento de Zoologia, UFRGS, 2486.

Site. branchial chambers and body surface; eggs cemented over external surfaces of exoskeleton, in different regions of the ventral side: perioral area, pleural strips, esternal plates, pereiopods and chelipods; to a lesser extent on the dorsal side of cephalothorax and dorsal side of uropods.

Type locality. creek and small reservoir in Utopia II Farm, 5 km East of the State Road RS-020, locality of Tainhas, Municipality of Cambará do Sul, Rio Grande do Sul, Brazil (29º15'10"S, 50º15'45"W).

Specimens deposited. CHIOC # 36199 – holotype, # 36197, 36198, 36200-36202 – paratypes, # 36203, 36204 – cirrus in de Faure´s mounting medium.

Etymology. The specific epithet '*cyanoglandula*' is a composite word and refers to the very distinctive appearance of the cyanophilous glands, which, in this species, appear as two irregular-shaped, grape-like bunches of cells, located in the anterior portion of the body, at the level of mouth and pharynx.

Remarks

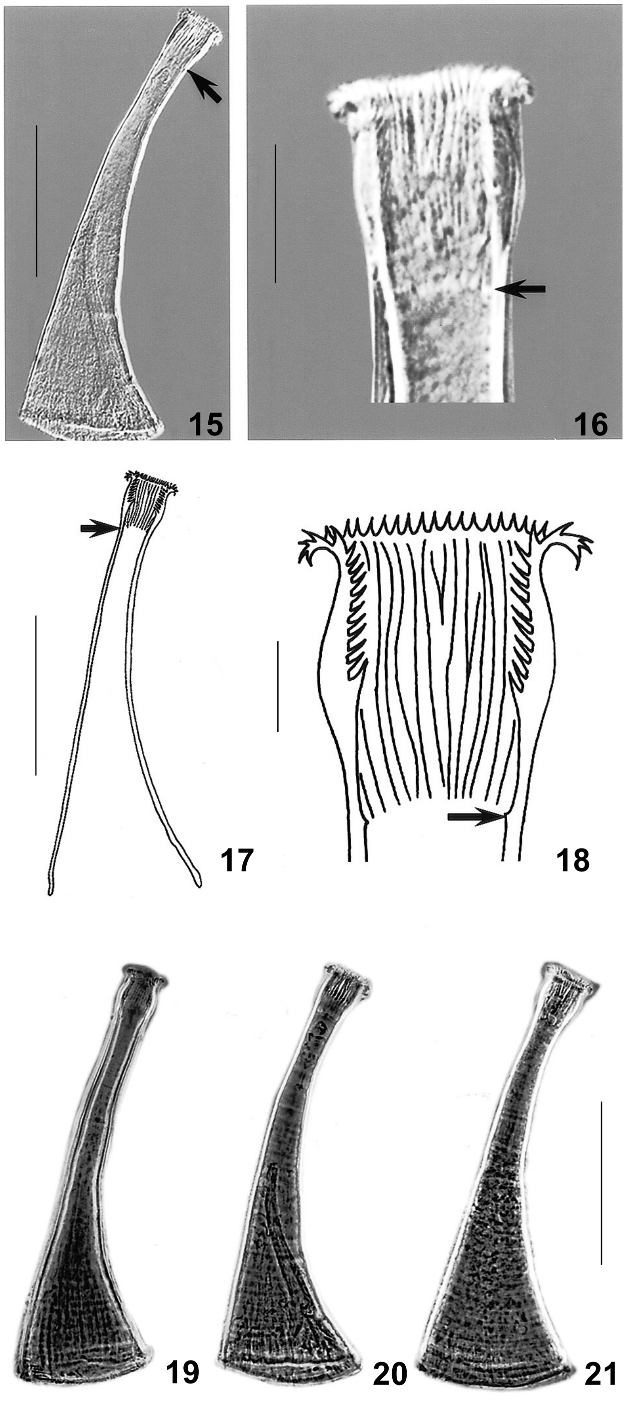
Temnocephala cyanoglandula **sp. nov.** is the second species described from an anomuran crustacean from Brazil. It is a distinct species in various aspects. The specific name refers to the cyanophilous glands, which stand out as deep-blue (affinity with hematoxylin), irregular-shaped, grape-like bunches which send their common ducts (Figs 5, 10, 11 and 26 CDG) forward in the direction of the mouth and backward, in the direction of the adhesive disk, winding back and forth while their diameter decreases in size. Williams (1980a), in the description of *Temnosewellia geonoma* (Williams, 1980) discussed at length these glands and their ducts, which are present in the New Zealand species, although she has drawn them diagrammatically not allowing a good comparison with what are observing in the specimens.

As *T. cyanoglandula* **sp. nov.** is the first Brazilian species to be studied through a suit of different techniques, which require live specimens to be killed with different fixatives, it is not possible to compare the many features observed with the same features observed by other authors in other species, except the 'excretory' syncytial plates, seen under SEM or through SN staining. In regard to this character the species differs from the seven species illustrated by Damborenea & Cannon (2001) by being elliptical, having a partially sinuous perimeter and by having the nephridiopore displaced toward the inner side of the syncytial plate. The syncycial plates also differ from those observed in the specimens of *T. lutzi* by being elliptical and not elongated, as Damborenea & Cannon (2001) have shown for *T. microdactyla*.

A photomicrograph taken from a live specimen (Fig. 7) shows that *T. cyanoglandula* **sp. nov.** has a well-marked pattern of faint orange pigmentation, which together with the red pigment of the eyes (Fig. 7) dissolves when the specimens are transferred to ethanol. The red pigment of the eyes is not affected by the fixation with formalin remaining with same size and intensity if left in that fixative.

So far, authors have not considered the size, distribution and position of the rhabdite glands to be of much systematic value. These cells and the rhabdites they secrete have been well studied by Williams (1980a), Williams & Ingerfeld (1988) and illustrated by Cannon (1993), Sewell & Cannon (1998) when describing new species of temnocephalans from Australia, while Ponce de Léon (1979, 1989) illustrated them when describing *T. rochensis* and *T. haswelli*, respectively, from Uruguay. It was also observed that the specimens of *T. iheringi* have the rhabdite producing glands completely anterior to testes.

The cyanophilous secretion produced by the cyanophilous glands has been referred by Williams (1980b) to form a film covering dorsal and ventral surfaces of temnocephalans staining selectively with alcian blue by the method of Steedman (1950), but they also are stained selectively by Delafield hematoxylin, as it is demonstrated in *T. cyanoglandula* **sp. nov.** The new species, characteristically, shows not only the unicellular cyanophilous glands but also the paths of their ducts (Figs 5, 11, 24 and 26), and a multitude of ductules filled with blue stained mucus. Figure 30 shows some of the accumulated secretion bodies with the "varied or polyhedral" shape described



Figures 15-21. *Temnocephala cyanoglandula* **sp. nov.** (15-16) Photomicrographs of cirri, seen with Nomarski´s interference microscopy, showing the entire organ and the introvert, bars = 100µm and 25 µm, respectively; (17-18) line drawings of entire cirrus and introvert, respectively, bars = 100 µm and 25 µm; (19-21) intraspecific variation of the cirrus of three specimens, seen in phase contrast, bar = 100 µm. Arrows indicate proximal limit of introvert.

by Williams (1975).

The cirrus in *T. cyanoglandula* **sp. nov.** is characteristic of the new species, not only in the shape and size of the shaft, but in the characteristics of the introvert. None of the cirri photographically illustrated with Nomarski´s microscopy by Damborenea & Cannon (2001) appear to be similar. Figures 15-21 show that one of the sides of the shaft is nearly straight while the other is curved. This can mislead the observer when this cirrus is seen in the dorsal or ventral position. With the micro-dissection of the cirrus and mounting in deF, one can observe it from the side, removed from the parenchyma. Figures 15-16 show the introvert seen with Nomarski´s interference contrast filters, allowing the observation of the posterior limit of the spine rows of the introvert. Figure 23, shows a transversal section of the introvert with 29 rows of spines. The distal rim of the introvert bends out as an open carnation. So far, none of the specimens studied have shown the introvert completely extroverted, as is seen in many Australian species, or in *T. chilensis* (Moquin-Tandon, 1849) as shown by Damborenea & Cannon (2001). Maybe in *T. cyanoglandula* the introvert does not extrovert completely. There is a slight intraspecific variation in the shape of the cirrus introvert, which can be seen in figures 19-21. As the same techniques are applied to other species of *Temnocephala* it will be possible to gain insight in relation to the specific variation of many characters clearly visible in *T. cyanoglandula* **sp. nov.**

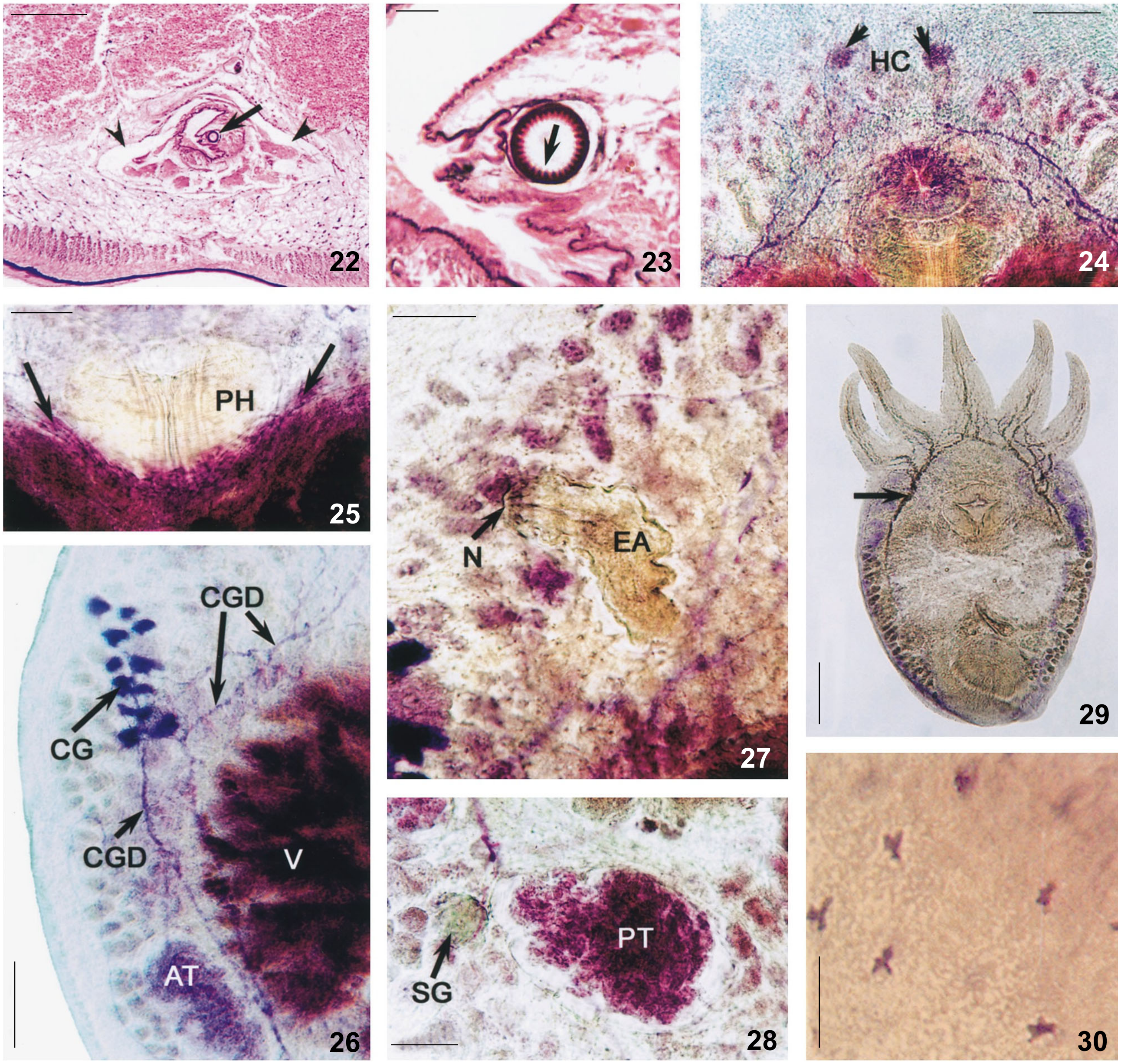
ACKNOWLEDGEMENTS

To Dr. Georgina Bond-Buckup (UFRGS), for identifying the species of *Aegla*, as well as for providing GPS data and to Dr. Ludwig Buckup (UFRGS), for providing the first hosts positive for temnocephalans and access to Romeis (1968); to the Departamento de Zoologia, UFRGS, for making space available to the senior author; to the Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS) for the grant given to the second author; and to PIBIC/CNPq/UFRGS, for the scholarship given to the junior author; to Prof. Abner Chiquieri, Departamento de Línguas, Universidade Federal Rural do Rio de Janeiro, Seropédica, Rio de Janeiro, for his advice on the correct formation of the specific name of the new species; to Dr. L.R.G. Cannon, Queensland Museum, Australia, for providing literature; to Dr. M.C. Damborenea, Curator of Helminths, Museo de La Plata, La Plata, Republica Argentina, for the loan of specimens; to Dr. Gilson R. Moreira for the aid with the SEM, as well as to the technicians of the Centro de Microscopia Eletrônica, UFRGS, for their expert assistance; to Mr Márcio C. Poisl, owner of the Utopia II Farm, for collecting some hosts with traps and allowing us to collect host specimens in his property; to Samantha A. Seixas and Cassandra de M. Monteiro for their invaluable help; and to Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Brazil, for the Senior Visiting Research Fellowship given to the senior author, from 1997 to 1999.

REFERENCES

Buckup, L. & G. Bond-Buckup. 1999. **Os crustáceos do Rio Grande do Sul.** Porto Alegre, Editora Universidade, UFRGS. 503p.

Cannon, L.R.G. 1993. New temnocephalans (Platyhelminthes): ectosymbionts of freshwater crabs and shrimps. **Memoirs of the Queensland Museum**, South Brisbane **33**: 17-40.



Figures 22-30. *Temnocephala cyanoglandula* **sp. nov.** (22) Sagital, histological section, showing the genital capsule (head arrows) and a transversal section of the cirrus introvert (arrow), bar = 100 µm; (23) transversal section through the cirrus introvert (arrow), in higher magnification, where is possible to count 29 spine rows, bar = 25 µm; (24) mouth region, showing the Haswell's cells (HC – arrows) and ducts of the cyanophilous glands, bar = 200 µm; (25) pharynx (PH) and esophageal glands (arrows), bar = 100 µm; (26) lateral side of holotype showing the irregular-shaped, grape-like bunch of cyanophilous glands, its ducts, anterior testis (AT), vitellaria (V) over the intestinal sac and the rhabdite glands, bar = 200 µm; (27) excretory ampullae (EA) with the characteristic boomerang shape – arrow indicates the nephridiopore (N), bar = 100 µm; (28) posterior testis (PT) and one of the brownish, shell glands (SG – arrow), bar = 100 µm; (29) rhabdite glands extending to midlevel of the adhesive disk and its ducts entering the tentacles (arrow), bar = 500µm; (30) tegument bodies in the adhesive disc, showing irregular shape, bar = 20 µm.

CANNON, L.R.G. & K.B. SEWELL. 1995. Craspedellinae Baer, 1931 (Platyhelminthes: Temnocephalida) ectosymbionts from the branchial chamber of Australian crayfish (Crustacea: Parastacidae). **Memoirs of the Queensland Museum**, South Brisbane, **38**: 397-418.

DALLWITZ, M.J. & T.A. PAINE. 1986. **User's guide to the Delta system: a general system for processing taxonomic descriptions.** Camberra, CSIRO Entomology. **Report 13**, 106p.

DAMBORENEA, M.C. & L.R.G. CANNON. 2001. On neotropical *Temnocephala* (Platyhelminthes). **Journal of Natural History**, London, **35**: 1103-1118.

DIONI, W.L. 1967. Temnocephalas Uruguayas II Descripción de *T. talicei* **sp. nov.** y notas sobre *T. axenos* Monticelli (Platyhelmintha). **Physis**, Buenos Aires, **26** (73): 477-484.

HUMASON, G. 1972. **Animal Tissue Techniques.** San Francisco, W. H. Freeman, 3rd ed., XV+641p.

JOFFE, B.I.; I.V. SOLOVEI & L.R.G. CANNON. 1995. The structure of the epidermis in *Didymorchis* (Temnocephalida: Platyhelminthes). **Australian Journal of Zoology**, Victoria, **43**: 631-641.

PONCE DE LEÓN, R. 1979. Especies americanas de Temnocephalidea Benham (Platyhelmintha), I. Descripción de *Temnocephala rochensis* **sp. nov.** de la camara paleal de *Pomacea canaliculata* (Lamarck). **Revista de Biología del Uruguay**, Montevideo, **7**: 39-48.

————. 1989. Description of *Temnocephala haswelli* **sp. nov.** (Platyhelminthes) from the mantle cavity of *Pomacea canaliculata* (Lamarck). **Journal of Parasitology**, Lawrence, **75**: 524-526.

ROMEIS, B. 1968. **Taschenbuch der microskopischen Technik.** 16. Aufl. Oldenbourg, Munchen. XIV+695p.

SEWELL, K.B. & L.R.G. CANNON. 1998. New temnocephalans from the branchial chamber of Australian *Euastacus* and *Cherax* crayfish hosts. **Proceedings of the Linnaean Society**, New South Wales, **119**: 21-36.

STEEDMAN, H.F. 1950. Alcian blue 8GS: a new stain for mucin. **Quarterly Journal of Microscopical Science**, London, **91**: 477-480.

WILLIAMS, J.B. 1975. Studies on the epidermis of *Temnocephala* I. Ultrastructure of the epidermis of *Temnocephala novaezaelandiae*. **Australian Journal of Zoology**, Victoria, **23**: 321-331.

————. 1980a. Morphology of a species of *Temnocephala* (Platyhelminthes) ectocommensal on the isopod *Phreatoicopsis terricola*. **Journal of Natural History**, London, **14**: 183-199.

————. 1980b. Studies on the epidermis of *Temnocephala* V. Further observations on the ultrastructure of the epidermis of *Temnocephala novae-zealandiae*, including notes on the glycocalyx. **Australian Journal of Zoology**, Victoria, **28**: 43-57.

WILLIAMS, J.B. & M. INGERFELD. 1988. Cells in the parenchyma of *Temnocephala*: rhabdite-secreting cells of *Temnocephala novaezealandiae* (Temnocephalidae: Platyhelminthes). **International Journal for Parasitology**, Elmsford, **18**: 651-659.

Received in 18.III.2003; accepted in 21.VIII.2003.