

## ***Nitella (Streptophyta, Characeae) from southern Brazil***

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**ABSTRACT** – In this study, we analyzed 100 samples of *Nitella* C. Agardh *emend.* A. Braun *emend.* Leonhard from four different Herbaria (ICN, SP, UNOP-Algae and UPCB), which represented 66 different locations from the southern region of Brazil. Twenty-two species of *Nitella* (*Streptophyta, Characeae*) were described and illustrated from the states Paraná, Santa Catarina and Rio Grande do Sul, eight of which are new records for the region. Considering the three states studied, three species were new records for Paraná [*N. flagellifera* J. Groves & G.O. Allen, *N. mucronata* (A. Braun) Miquel *in* H.C. Hall and *N. opaca* (C. Agardh *ex* Bruzelius) C. Agardh], three species for Santa Catarina (*N. blankinshipii* T.F. Allen, *N. flagellifera* J. Groves & G.O. Allen and *N. subglomerata* A. Braun) and another two for Rio Grande do Sul [*N. flexilis* (L.) C. Agardh and *N. microcarpa* A. Braun].

**Keywords:** *Charales*, green algae, morphology, taxonomy

**RESUMO** – *Nitella (Streptophyta, Characeae) do sul do Brasil.* O estudo envolveu a análise de 100 amostras de *Nitella* C. Agardh *emend.* A. Braun *emend.* Leonhard, depositadas em quatro herbários (ICN, SP, UNOP-Algae e UPCB) representando 66 localidades da região sul do Brasil. Vinte e duas espécies de *Nitella* (*Streptophyta, Characeae*) foram descritas e ilustradas para os estados do Paraná, Santa Catarina e Rio Grande do Sul, oito são novos registros para a região. Considerando os três estados estudados, três espécies são novos registros para o Paraná [*N. flagellifera* J. Groves & G.O. Allen, *N. mucronata* (A. Braun) Miquel *in* H.C. Hall e *N. opaca* (C. Agardh *ex* Bruzelius) C. Agardh], três para Santa Catarina (*N. blankinshipii* T.F. Allen, *N. flagellifera* J. Groves & G.O. Allen e *N. subglomerata* A. Braun) e outras duas para o Rio Grande do Sul [*N. flexilis* (L.) C. Agardh e *N. microcarpa* A. Braun].

**Palavras-chave:** *Charales*, algas verdes, morfologia, taxonomia

## **INTRODUCTION**

The genus *Nitella* C. Agardh *emend.* A. Braun *emend.* Leonhard includes a diverse and complex group of macroalgae found in all continents except Antarctica (Wood & Imahori 1959, 1964, 1965; Mann *et al.* 1999; Martin *et al.* 2003). Studies on charophytes are relevant to understand the evolution of land plants. Ruhfel *et al.* (2014) found streptophytic algae to be monophyletic with land plants, for example, *Zygnematophyceae* are sister to land plants, *Coleochaetophyceae* are sister to *Zygnematophyceae + Embryophyta*, *Charophyceae* are sister to *Coleochaetophyceae + (Zygnematophyceae + Embryophyta)*, and a clade of *Mesostigmatophyceae + Chlorokybophyceae* is sister to all other *Streptophyta*. Phylogenetic studies support that embryophytes were derived from the charophytes, the stoneworts being the closest living relatives to the land plants (Karol *et al.* 2001). Furthermore, charophytes are an important group of submerged aquatic vegetation with several important roles in both brackish and freshwater ecosystems (Coops 2002), and some species can be considered as good water quality indicators due to their sensitivity to eutrophication

(Torn *et al.* 2004; Cirujano *et al.* 2008).

In Brazil, studies on *Characeae* began in the 1960's with Prof. Dr. Rosa Maria Teixeira Bicudo dedicating her work to the taxonomy of the group (Bicudo 1968a, b, 1969). Later, other researchers dedicated their study to the group involving its taxonomy and ecology, though the study of *Characeae* in Brazil is still rare.

Studies on *Nitella* in Brazil include those of Bicudo & Yamaoka (1978), Picelli-Vicentim & Bicudo (1990, 1993), Branco & Necchi-Júnior (1996, 1998), Branco *et al.* (2005), Bueno & Bicudo (1997, 2006, 2008), Dias & Araújo (2001), Torgan *et al.* (2001), Vieira-Júnior & Necchi-Júnior (2002, 2006), Vieira-Júnior *et al.* (2002), Prado (2003), Picelli-Vicentim *et al.* (2004), Prado & Baptista (2005), Araujo *et al.* (2010), Bicudo & Bueno (2011) and Bueno *et al.* (2011). Considering the relevance of this group in aquatic ecosystems and the importance of understanding species diversity, we carried out a taxonomic survey of the Brazilian charophyte flora focusing on the southern region of Brazil that includes the states Paraná, Santa Catarina and Rio Grande do Sul. We present the morphological analysis and variation of specimens of *Nitella* and briefly compare them with those recorded in previous publications.

## MATERIAL AND METHODS

One hundred samples were collected from 66 different locations in the state Rio Grande do Sul and four others in each one of the states Paraná and Santa Catarina, collected in years 1975, 1978, 1980, 1996, 2000, 2001, 2002, 2008 and 2009. Specimens examined were borrowed from the following herbaria: SP (Herbário do Estado "Maria Eneyda P. Kaufmann Fidalgo", Institute of Botany), UPCB (Herbarium of the Botany Department, Federal University of Paraná), UNOP-Algae (Herbarium of the State University of Western Paraná) and ICN (Herbarium of the Institute of Biosciences, Federal University of Rio Grande do Sul). Herbaria abbreviations follow *Index Herbariorum* (Thiers 2015). Morphological characteristics and their intra and interspecific variations were studied and compared with those recorded in previous publications for Brazil. Sampling sites are illustrated in Figure 1.

Rio Grande do Sul State specimens were collected from natural marshes, reservoirs and canals built for the irrigation of the surrounding crops. These environments have muddy bottoms and their depth varied from 0.2 to 1.5 m. Specimens from the coastal region (lagoons and

lakes) were living on sandy bottoms, in clear water, at depths of 1.5 to 2 m, and were subject to influence from the sea. In all sites, the *Characeae* formed large meadows or patches among other macrophytes. The specimens from the states Paraná and Santa Catarina were collected from lotic and lentic environments, and were already deposited in herbaria without precise information on the environment where they were collected.

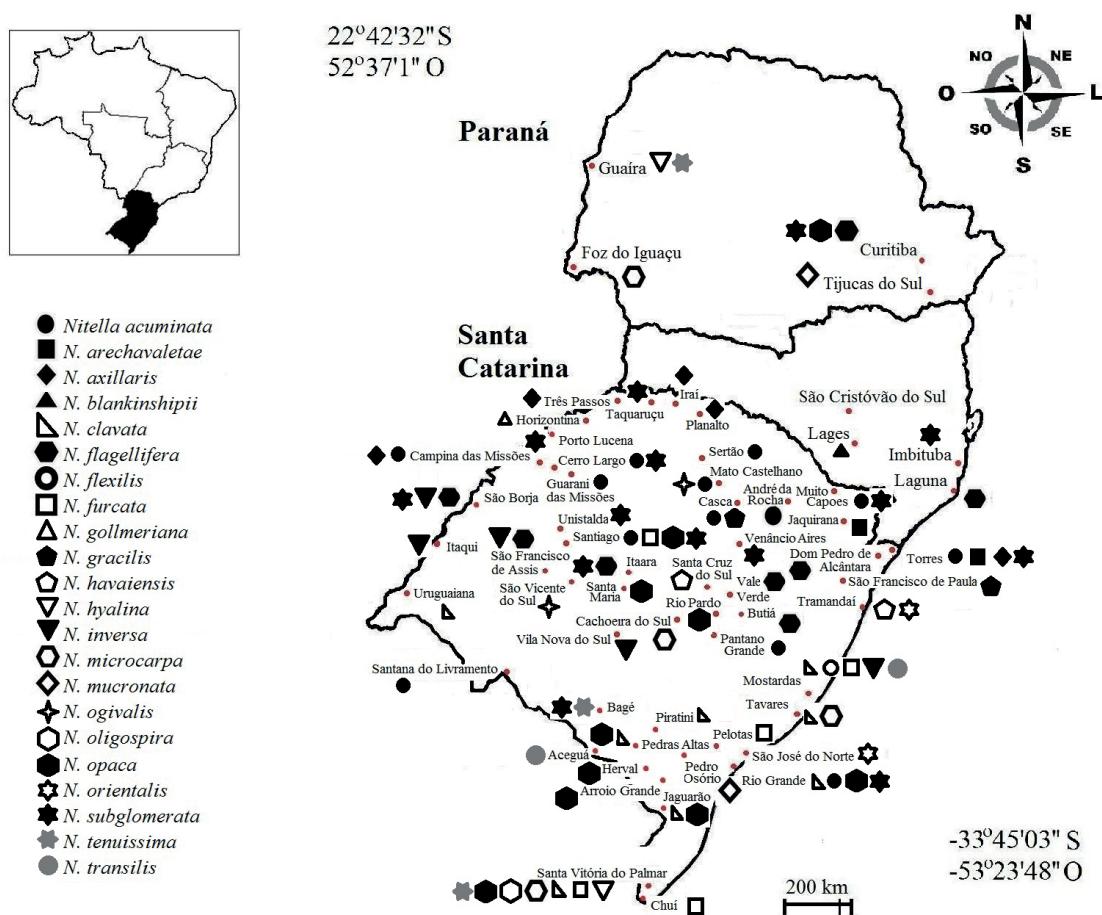
Samples deposited in ICN were collected and taken directly to the laboratory, where the specimens were cleaned with fresh water to be analyzed under the light microscope. SP, UPCB and UNOP-Algae herbaria were specimens loaned. These specimens were preserved as exsiccatae and/or in Transeau solution. Plant examination followed the methodology in Bicudo & Menezes (2006); morphometric characteristics used in the identifications were taken from each plant examined. The classification used here followed Krause (1997) and the following literature was consulted to identify and discuss the specimens: Groves & Groves (1911), Wood & Imahori (1964, 1965) Moore (1986), Sakayama *et al.* (2002), Schubert & Blindow (2004), Picelli-Vicentim *et al.* (2004), Cirujano *et al.* (2008) and Scribailo & Alix (2010).

## RESULTS AND DISCUSSION

### Key for the species of *Nitella* studied in Paraná, Santa Catarina and Rio Grande do Sul States, Brazil.

1. 1-celled dactyls.
  2. Plant monoecious.
    3. Branchlets monomorphic.
      4. Coronula persistent ..... *N. acuminata*
      4. Coronula deciduous ..... *N. flexilis*
    3. Branchlets dimorphic.
      5. Heads lax, loose ..... *N. subglomerata*
      5. Heads dense, thick.
        6. Dactyls inflated ..... *N. clavata*
        6. Dactyls not inflated ..... *N. gollmeriana*
    2. Plant dioecious.
      7. Branchlets dimorphic ..... *N. blankinshipii*
      7. Branchlets monomorphic ..... *N. opaca*
  1. 2-3-more-celled dactyls.
    8. Different branchlets at the same whorl (heteroclemous) ..... *N. hyalina*
    8. Similar branchlets at the same whorl (homoclemous).
      9. Branchlets monomorphic.
        10. Dactyls dominantly abbreviated.
          11. Oogonia inverse, terminal ..... *N. inversa*
          11. Oogonia in normal position.
            12. 2-3 oogonia at a furcation ..... *N. microcarpa*
            12. Up to 6 oogonia at a furcation ..... *N. furcata*
        10. Dactyls dominantly elongated.
          13. Dactyls 2-celled.
            14. Micro and macro dactyls predominantly elongated, 2-3 oogonia at a branchlet furcation ..... *N. flagellifera*
            14. Regularly elongated dactyls.
              15. Gametangia absent from 1<sup>st</sup> furcation.

16. Oogonia 1-3 at a node; oospore wall granulate ..... *N. orientalis*  
 16. Oogonia solitary; oospore wall reticulate to papillate.  
   17. Oospore wall reticulate to papillate ..... *N. tenuissima*  
   17. Oospore wall reticulate ..... *N. transilis*  
 15. Oogonia at the base of all furcations.  
   18. Oogonia solitary or geminated;  
     branchlets occasionally reduced ..... *N. mucronata*  
   18. Oogonia 1-3 at a node, occasionally 3-celled dactyls ..... *N. oligospira*  
 13. Dactyls 2 or more celled.  
   19. Gametangia present at the base of all furcations ..... *N. ogivalis*  
   19. Gametangia present at the base of 2<sup>nd</sup>-4<sup>th</sup> furcation, absent at the 1<sup>st</sup> ..... *N. gracilis*  
 9. Branchlets dimorphic.  
 20. Branchlets with 1 furcation; 2-celled dactyls ..... *N. axillaris*  
 20. Branchlets with 1-3 furcations; multicellular dactyls.  
 21. Branchlets with 1-2 furcations; dactyls 2-3(-4)-celled,  
   1-3 oogonia at a node; oospore finely reticulate ..... *N. arechavaletae*  
 21. Branchlets with 1-3 furcations; 3 oogonia at a node;  
   oospore granulate ..... *N. havaiensis*



**Fig. 1.** Geographical distribution of *Nitella* in Paraná, Santa Catarina e Rio Grande do Sul States.

**Nitella acuminata** A. Braun ex Wallman, Försök Syst. Char. 35. 1853.

(Figs. 2 A-F)

Plants monoecious. Stem: 10-56 cm tall. Axes: 620-880 µm diam., without incrustation. Internodes: 0.5-7 cm long. Branchlets: monomorphic, furcated. Sterile branchlets: 6-8, 1-furcate, 1-2.8 cm long × 630-800 µm diam., primary rays 6-8, 0.75-1.85 cm long. Dactyls: 2-5, 1-celled, 2-10 mm long × 200-450 µm diam. Fertile branchlets: 7-9, 3-6 mm long × 220-340 µm diam., 1-furcate. Dactyls 2-5, 1-celled, 1100-2800 µm long × 110-200 µm diam. Heads: not formed. Gametangia: conjoined, sessile or sometimes with stalk. Oogonia: 1-2 at a node, 320-530 µm long × 310-430 µm diam., convolutions 8, coronula 35-55 µm long × 60-70 µm diam. Oospores: 260-275 µm long × 200-250 µm diam., striae of 6-7 ridges, fossa 30-70 µm across, membrane granulate. Antheridia: 330-400 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, André da Rocha, 21.V.2000, J.F. Prado (ICN91603); Campina das Missões, 19.xii.2000, J.F. Prado (ICN91542); Casca, 02.vi.2000, J.F. Prado (ICN91543); Cerro Largo, 15.xi.2000, J.F. Prado (ICN91544); Guarani das Missões, 14.xi.2000, J.F. Prado (ICN91545); Mato Castelhano, 03.vi.2000, J.F. Prado (ICN91546); Muitos Capões, 20.v.2000, J.F. Prado (ICN91547); Pântano Grande, 21.ii.2002, J.F. Prado (ICN91548); Rio Grande, 22.iii.2001, J.F. Prado (ICN91549); Santana do Livramento, 20.iii.2001, J.F. Prado (ICN91550); Santiago, 28.xi.2000, J.F. Prado (ICN 91551), 20.xii.2000, J.F. Prado (ICN91522); Sertão, 28.xi.2000, J.F. Prado (ICN91553); Torres, 18.xi.1999, J.F. Prado (ICN91554).

**Distribution in Brazil:** Rio de Janeiro (Bicudo 1969, Bicudo & Yamaoka 1978); São Paulo (Bicudo 1969, Bicudo & Yamaoka 1978, Picelli-Vicentim & Bicudo 1993, Picelli-Vicentim *et al.* 2004, Bicudo & Bueno 2011); Mato Grosso do Sul (Bueno & Bicudo 1997); Paraná (Meurer & Bueno 2012); Rio Grande do Sul, (Prado 2003, Araújo *et al.* 2010, Rolon *et al.* 2011, This study).

**Comments:** The specimens analyzed showed all diagnostic features reported for *N. acuminata*. The upper and lower metric limits of the coronula length were extended from 37-47 µm reported by Bueno & Bicudo (1997) to 35-55 µm. The upper and lower metric limits of the fossa width also differed from the measurements provided by Bueno & Bicudo (1997), being extended from 42.5-52.5 µm long to 30-70 µm, and the upper limit of the antheridia diameter was extended from 227.5 µm (Bueno & Bicudo 1997) to 400 µm. *Nitella acuminata* is a cosmopolitan species occurring in North, Central and South America, and occasionally in Asia and Africa (Wood & Imahori 1965).

**Nitella arechavaletae** C. Spegazzini, Anales Soc.Cient. Argentina 15: 224. 1883.

(Figs. 2 G-P)

Plants monoecious. Stem: 25 cm tall. Axes: 290-680

µm diam., without incrustation. Internodes: 0.5-4 cm long. Branchlets: dimorphic, furcated. Sterile branchlets: 6-8, 1-2-furcate; 0.7-2 cm long × 100-150 µm diam., primary ray 6-8, as long as the branchlets, 0.2-1 cm long × 100-150 µm diam., secondary ray 3-5, tertiary 2-4. Dactyls: 3-4, 2-4-celled, 1-11 mm long × 85-120 µm diam., end cell conical, acute or mucronate, 10-210(-500) µm long × 35-48 µm diam. Fertile branchlets: 5-7(-8), compacted into axillary heads, short stipitate, with mucus, 1050-1245 µm long × 100-120 µm diam. Dactyls: 2-4-celled, 560-1260 µm long × 40-70 µm diam., end cells 95-279 µm long × 29-45 µm diam. Heads: 3-6, 1-2.5 mm long × 0.5-2.3 mm diam., formed by 3-5 fertile whorls, stalk 1-3 mm long. Gametangia: stipitate, conjoined or sejoined, at lowest furcation of fertile branchlets whorl, enveloped with mucus. Oogonia: 1-3 at a node, 220-400 µm long × 130-390 µm diam., convolutions 8-10, coronula 35-40 µm long × 50-60 µm diam., stalk 70-110 µm long. Oospores: 265-280 µm long × 240-260 µm diam., striae of 7-8 ridges, fossa 20-40 µm across, membrane coarsely reticulate. Antheridia: 195-250 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Jaquirana, 10.v.2000, J.F. Prado (ICN915943, ICN91594); Torres, 23.vi.2000, Faxinal, J.F. Prado (ICN91595).

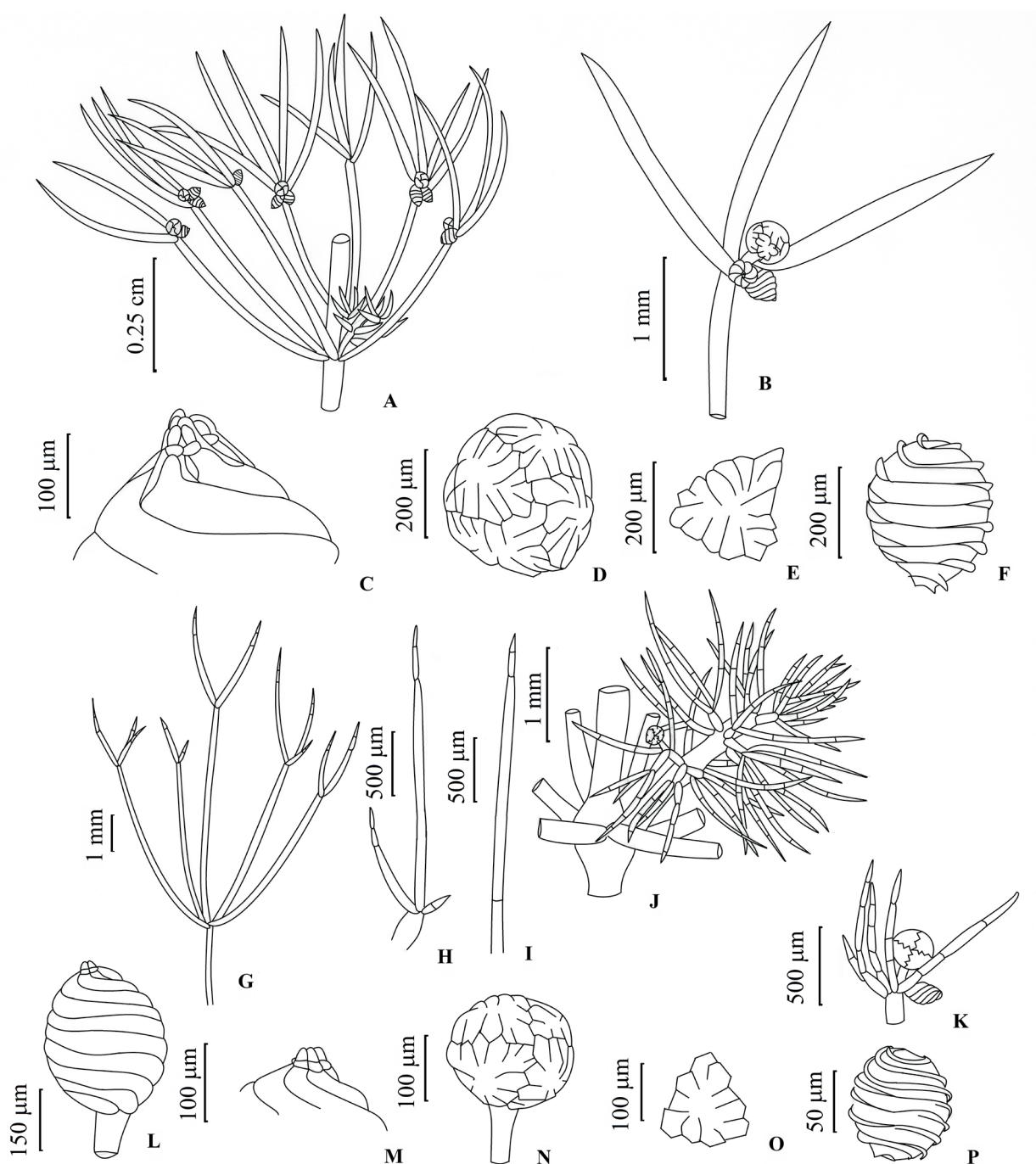
**Distribution in Brazil:** Rio Grande do Sul (Rantzien 1949, Torgan *et al.* 2001, Prado 2003, Araújo *et al.* 2010, This study).

**Comments:** *Nitella arechavaletae* is characterized by the presence of multicellular dactyls, gametangia enveloped in mucilage, fertile branchlets reduced into small heads and the oospore membrane coarsely reticulated. The metric limits of the reproductive structures somewhat differed from Wood & Imahori (1965) from specimens collected in Uruguay. Those of the oogonia increased from 250-340 µm long and 215-365 µm in diameter in Wood & Imahori (1965) to 220-400 µm and 130-390 µm, respectively, in our specimens. The upper limits of the coronula length and diameter increased from 30 µm and 45 µm in Wood & Imahori (1965) to 40 µm and 60 µm, respectively; the upper limits of the oospore length and diameter increased from 270 µm and 240 µm to 280 µm and 260 µm, respectively; and the upper limits of the fossa width and the antheridia diameter also increased from 30 µm and 165 µm to 40 µm and 250 µm, respectively. *Nitella arechavaletae* is restricted to South America (Wood & Imahori 1965).

**Nitella axillaris** A. Braun, Monat. der Deutschen Akad. der Wiss. zu Berlin 1858: 356. 1858.

(Figs. 3 A-G)

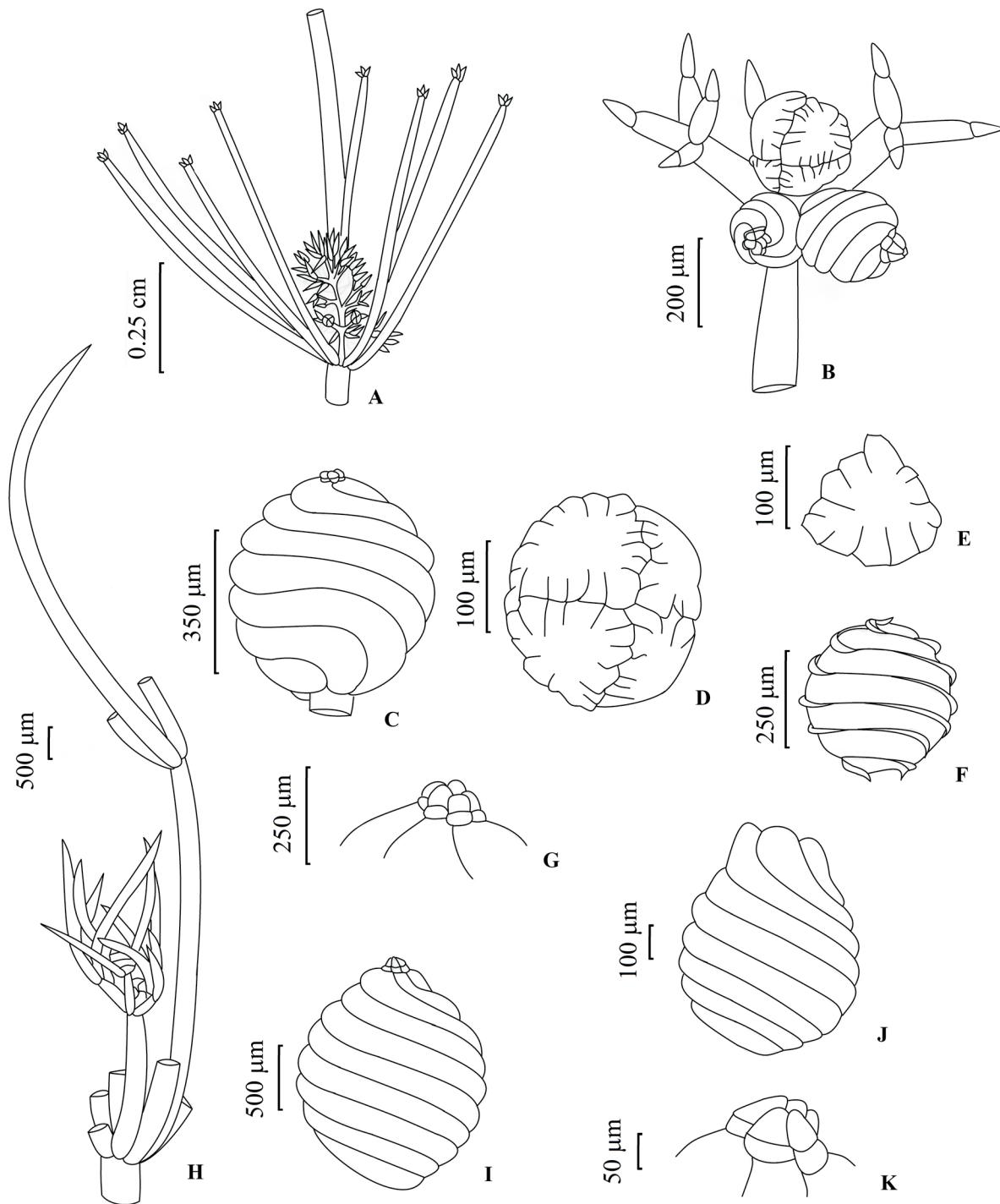
Plants monoecious. Stem: 10-38 cm tall. Axes: 470-1010 µm diam., without incrustation. Internodes: 2.5-7 cm long. Branchlets: dimorphic, apparently simple, although



**Figs. 2 A-P.** **A-F.** *Nitella acuminata*. **A.** Whorl with fertile branchlets; **B.** Fertile node; **C.** Coronula; **D.** Antheridium; **E.** Triangular scute; **F.** Oospore. **G-P** *Nitella arechavaletae*. **G.** Two-furcated sterile whorl; **H.** Two to three-celled dactyl; **I.** Four-celled dactyl; **J.** Axillary Head; **K.** Fertile Branchlet; **L.** Short-stipitate oogonia; **M.** Coronula; **N.** Antheridium with eight scutes; **O.** Triangular scute; **P.** Oospore.

1-furcated, fertile reduced. Sterile branchlets: 6-9, 1-furcate, appearing simple, 0.5-3.4 cm long  $\times$  190-970  $\mu\text{m}$  diam., primary ray 6-9, as long as the branchlets, 0.4-3.4 cm long. Dactyls: 3-5, 2-celled, apex mucronate, 220-1000  $\mu\text{m}$  long  $\times$  70-190  $\mu\text{m}$  diam. Fertile branchlets: 7-8, 1-furcate, 900-1000  $\mu\text{m}$  long  $\times$  80-90  $\mu\text{m}$  diam., reduced into heads.

Dactyls: 3-5, 2-celled, 200-340  $\mu\text{m}$  long  $\times$  50-115  $\mu\text{m}$  diam. Heads: axillary, 1-3 per whorl, sometimes stipitate, heads 6 mm long  $\times$  0.5-4 mm diam. Gametangia: conjoined. Oogonia: 1-3 at a node, 230-520  $\mu\text{m}$  long  $\times$  180-460  $\mu\text{m}$  diam., convolutions 7-8, coronula 40-45  $\mu\text{m}$  long  $\times$  70-80  $\mu\text{m}$  diam. Oospores: 250-370  $\mu\text{m}$  long  $\times$  180-340  $\mu\text{m}$



**Figs. 3 A-K. A-G.** *Nitella axillaris*. **A.** Whorl with one-furcated branchlets, sterile normal and fertile reduced into heads; **B.** Fertile branchlet; **C.** Short stipitate oogonium; **D.** Antheridium with eight scutes; **E.** Triangular scute; **F.** Oospore; **G.** Coronula. **H-K** *Nitella blankinshipii*. **H.** Whorl with fertile and sterile branchlets; **I.** Oogonium; **J.** Oogonium with deciduous coronula; **K.** Coronula.

diam., membrane reticulate, striae of 6-7 ridges, fossa 40-70 µm across. Antheridia: 200-250 µm diam., octoscutate, shields triangular.

(ICN91597, ICN91598); Planalto, 17.xi.2000, J.F. Prado (ICN91599); Três Passos, 16.xi.2000, J.F. Prado (ICN91600, ICN91601).

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Campina das Missões, 19.xii.2000, J.F. Prado (ICN91596); Iraí, 17.xi.2000, J.F. Prado

**Distribution in Brazil:** *Nitella axillaris* [= *N. translucens* (Persoon) C. Agardh emend. R.D. Wood subsp. *translucens* var. *axillaris* (A. Braun) R.D. Wood f. *axillaris*]. Pernambuco

(Wood & Imahori 1965); São Paulo (Picelli-Vicentim & Bicudo 1993, Picelli-Vicentim *et al.* 2004, Bicudo & Bueno, 2011); Mato Grosso do Sul (Bueno & Bicudo 1997), Paraná (Meurer & Bueno 2012), Rio Grande do Sul (Araújo *et al.* 2010, Prado 2003, This study as *Nitella axillaris*).

**Comments:** *Nitella axillaris* [= *N. translucens* (Persoon) C. Agardh *emend.* R.D. Wood subsp. *translucens* var. *axillaris* (A. Braun) R.D. Wood f. *axillaris*] was considered a taxonomic variation of *N. translucens* by Wood & Imahori (1965) because of similar vegetative structures. Moore (1986) reported that *N. translucens* oospores are elongated, ovoid, with a finely reticulate ornamentation and five to six striae. In contrast, Sakayama *et al.* (2002) described *N. axillaris* oospores ovoid, with a strongly reticulate ornamentated wall, and six to seven striae. According to molecular phylogenetic analyses, *N. axillaris* and *N. translucens* are different species (Sakayama *et al.* 2004). The material presently studied was identified as *N. axillaris* by the presence of 1-furcate branchlets and two to four abbreviated 2-celled dactyls, with a conical terminal cell, as described by Sakayama *et al.* (2004). *Nitella acuminata* is a cosmopolitan species occurring in North and South America, Asia and Africa (Wood & Imahori 1965).

***Nitella blankinshipii*** T.F. Allen, Char. America 2(1): 5, 1892.

(Figs. 3 H-K)

Plants dioecious. Axes: 417-520 µm diam., without incrustation. Internodes: 2-2.5 cm long. Branchlets: dimorphic, furcated. Sterile branchlets: 6, 1.2-1.4 cm long × 250 µm diam., 1-furcate, primary ray 0.5 cm long × 290 µm diam., 0.2-2 times the branchlets. Dactyls 2-4, 1-celled, acuminate, 7-9 mm long × 170-208 µm diam. Fertile branchlets: 6, 2-6 mm long × 167-271 µm diam., 1-furcate. Dactyls 2-4, 1-celled, 1.5-2.8 mm long × 125-166 µm diam. Heads: well formed, lax. Oogonia: 1-(2) at a node, 562-708 µm long × 516-583 µm diam., corona deciduous, 42-45 µm long × 67-75 µm diam., convolutions 8-10. Oospores: not observed. Antheridia: not observed.

**Specimens examined:** BRAZIL, SANTA CATARINA, Lajes, 22.vii.1980, R.M.T. Bicudo (SP162066).

**Distribution in Brazil:** São Paulo (Picelli-Vicentim *et al.* 2004); Santa Catarina (This study).

**Comments:** This is the first citation of this species in Santa Catarina. Analysis of our material increased the lower metric limit of the stalk diameter from 465.5-417 µm, the upper metric limit of the sterile branchlet dactyl length from 8-9 mm, and the lower metric limit of the fertile branchlet dactyl length from 1.5-2 mm, compared with measurements provided by Picelli-Vicentim *et al.* (2004). *Nitella blankinshipii* is a species occurring in North and South America (Wood & Imahori 1965).

***Nitella clavata*** Kützing, Syst. Alg. 518. 1849.

(Figs. 4 A-H)

Plants monoecious. Stem: 11-30 cm tall. Axes: 687-1900 µm diam., without incrustation. Internodes: 1-8.5 cm long. Branchlets: heterocleous. Sterile branchlets: 1-furcate; 0.8-4 cm long × 800-1450 µm diam., primary ray 4-7, nearly the branchlet length. Dactyls: 2-5, 1-celled, inflated, acuminate to mucronate, 0.1-1 cm long × 900-1590 µm diam. Accessory branchlets: simple, inflated, 5-9, 0.3-1.3 cm long × up to 1 mm diam., between each normal ones, acuminate to mucronate. Fertile branchlets: forming heads, 6-8 per whorl, 1-furcate, 0.5-6.8 mm long × 140-200 µm diam.; dactyls 0.5-3 mm long, acuminate or mucronate. Heads: compact, dense, numerous, axillary or terminal, 1-15 mm long × 1-9 mm diam. Gametangia: short-stipitate, conjoined or sejoned, at the nodes of fertile branchlets, absent on accessory branchlets, stalk 50-116 µm long. Oogonia: 1-2(-3) at a node, 420-600 µm long × 358-500 µm diam., convolutions 7-10, corona 50-92 µm long × 65-92 µm diam. Oospore: 340-380 µm long × 320-370 µm diam., fossa 30-75 µm across, striae of 7-8 ridges. Antheridia: 420-625 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Jaguarão, 21.x.2000, J.F. Prado (ICN91565); Mostardas, ix.2009, A.S. Rolon (UNOP-Algae2883); Pedras Altas, 22.x.2000, J.F. Prado (ICN91566); Piratini, 20.xi.1999, J.F. Prado (ICN91567); Rio Grande, 20.x.2000, J.F. Prado (ICN91568); Santa Vitória do Palmar, 14.vii.1980, R.M.T. Bicudo (SP162048), 14.x.2002, J.F. Prado (ICN91570); Tavares, viii.2008, A.S. Rolon (UNOP-Algae2881, UNOP-Algae2882); Uruguaiana, 13.xi.1999, J.F. Prado (ICN91569).

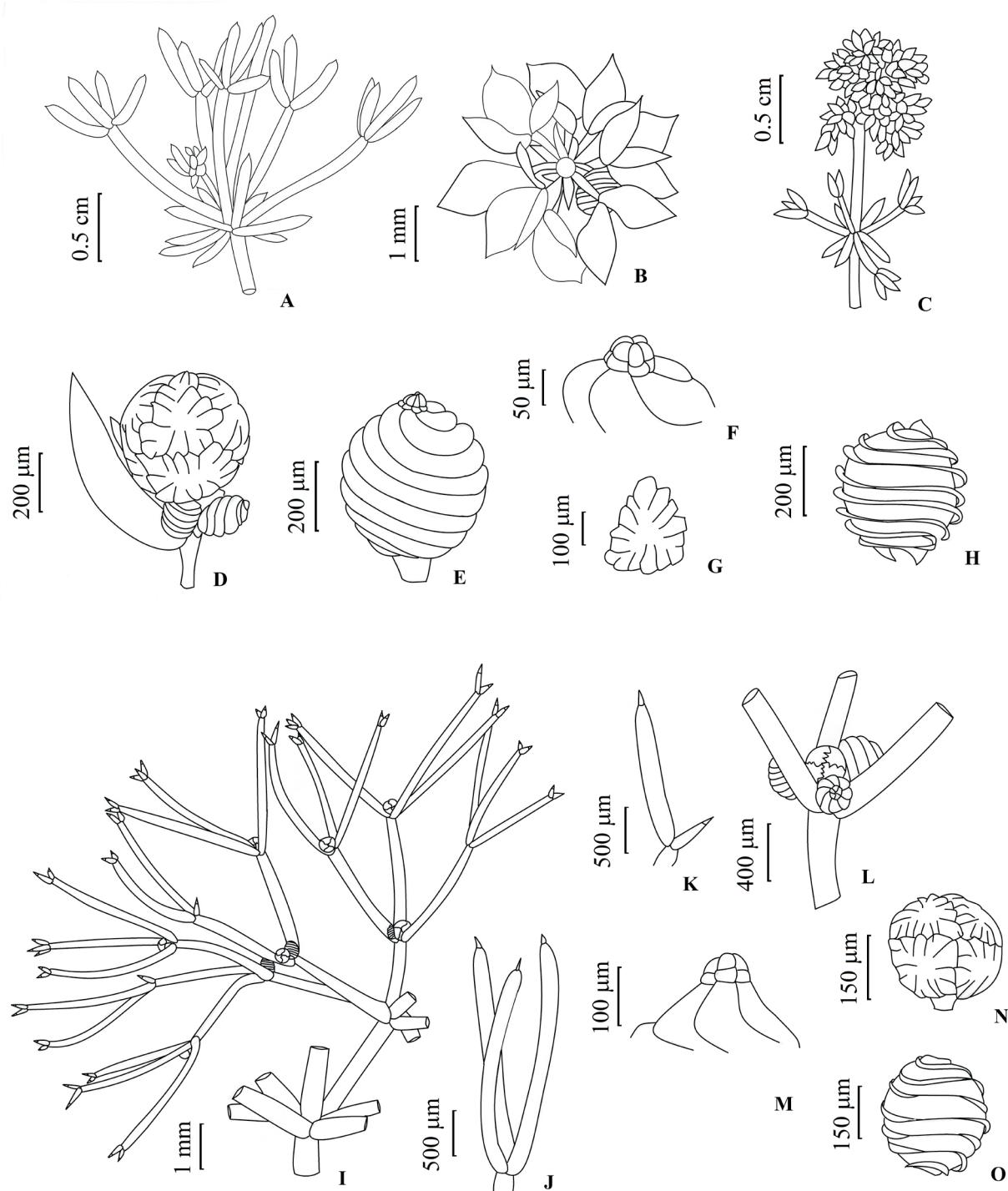
**Distribution in Brazil:** Espírito Santo (Kützing 1849, Bicudo & Yamaoka, 1978, Araújo *et al.* 2010); Rio Grande do Sul (Astorino 1983, Torgan *et al.* 2001, Rolon *et al.* 2011, This study).

**Comments:** *Nitella clavata* was proposed by Kützing (1849) based on material collected in "Vittoria" (Wood & Imahori 1965), probably in Santa Vitória do Palmar. *Nitella clavata* occurs in South America (Wood & Imahori 1965).

***Nitella flagellifera*** J. Groves & G.O. Allen, Jour. Bot. 65: 337. 1927.

(Figs. 4 I-O)

Plants monoecious. Stem: 10-26 cm tall. Axes: 325-830 µm diam., without incrustation. Internodes: 0.5-3.5 cm long. Branchlets: monomorphic, 7-10(-12), 2-4-furcate, 1.1-2.3 cm long, primary ray 6-8, 0.4-0.5 cm long × 270-312 µm diam., secondary ray 4-6, of which one may be central and of different size, sometimes replaced by an



**Figs. 4 A-O.** A-H. *Nitella clavata*. A. Sterile whorl with developing head; B. Fertile branchlet apical view; C. Fertile whorl, head; D. Fertile node; E. Oogonium; F. Coronula; G. Triangular scute; H. Oospore. I-O *Nitella flagellifera*. I. Fertile whorl with three to four-furcated branchlets; J-K. One-celled dactyls; L. Fertile node; M. Coronula; N. Antheridium; O. Oospore.

antheridia, tertiary ray 3-5, quaternary ray 3-5. Dactyls: (2)-3-5, 2-celled, 290-3700 µm long × 60-209 µm diam., usually elongate, basal cell gradually tapering distally, end cell conical to acute. Heads: occasionally formed. Gametangia: conjoined or sejoned, at the 2<sup>nd</sup> and 3<sup>rd</sup> lowest

nodes. Oogonia: 1-2(-3), 325-650 µm long × 258-500 µm diam., convolutions 7-8, coronula 45-60 µm long × 70-85 µm diam. Oospores: 320-350 µm long × 260-310 µm diam., striae of 6 ridges, fossa 30-80 µm across. Antheridia: 187-383 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, estrada Curitiba-Joinville, km 42, 26.viii.1966, C.E.M. Bicudo (SP96636). Santa Catarina, Laguna, 17.x.1979, R.M.T. Bicudo (SP155079). RIO GRANDE DO SUL, Butiá, 21.ii.2002, J.F. Prado (ICN91571); Dom Pedro de Alcântara, 23.vi.2000, J.F. Prado (ICN91572); Itaara, 18.xii.1999, J.F. Prado (ICN91573); São Borja, 03.iv.2001, J.F. Prado (ICN91574); São Francisco de Assis, 20.xii.2000, J.F. Prado (ICN91575); Vale Verde, 21.ii.2002, J.F. Prado (ICN91576).

**Distribution in Brazil:** *Nitella flagellifera* [= *N. furcata* (Roxburgh ex Bruxelius) C. Agardh emend. R.D. Wood subsp. *flagellifera* (J. Groves & G.O. Allen) R.D. Wood]. São Paulo (Vieira-Júnior & Necchi-Júnior 2002; Picelli-Vicentim *et al.* 2004; Bicudo & Bueno 2011); Mato Grosso do Sul (Bueno & Bicudo 1997); Paraná (This study as *Nitella flagellifera*); Santa Catarina (This study as *Nitella flagellifera*); Rio Grande do Sul (Araújo *et al.* 2010, This study as *Nitella flagellifera*).

**Comments:** This is the first citation this species for the states of Paraná and Santa Catarina. The upper metric limits of the stalk diameter were extended from 814 µm to 830 µm and those of the oogonia from 500 µm long × 431 µm in diameter to 650 µm × 500 µm, compared to data in Picelli-Vicentim *et al.* (2004). The upper metric limits of the fossa width were extended from 70 µm to 80 µm and that of the antheridia diameter from 350 µm to 358 µm compared to data provided by Vieira-Júnior & Necchi-Júnior (2002). *Nitella flagellifera* is a species occurring in South America and Asia (Wood & Imahori 1965).

***Nitella flexilis* (L.) C. Agardh, Syst. Alg. 124. 1824.**

(Figs. 5 A-E)

Plants monoecious. Stem: 15 cm tall. Axes: 521-583 µm diam., without incrustation. Internodes: 2-4 cm long. Branchlets: monomorphic, 1.7-3.5 cm long, 1-furcate, primary ray 7, 0.9-1.4 cm long. Dactyls: 2, 1-celled, generally elongate, acute, 0.4-8.8 mm long × 250-354 µm diam. Heads: absent. Gametangia: conjoined or sejoned. Oogonia: 1-2 at a node, 433-750 µm long × 374-560 µm diam., convolutions 8, coronula deciduous, 42 µm long × 58 µm diam. Oospores: 525 µm long × 383 µm diam., striae of 7 ridges, fossa 77 µm across. Antheridia: 283-333 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Mostardas, 08.viii.2009, A.S. Rolon (UNOP-Algae2884).

**Distribution in Brazil:** Pernambuco (Bicudo & Yamaoka 1978, Araújo *et al.* 2010); Rio Grande do Sul (This study).

**Comments:** This is the first citation of this species for Rio Grande do Sul. *Nitella flexilis*, due to frequent protandry, is commonly identified as *N. opaca* (C. Agardh ex Bruxelius) C. Agardh (dioecious). In our material, the upper metric limits of the axis diameter were increased from 560 µm (Bicudo & Yamaoka 1978) to 583 µm (UNOP-Algae2884).

The length and the diameter limits of dactyls were increased from 2.4-3.2 mm and 256-320 µm (Bicudo & Yamaoka, 1978) to 0.4-8.8 mm and 250-354 µm, respectively. *Nitella flexilis* is a cosmopolitan species occurring in North and South America, Africa, Asia and Europe (Schubert & Blindow 2004).

***Nitella furcata* (Roxburgh ex Bruxileus) C. Agardh, Syst. Alg. 124. 1824.**

(Figs. 5 F-N)

Plants monoecious. Stem: 10-22.5 cm tall. Axes: 408-985 µm diam., without incrustation. Internodes: 0.5-4.3 cm long. Branchlets: monomorphic, 5-8, 1.1-2 cm long × 310-560 µm diam., 2-3(-4)-furcate; primary ray 6, 0.35-0.6 cm long × 310-560 µm diam., secondary ray 4-5, tertiary 3-4, quaternary 2-3. Dactyls: 2-3(-4), forming a crown, 2-celled, end cell conical, acuminate or acute, usually abbreviated, 140-390(-1260) µm long × 50-100 µm diam. Heads: absent. Gametangia: conjoined or sejoned, sometimes short-stipitate. Oogonia: (1-)2-4-(6) at a node, 375-590 µm long × 325-396 µm diam., convolutions 6-8, coronula convergent or divergent, 58-90 µm long × 52-98 µm diam. Oospores: 260-317 µm long × 215-275 µm diam., striae 6-7, fossa 30-70 µm across, membrane reticulate. Antheridia: 250-310 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Chuí, 16.x.2002, J.F. Prado (ICN91577); Santa Vitória do Palmar, 16.x.2002, J.F. Prado (ICN91578, 91602); Santiago, 28.xi.2000, J.F. Prado (ICN91579, SP127680); Pelotas, 17.xi.1978, col.? (SP152634); Mostarda, Lagoa da Figueira, 30.xii.1978, col.? (SP154998).

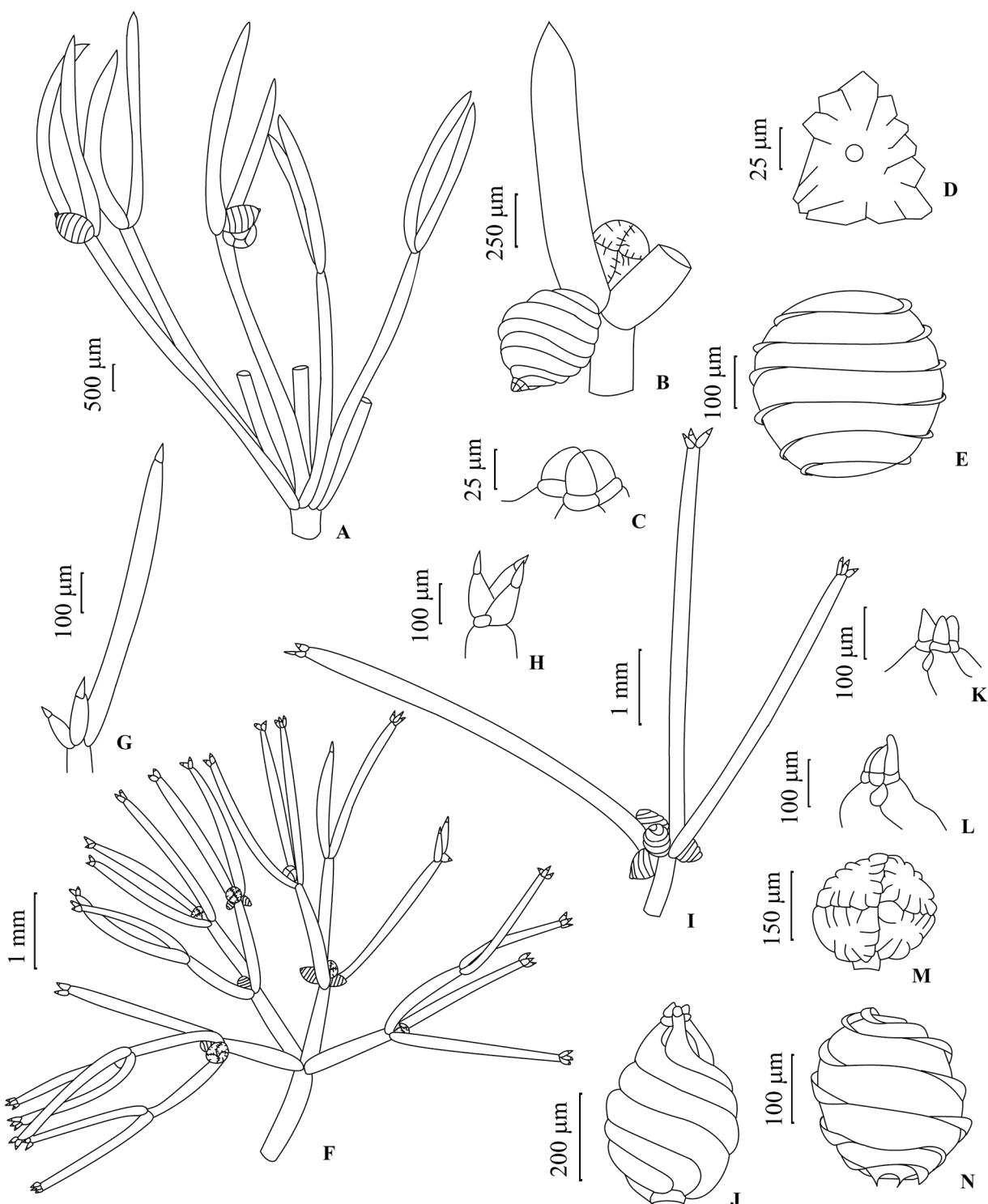
**Distribution in Brazil:** locality not specified (Guerlersquin 1981, Araújo *et al.* 2010); Paraná (Meurer & Bueno 2012); Rio Grande do Sul (Rolon *et al.* 2011, This study).

**Comments:** *Nitella furcata* is a cosmopolitan species occurring in Africa, Asia and South America (Wood & Imahori 1965, Wood 1978).

***Nitella gollmeriana* A. Braun, Monats. der Deutschen Akad. der Wiss. zu Berlin 1858: 355. 1859.**

(Figs. 6 A-E)

Plants monoecious. Stem: 14-28 cm tall. Axes: 790-860 µm diam., without incrustation. Internodes: 2-5.5 cm long. Branchlets: dimorphic, furcate. Sterile branchlets: 6-9, 1-furcate, 0.8-3.65 cm long × 200-800 µm diam., primary ray 6-9, nearly as long as the branchlet, 0.6-2.6 cm long. Dactyls: 3-4, 1-celled, acuminate, 1-6 mm long × 170-400 µm diam. Fertile branchlets: 6-9, 1-furcate, 970-1050 µm long × 150-170 µm diam., reduced into fertile heads. Dactyls: 2-3, 1-celled, abbreviated, acuminate, 540-800 µm long × 120-210 µm diam. Heads: dense, axillary, stipitate, numerous, 1-3 per whorl, without mucus, semi-spherical to

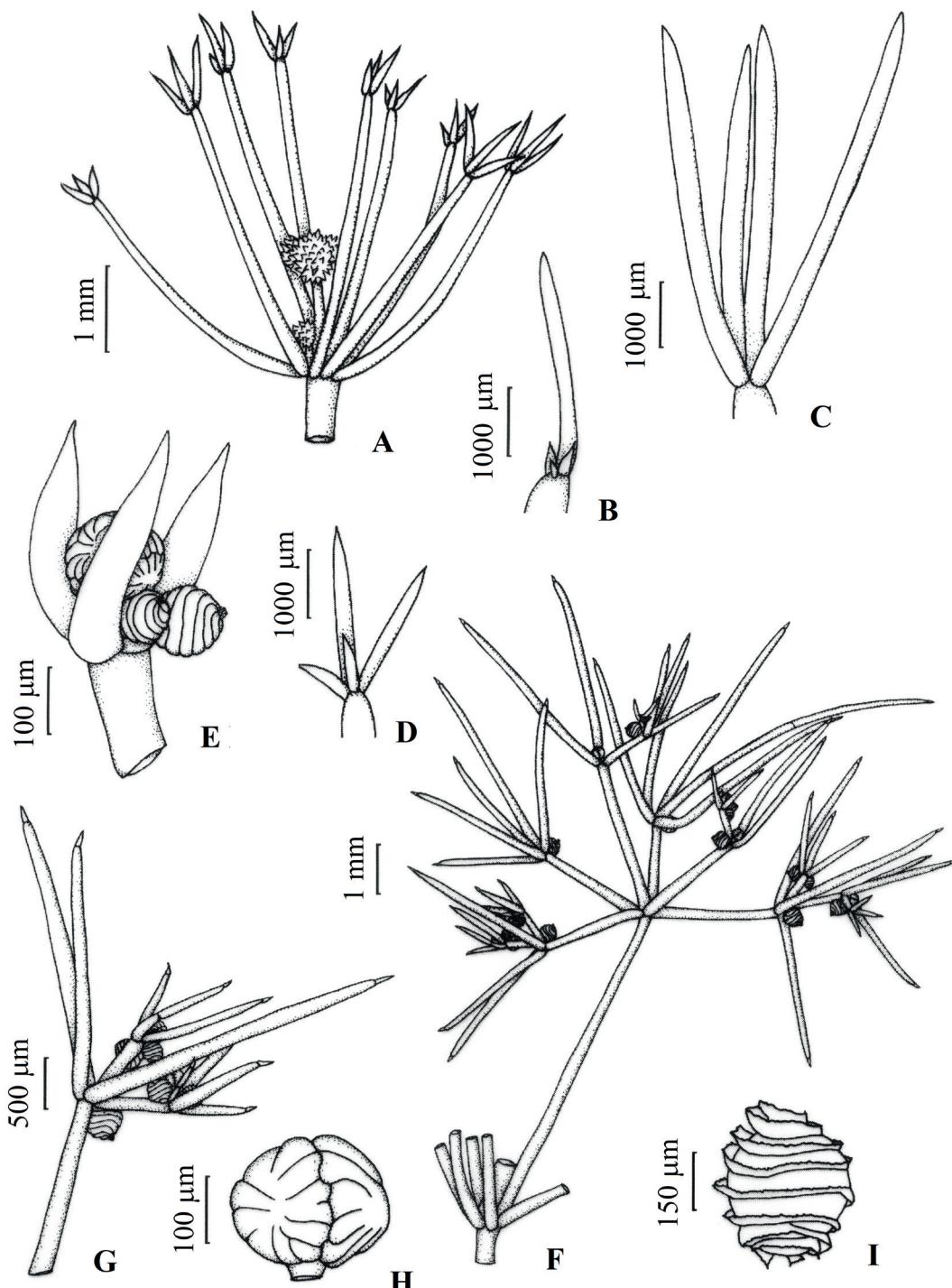


**Figs. 5 A-N.** A-E. *Nitella flexilis*. A. Sterile and fertile branchlets; B. Fertile node; C. Coronula; D. Triangular Scute; E. Oospore. F-N *Nitella furcata*. F. Whorl with three and four-furcated branchlets; G.-H. Dactyls; I. Fertile node; J. Oogonium; K-L. Coronula; M. Antheridia with eight scutes; N. Oospore.

spherical, 2-3 mm diam. Gametangia: conjoined, sessile, sometimes in the heads. Oogonia: 1-2 at a node, 210-375.5  $\mu\text{m}$  long  $\times$  150-200  $\mu\text{m}$  diam., convolutions 7-8, coronula 35-40  $\mu\text{m}$  long  $\times$  45-50  $\mu\text{m}$  diam. Oospores:

not observed. Antheridia: 260-300  $\mu\text{m}$  diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Horizontina, 16.xi.2000, J.F. Prado (ICN91555).



**Figs. 6 A-I.** A-E. *Nitella gollmeriana*. A. Whorl with sterile and fertile branchlets; B-D. One-celled dactyls; E. Fertile node. F-I *Nitella gracilis*. F. Whorl with two to four-furcated fertile branchlets; G. Fertile branchlet; H. Antheridium with four scutes; I. Oospore.

**Distribution in Brazil:** São Paulo (Bicudo 1969, Bicudo & Yamaoka 1978, Branco & Necchi-Júnior 1996, Necchi-Júnior *et al.* 2000, Picelli-Vicentim *et al.* 2004); Mato Grosso do Sul (Bueno & Bicudo 1997); Rio Grande do Sul, (Araújo *et al.* 2010, This study).

**Comments:** *Nitella gollmeriana* is morphologically similar to *N. acuminata* and *N. subglomerata* A. Braun, from which it differs by having dimorphic branchlets and fertile heads, varying from compact to dense (Bicudo & Yamaoka 1978, Picelli-Vicentim *et al.* 2004). *Nitella gollmeriana* is a species restricted to South America (Wood & Imahori 1965).

*Nitella gracilis* (Smith) C. Agardh, Syst. Alg. 125. 1824.

(Figs. 6 F-I)

Plants monoecious. Stem: 9-16 cm tall. Axes: 290-360  $\mu\text{m}$  diam., slender. Internodes: 0.5-5 cm long. Branchlets furcated, without incrustation. Sterile branchlets: 5-6 per whorl, 0.7-2.2 cm long  $\times$  200-270  $\mu\text{m}$  diam., 2-3-furcate, primary ray 5-6, secondary ray 5-6, one of which may be central, tertiary 2-4, quaternary 2-3. Dactyls 2-5, 2-3-celled, 600-6000  $\mu\text{m}$  long  $\times$  70-260  $\mu\text{m}$  diam., usually elongated, end cell conical to acute. Fertile branchlets: 5-6, 2-3-(4)-furcate, primary ray 5-6, secondary ray 5-6, tertiary ray 3-5, quaternary ray 1-3, of which 1 or 2 may 2-furcate into 2-3 quinary rays. Dactyls: 2-5, 2-3-celled. Heads: not formed. Gametangia: short-stipitate, conjoined or sejoined, at the base of 2<sup>nd</sup>-4<sup>th</sup> furcation. Oogonia: 1-2 at a node, 390-510  $\mu\text{m}$  long  $\times$  300-410  $\mu\text{m}$  diam., convolutions 7-8, corona persistent, 35-40  $\mu\text{m}$  long  $\times$  50-60  $\mu\text{m}$  diam. Oospores: light to dark brown, 280-330  $\mu\text{m}$  long  $\times$  240-330  $\mu\text{m}$  diam., striae of 6 ridges, fossa 40-70  $\mu\text{m}$  across, membrane granulate. Antheridia: 150-270  $\mu\text{m}$  diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Casca, 02.vi.2000, J.F. Prado (ICN91588); São Francisco de Paula, 11.iv.2000, J.F. Prado (ICN91589).

**Distribution in Brazil:** Paraná (Meurer & Bueno 2012); Rio Grande do Sul (Astorino 1983, Araújo *et al.* 2010, This study).

**Comments:** *Nitella gracilis* is a cosmopolitan species found in Europe, North and South America, but rare in southern Africa (Corillion 1975, Wood & Imahori 1965, Wood 1975).

*Nitella havaiensis* Nordstedt, Minneskr. Fys. Sällsk. Lund 7: 1-24. 1878.

(Figs. 7 A-H)

Plants monoecious. Stem: 10 cm tall. Axes: 248-500  $\mu\text{m}$  diam. Internodes: 0.3-2.8 cm long. Branchlets: dimorphic. Sterile branchlets: 6, (1)-(2)-(3)-furcate, primary ray 6, 2/4 times the length of whorl branchlet, secondary ray (2)-(4)-(5), tertiary ray (2)-5. Fertile branchlet: 6, usually forming dense heads at the terminal portion of the plant. Dactyls: 4, 2-4-celled, acuminate, normal to abbreviated, usually elongated. Heads: fertile branchlets reduced, without mucus. Gametangia: absent at the basal node of branchlets. Oogonia: 3 at a node, 263-334  $\mu\text{m}$  long  $\times$  251-304  $\mu\text{m}$  diam., convolutions 8-9, corona 38-52  $\mu\text{m}$  long  $\times$  33-43  $\mu\text{m}$  diam. Oospore: 215-236  $\mu\text{m}$  long  $\times$  195-220  $\mu\text{m}$  diam., striae of 6 ridges, fossa 44-52  $\mu\text{m}$  across, membrane granulate. Antheridia: 134-234  $\mu\text{m}$  diam., occasionally short-stipitate, stalk 66  $\mu\text{m}$  long, octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Santa Cruz do Sul, 18.vii.1980, R.M.T. Bicudo (SP162058); Tramandaí, 11.viii.1980, R.M.T. Bicudo (SP162032).

**Distribution in Brazil:** Rio Grande do Sul (Astorino 1983, Araújo *et al.* 2010, This study).

**Comments:** *Nitella havaiensis* is found in Central and South America (Wood & Imahori 1965).

*Nitella hyalina* (DC.) C. Agardh, Syst. Alg. 126. 1824.

(Figs. 7 I-O)

Plants monoecious. Stem: 2.5-4.7 cm long. Axes: 250-646  $\mu\text{m}$  diam., without incrustation, delicate. Internodes: 0.3-2.5 cm long, up to 9 times the branchlets length. Branchlets: heterocleous, furcated. Sterile branchlets: 5-8, 2-furcate, 1-9 mm long, primary ray 5-8, 1-4.3 mm long  $\times$  283-325  $\mu\text{m}$  diam., secondary ray 3-6, tertiary ray 3, usually elongate. Dactyls: 3, 2-celled, 550-2708  $\mu\text{m}$  long  $\times$  150-250  $\mu\text{m}$  diam. Accessory branchlets: in 2 rows, 4-6, 0-1-furcate,  $\frac{1}{2}$  the length of primary ray. Heads compact, 0.2-0.6 cm long. Gametangia: sejoined, short-stipitate, involved with mucus. Oogonia: 1 at a node, 233-680  $\mu\text{m}$  long  $\times$  150-450  $\mu\text{m}$  diam., convolutions 7-8, corona 49-51  $\mu\text{m}$  long  $\times$  45-80  $\mu\text{m}$  diam. Oospore: 275-400  $\mu\text{m}$  long  $\times$  284-340  $\mu\text{m}$  diam.; striae of 6-7 ridges, fossa 51-70  $\mu\text{m}$  across, membrane fibrous. Antheridia: 190-380  $\mu\text{m}$  diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Guaíra, 06.x.1978, R.M.T. Bicudo (SP152549). RIO GRANDE DO SUL, Santa Vitória do Palmar, 14.xii.1999, J.F. Prado (ICN91592).

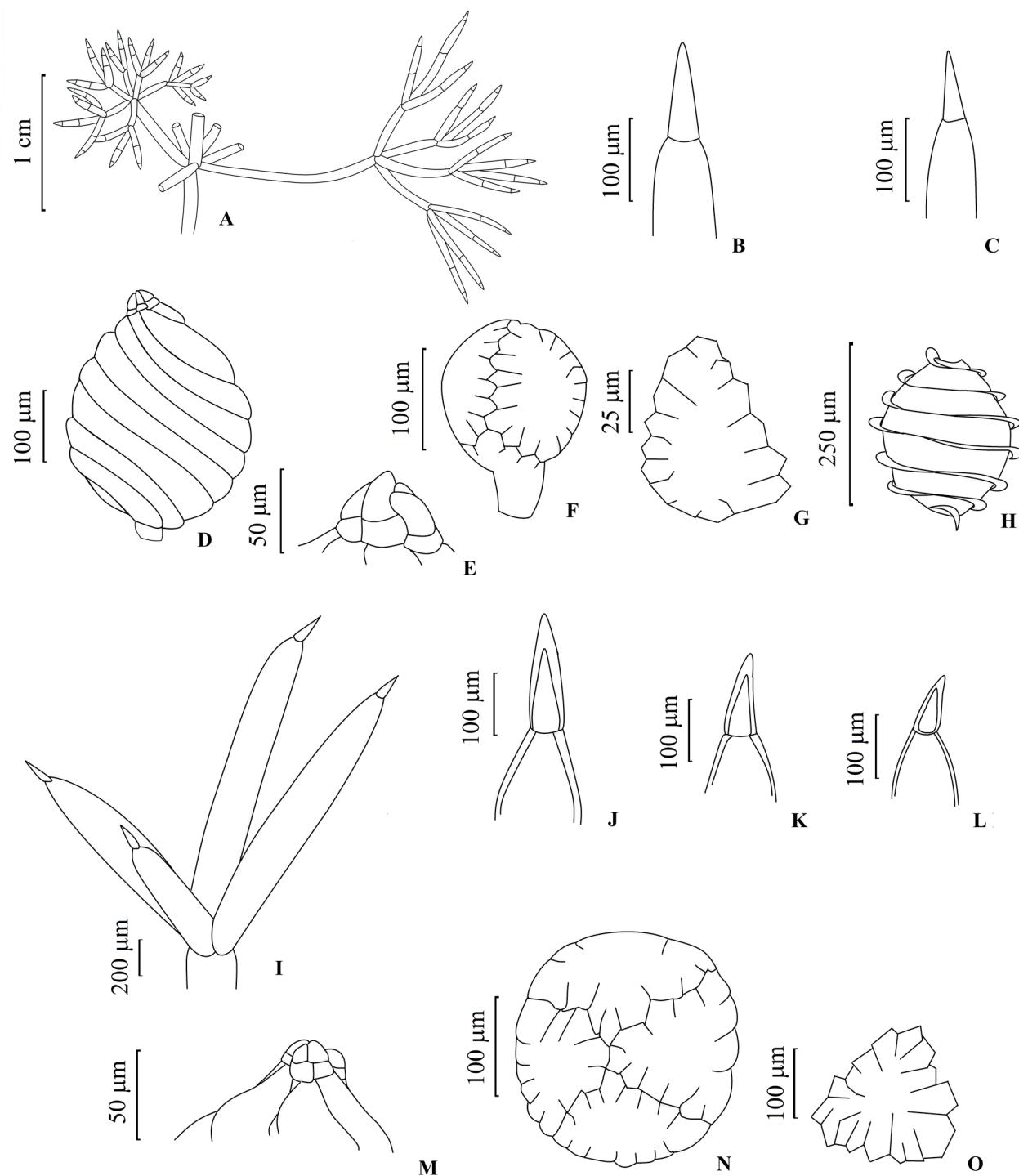
**Distribution in Brazil:** Paraná (Meurer & Bueno 2012, Present study); Rio Grande do Sul (Prado & Baptista 2005, Araújo *et al.* 2010, This study).

**Comments:** The lower limit of oogonia length differed from 300  $\mu\text{m}$  reported by Prado & Baptista (2005) to 233  $\mu\text{m}$  in our material, as well as the lower limit of the oospore length from 370  $\mu\text{m}$  to 275  $\mu\text{m}$ . *Nitella hyalina* is a cosmopolitan species, but not abundant, occurring in North and South America, southwestern and southern Africa and Asia (Corillion 1957, Wood & Imahori 1965, Wood 1978, Moore 1986, Krause 1997, Schubert & Blindow 2004).

*Nitella inversa* Imahori, Japanese Char. 125, pl. 31, fig. 44. 1954.

(Figs. 8 A-K)

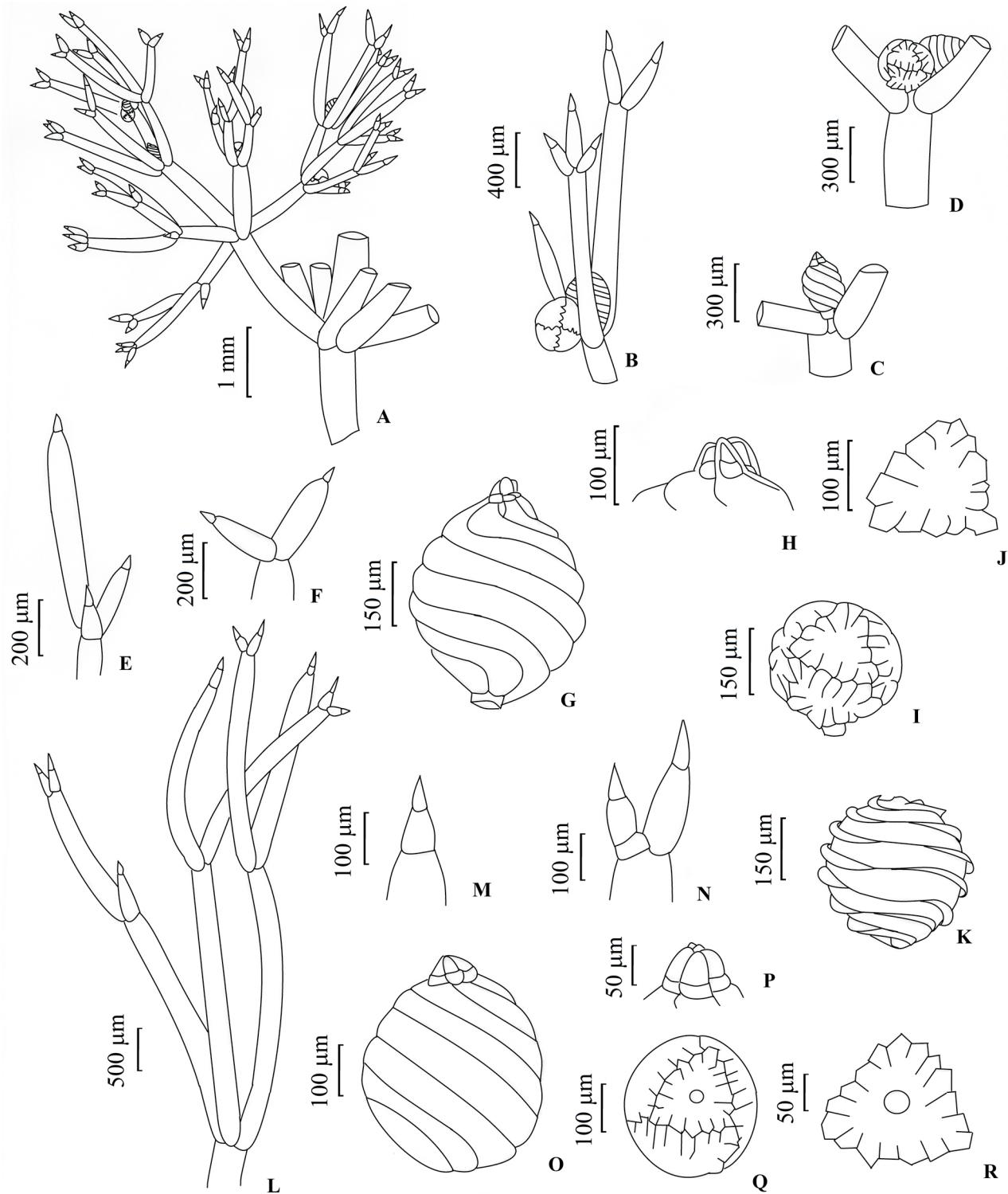
Plants monoecious. Stem: 10-30 cm tall. Axes: 390-900  $\mu\text{m}$  diam., without incrustation. Internodes: 0.8-4.7 cm long. Branchlets: monomorphic, 1.1-2.8 cm long  $\times$  240-470  $\mu\text{m}$  diam., 3-4-furcate, primary ray 5-7, 0.5-1.1



**Figs. 7 A-O.** A-H. *Nitella havaiensis*. A. Whorl; B-C. Dactyls apices; D. Oogonium; E. Coronula; F. Antheridium; G. Scute; H. Oospore. I-O *Nitella hyalina*. I. Two-celled dactyls; J-L. Dactyls apices; M. Coronula; N. Antheridium; O. Scute.

cm long  $\times$  240-470 µm diam., secondary ray 4-6, tertiary ray 3-4, quaternary ray 2-3. Dactyls: 2-3, 2-3(-4)-celled, 70-2950 µm long  $\times$  35-180 µm diam., usually abbreviated, end cell conical to acute, 79-100 µm long  $\times$  35-42 µm diam. Heads: not formed. Gametangia: conjoined or sejoined, at the base of all branchlet's furcation, sometimes short-

stipitate. Oogonia: 1-2 at a node, terminal, 320-590 µm long  $\times$  260-450 µm diam., convolutions 7-8, coronula 50-65 µm long  $\times$  75-80 µm diam. Oospore: 320-345 µm long  $\times$  290-315 µm diam., fossa 30-70 µm across, membrane reticulate. Antheridia: terminal or lateral, 270-310 µm diam., octoscutate, shields triangular.



**Figs. 8 A-R.** A-K. *Nitella inversa*. A. Whorl; B. Fertile branchlet; C-D. Fertile nodes; E-F. Dactyls; G. Oogonium; H. Coronula; I. Antheridium; J. Scute; K. Oospore. L-R *Nitella microcarpa*. L. Whorl; M-N. Dactyls; O. Oogonium; P. Coronula; Q. Octo-scute Antheridium with eight scutes; R. Triangular scute.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Itaqui, 04.iv.2001, J.F. Prado (ICN91580); Mostardas, 05.xii.1999, J.F. Prado (ICN91581); São Borja, 03.iv.2001, J.F. Prado (ICN91582, ICN91583); São Francisco de Assis, 20.xii.2000, J.F. Prado (ICN91584); Vila Nova do Sul, 19.iii.2001, J.F. Prado (ICN91585).

**Distribution in Brazil:** São Paulo (Picelli-Vicentim *et al.* 2004, Bicudo & Bueno 2011); Paraná (Meurer & Bueno 2012); Rio Grande do Sul (This study).

**Comments:** The most important metric variation was detected in the diameter of the coronula that varied from 90  $\mu\text{m}$  in Picelli-Vicentim *et al.* (2004) to 75-80  $\mu\text{m}$ . *Nitella inversa* occurs in Asia and South America (Wood & Imahori 1965).

*Nitella microcarpa* A. Braun, Monat. der Deutschen Akad. der Wiss. zu Berlin 1858: 357. 1858.

(Figs. 8 L-R)

Plants monoecious. Stem: 10 cm tall. Axes: 416-855  $\mu\text{m}$  diam., without incrustation. Internodes: 0.7-2.1 cm long. Branchlets: monomorphic, 7-8, 2-3-furcated, 0.6-2.3 cm long, primary ray 7-8, 4.4-6.7  $\mu\text{m}$  long  $\times$  333-354  $\mu\text{m}$  diam., secondary ray 4-5, 1 of which may be central and reduced, tertiary ray 3-4, quaternary ray 2-3. Dactyls: 2-3, 2-3-celled, 186-1978  $\mu\text{m}$  long  $\times$  62-146  $\mu\text{m}$  diam., usually abbreviated, end cell conical to acute. Heads: not formed. Gametangia: conjoined, at the base of all branchlet's furcation. Oogonia: 1(-2) at a node, 416-458  $\mu\text{m}$  long  $\times$  354-375  $\mu\text{m}$  diam., convolutions 8, coronula 41-43  $\mu\text{m}$  long  $\times$  50-57  $\mu\text{m}$  diam. Oospore: 383-408  $\mu\text{m}$  long  $\times$  316-341  $\mu\text{m}$  diam., fossa 42-58  $\mu\text{m}$  across, membrane reticulate. Antheridia: 192-250  $\mu\text{m}$  diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Foz do Iguaçu, 21.v.1966, B. Lowy (SP96238). RIO GRANDE DO SUL, Cachoeira do Sul, 17.vii.1980, R.M.T. Bicudo (SP162057); Santa Vitória do Palmar, 11.vi.1980, R.M.T. Bicudo *et al.* (SP162031); Cachoeira, 02.xi.1893, G.A. Malme (SP114568); Tavares, 08.viii.2008, A.S. Rolon (UNOP-Algae2879); 10.x.2009, A.S. Rolon (UNOP-Algae2880).

**Distribution in Brazil:** São Paulo (Necchi-Júnior *et al.* 2000, Vieira-Júnior *et al.* 2002, Picelli-Vicentim *et al.* 2004, Bicudo & Bueno 2011); Mato Grosso do Sul (Bueno & Bicudo, 1997); Paraná (Present study); Rio Grande do Sul, (Araújo *et al.* 2010, This study).

**Comments:** This is the first citation of this species in the state of Rio Grande do Sul. In the present material, the maximum oogonia measurements were extended to 600  $\mu\text{m}$  long and 375  $\mu\text{m}$  in diameter, the oospore limits to 366  $\mu\text{m}$  long and 308  $\mu\text{m}$  in diameter, and the antheridia to 333  $\mu\text{m}$  in diameter (Bueno & Bicudo 1997, Picelli-Vicentim *et al.* 2004). *Nitella microcarpa* occurs in North and South America (Wood & Imahori 1965).

*Nitella mucronata* (A. Braun) Miquel in H.C. Hall, Fl. Belg. Sept. 2: 428. 1840.

(Figs. 9 A-G)

Plants monoecious. Stem: 15 cm tall. Axes: 358-632  $\mu\text{m}$  diam., without incrustation. Internodes: 0.8-1.5 cm long. Branchlets: monomorphic, 5-7, 0.8-1 cm long, 2-3-furcated,

primary ray 5-7, 3.5-5.6  $\mu\text{m}$  long  $\times$  250-270  $\mu\text{m}$  diam., secondary ray 3-5, tertiary ray 2-3. Dactyls: 2-3, 2-celled, 283-1,875  $\mu\text{m}$  long  $\times$  108-125  $\mu\text{m}$  diam., elongated, end cell conical to acute. Heads: not formed, terminal whorls occasionally compact, no mucus. Gametangia: conjoined or sejoned, at the base of all branchlet's furcation. Oogonia: 1(-2) at a node, 416-458  $\mu\text{m}$  long  $\times$  354-375  $\mu\text{m}$  diam., convolutions 8, coronula 41-43  $\mu\text{m}$  long  $\times$  50-57  $\mu\text{m}$  diam. Oospore: 383-408  $\mu\text{m}$  long  $\times$  316-341  $\mu\text{m}$  diam., fossa 42-58  $\mu\text{m}$  across, membrane reticulate. Antheridia: 192-250  $\mu\text{m}$  diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Tijucas do Sul, lago Panagro, 11.ii.1999, M.T. Shirata 3880 (SP371375). RIO GRANDE DO SUL, Pedro Osório, 10.xii.1975, M.A.C. Oliveira (SP127680).

**Distribution in Brazil:** São Paulo (Vieira-Júnior *et al.*, 2002; Picelli-Vicentim *et al.*, 2004; Bicudo & Bueno, 2011); Mato Grosso do Sul (Bueno & Bicudo, 1997); Paraná (Present study); Rio Grande do Sul, (Araújo *et al.* 2010, This study).

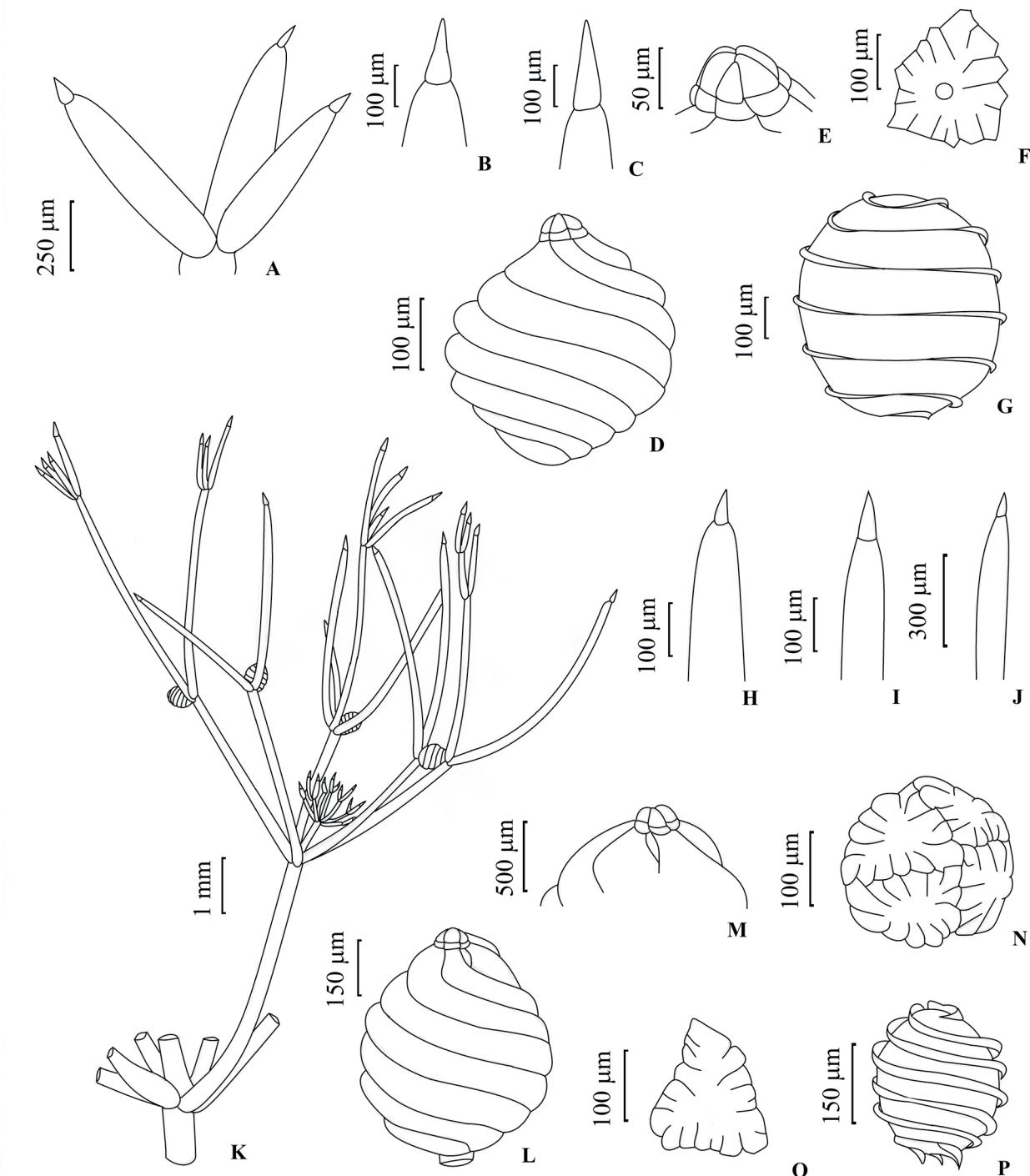
**Comments:** This is the first documentation of this species in the state of Paraná. The study of this material increased the metric limits of the oospores from 300  $\mu\text{m}$  long  $\times$  280  $\mu\text{m}$  in diameter reported by Picelli-Vicentim *et al.* (2004) to 408  $\mu\text{m}$   $\times$  341  $\mu\text{m}$ .

According to Caisová *et al.* (2008), *N. mucronata* is characterized by a mucronate terminal cell, reticulate oospore membrane and an isolated or geminated oogonia. Migula (1897) and Allen (1928) noted that *N. mucronata* is a polymorphic species. According to Urbaniak (2009), *N. mucronata* are plants with fertile branchlets divided 2-3 times to form dense heads, and have 2(-3)-celled dactyls, with mucronate and acute end cells, thus fitting the circumscription of the present material. Recent studies (Casanova 2009, Sakayama 2008) mentioned that the controversial classification proposed by Wood & Imahori (1964, 1965) was based on an incorrect species concept. Therefore, Migula's (1897) classification is adopted, supporting the natural classification of the group. *Nitella mucronata* [= *Nitella furcata* (Roxburgh ex Brzelius) C. Agardh emend. R.D. Wood subsp. *mucronata* (A. Braun) R.D. Wood var. *mucronata* f. *mucronata*] occurs in North, Central and South America, Africa, Asia and Europe (Wood & Imahori 1965, Wood 1978, Schubert & Blindow 2004, Cirujano *et al.* 2007).

*Nitella ogivalis* J. Groves & Stephens, Trans. Roy. Soc. S. Africa 21: 276, pl. 16. 1933.

(Figs. 9 H-P)

Plants monoecious. Stem: 25-45 cm tall. Axes: 250-540  $\mu\text{m}$  diam., without incrustation. Internodes: 1.3-3.2 cm long. Branchlets: monomorphic, 5-6, 2-3-furcated,



**Figs. 9 A-P.** A-G. *Nitella mucronata*. A. Two-celled dactyls; B-C. Dactyls apices; D. Oogonium; E. Coronula; F. Scute; G. Oospore. H-P *Nitella ogivalis*. H-J. Dactyls apices; K. Whorl with two and three-furcated branchlets; L. Oogonium; M. Coronula; N. Antheridium; O. Triangular scute; P. Oospore.

0.5-2.5 cm long  $\times$  180-435 µm diam., primary ray 5-6, 0.4-0.8 cm long  $\times$  200-435 µm diam., secondary ray 3-5(-6), from which 1 may be central, tertiary ray 2-4, quaternary ray 2-4. Dactyls: 2-4, 2-4-celled, 515-3500(-4500) µm long  $\times$  60-85 µm diam., elongated, basal cell tapering distally, end cell conical to acute. Heads: not formed. Gametangia: conjoined or sejoined, at the base of all branchlet's furcation, occasionally absent, no mucus.

Oogonia: 1 at a node, 400-560 µm long  $\times$  350-430 µm diam., convolutions (6)-7-8, corona 35-40 µm long  $\times$  55-65 µm diam. Oospore: light to dark brown, 290-350 µm long  $\times$  240-290 µm diam., striae of 5-7 ridges, fossa 40-65 µm across, membrane reticulate. Antheridia: 180-310 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Mato

Castelhano, 03.vi.2000, J.F. Prado (ICN91586); São Vicente do Sul, 20.xii.2000, J.F. Prado (ICN91587).

**Distribution in Brazil:** São Paulo (Picelli-Vicentim *et al.* 2004); Rio Grande do Sul (Araújo *et al.* 2010, This study).

**Comments:** The oogonia had their size increased from 243-446 µm long to 400-560 µm and antheridia from 296 µm diam. to 310 µm compared to information in Picelli-Vicentim *et al.* (2004). *Nitella ogivalis* is a species occurring in Africa and South America (Wood & Imahori 1965).

*Nitella oligospira* A. Braun, Monat. Deutschen Akad. der Wiss. zu Berlin 1858: 357. 1858.

(Figs. 10 A-F)

Plants monoecious. Stem: up to 25 cm tall. Axes: 416-1078 µm diam. Internodes: 29-69 µm long. Branchlets: dimorphic, furcated. Sterile branchlets: up to 1 cm long, 3-4-furcate; primary ray 7, 0.2-1 cm long, secondary ray (3)-5, 1 of which may be central, tertiary ray 2-3, quaternary ray 2-3(-5), quinary ray 2-3(-4). Dactyls (2)-3-5, 2-celled, confluent, persistent. Fertile branchlets: 6-7, 3(4)-furcate, dactyls 3-5, 2-celled, 937-1375 µm long × 63-104 µm diam. Heads: not formed. Gametangia: conjoined or sejoned, at all branchlet's furcation. Oogonia: 1-3 at a node, 333-590 µm long × 432-458 µm diam., convolutions: 7(-8), coronula 55-75 µm long × 70-94 µm diam., convergent. Oospore: 274-326 µm long × 235-312 µm diam., striae of 6 ridges, fossa 54-63 µm across, membrane reticulate. Antheridia: 211-321 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Santa Vitória do Palmar, 11.vii.1980, R.M.T. Bicudo (SP162030).

**Distribution in Brazil:** Bahia; Minas Gerais (Wood & Imahori 1965); São Paulo (Picelli-Vicentim *et al.* 2004, Bueno & Bicudo 2006, 2008, 2011); Rio Grande do Sul (Astorino 1983, This study).

**Comments:** *Nitella oligospira* [= *Nitella furcata* (Roxburgh ex Brizelius) C. Agardh emend. R.D. Wood var. *mucronata* (A. Braun) R.D. Wood f. *oligospira* (A. Braun) R.D. Wood] has 2-celled, elongate dactyls and a reticulate oospore membrane. The typical variations within the species are 2-3-furcate branchlets, 2-celled or occasionally 3-celled dactyls with a mucronate end cell and an accessory central ray that is not developed. In addition to these characteristics, the specimens were identified as *N. oligospira* by the size of the internodes and whorl branchlets, number of furcations, number of rays, and the presence of quinary rays, absence of heads, number of striae and type of oospore membrane. *Nitella oligospira* is a species widely distributed in southern and eastern Asia, North and South America, Australia, and in Africa and Madagascar (Wood & Imahori 1965, Wood 1978).

*Nitella opaca* (C. Agardh ex Brizelius) C. Agardh, Syst. Alg. 312. 1824.

(Figs. 10 G-K)

Plants dioecious. Stem: 12-28 cm tall. Axes: 420-896 µm diam., without incrustation. Internodes: 0.2-7 cm long. Branchlets: monomorphic; 0.8-5 cm long, 1-furcated; primary ray 6-10, 5-12 mm long × 250-396 µm diam. Dactyls: 2, 1-celled, usually elongated, acute, 0.2-2.3 cm long × 208-395 µm diam. Heads: not formed. Gametangia: in different plants. Oogonia: 1-2 at a node, 396-690 µm long × 325-580 µm diam., convolutions 6-8, coronula deciduous, 42-60 µm long × (33)-58-80 µm diam. Oospore: 308-370 µm long × 292-380 µm diam., striae of 5-6 ridges, fossa 40-80 µm across, membrane reticulate. Antheridia: 670-900 µm diam., octoscutate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Curitiba, 22.vi.1974, J. Rosa (SP116441). RIO GRANDE DO SUL, Arroio Grande, 21.x.2000, J.F. Prado (ICN91535); Herval, 22.x.2000, J.F. Prado (ICN91536); Jaguariaí, 21.x.2000, J.F. Prado (ICN91537); Pedras Altas, 22.x.2000, J.F. Prado (ICN91538); Rio Grande, 20.x.2000, J.F. Prado (ICN91539); Santa Maria, 15.x.1974, Z. Baldissera (SP116446); Rio Pardo, estância Boa Vista, 10.ix.1979, I. Ungaretti (SP155076); Santa Vitória do Palmar, 15.x.2002, J.F. Prado (ICN91540); Santiago, 20.xii.2000, J.F. Prado (ICN91541).

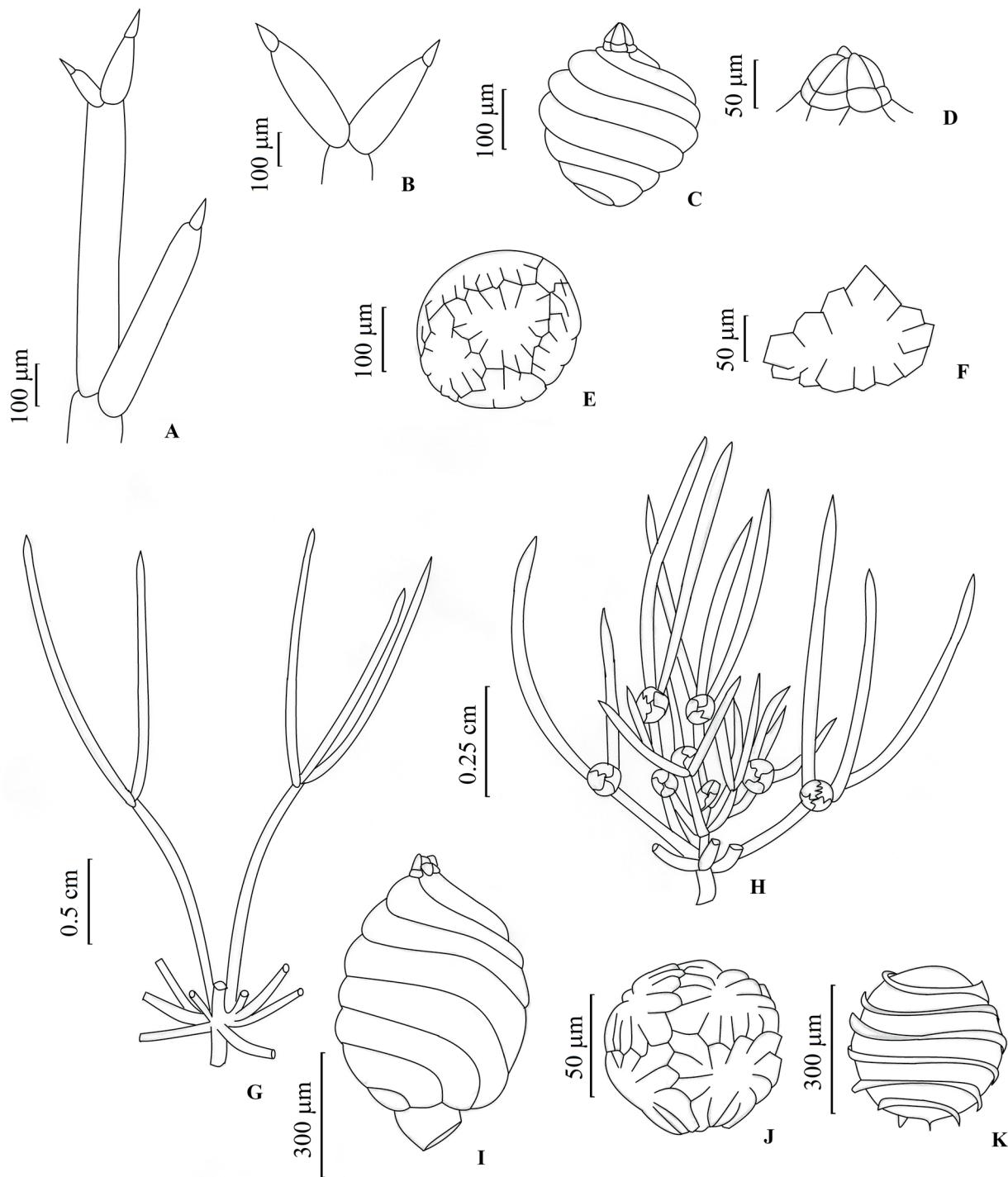
**Distribution in Brazil:** Paraná (Present study); Rio Grande do Sul (Bicudo & Yamaoka 1978, Araújo *et al.* 2010, This study).

**Comments:** *Nitella opaca* is a dioecious species, with 1-furcate branchlets and 1-celled dactyls. When sterile, it is easily mistaken for the monoecious *N. flexilis* (L.) C. Agardh (Bicudo & Yamaoka 1978, Krause 1997, Schubert & Blindow 2004). This is the second record of *Nitella opaca* in Rio Grande do Sul and the first one for Paraná. In Rio Grande do Sul, the samples from Santiago and Santa Vitória do Palmar had only female gametangia, and the ones from the other regions had both male and female. This is the first report of male plants in Brazil. *Nitella opaca* is a cosmopolitan species occurring in North and South America, Africa and Asia. In Europe it is widespread (Corillion 1957, Wood & Imahori 1965, Moore 1986, Krause 1997, Schubert & Blindow 2004).

*Nitella orientalis* T.F. Allen, Bull. Torrey Bot. Club 21(12): 524. 1894.

(Figs. 11 A-G)

Plants monoecious. Stem: 12-17 cm tall. Axes: 563-1043 µm diam., without incrustation. Internodes: 0.7-5.8 cm long, 0.5-1.4 times the branchlets length. Branchlets: dimorphic, furcated. Sterile branchlets: 6, 2-4-furcate, primary ray 6, 0.3-0.5 times the branchlet length, secondary ray (3)-4-5, 1 of which may be central, tertiary ray 3-4, quaternary ray 2-3, quinary ray 2(-3). Dactyls: 2-3, 2-3-celled, usually

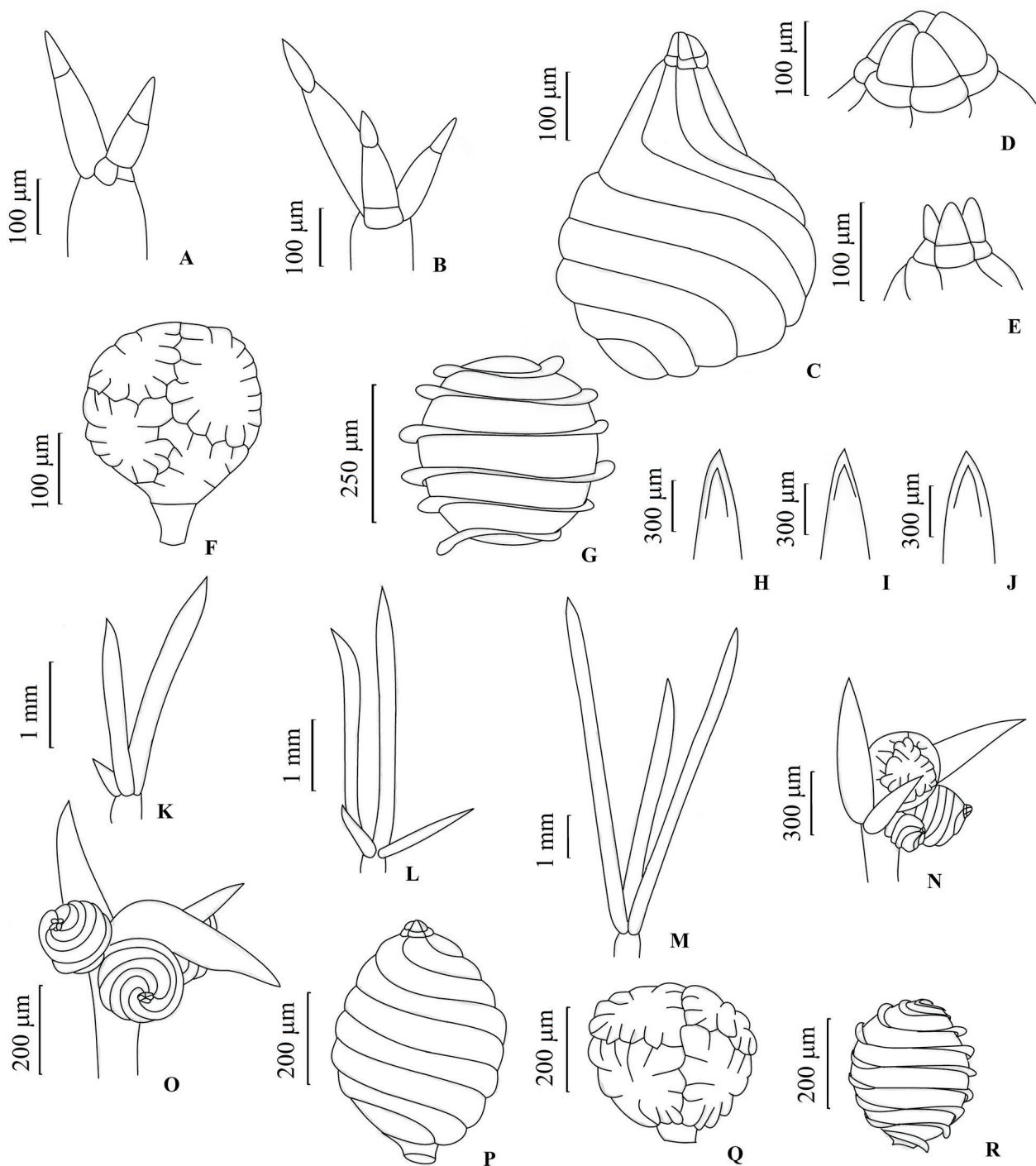


**Figs. 10 A-K.** A-F. *Nitella oligospira*. A-B. Dactyls; C. Oogonium; D. Coronula; E. Antheridium; F. Scute. G-K *Nitella opaca*. G. Sterile branchlets; H. Whorl with fertile branchlets; I. Oogonium; J. Antheridium; K. Oospore.

elongated, mucronate to slightly acuminate, 200-400 µm long. Fertile branchlets: 6, similar to the sterile, but reduced, 3-4-furcate. Heads: not formed. Gametangia: conjoined, at the base of all branchlet's furcation, usually absent at the basal furcation. Oogonia: 1-3 at a node, 314-540 µm long × 272-464 µm diam., convolutions 7-8, coronula persistent, 56-85 µm long × 78-93 µm diam. Oospore: 284-418 µm

long × 232-361 µm diam., striae of 6(-7) ridges, fossa 48-71 µm across, membrane granulate. Antheridia: 248-332 µm diam., tetrascute or octoscute.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Tramandaí, 08.vii.1980, R.M.T. Bicudo (SP162025); São José do Norte, 10.vii.1980, R.M.T. Bicudo (SP162029).



**Figs. 11 A-R.** A-G. *Nitella orientalis*. A-B. Dactyls; C. Oogonium; D-E. Coronula; F. Short-stipitate antheridium; G. Oospore. H-R *Nitella subglomerata*. H-J. Sterile dactyls apices; K-M. Dactyls; N-O. Fertile nodes; P. Oogonium; Q. Antheridia; R. Oospore.

**Distribution in Brazil:** Rio Grande do Sul (Astorino 1983, Araújo *et al.* 2010, This study).

**Comments:** *Nitella orientalis* [= *Nitella furcata* (Roxburgh ex Brzelius) C. Agardh emend. R.D. Wood subsp. *orientalis* (T.F. Allen) var. *orientalis* (T.F. Allen) R.D. Wood f. *orientalis*]. Wood & Imahori (1965) did not mention or

illustrate the number of antheridial scutates. The antheridia ornamentation also varied among sampled populations. The excisicta SP162025 had antheridia with four scutates, while sample unit SP162029 had antheridia with eight scutates. This species occurs in Asia and South America (Wood & Imahori 1965).

**Nitella subglomerata** A. Braun, Monat. der Deutschen Akad. der Wiss. zu Berlin 1858: 356. 1858.

(Figs. 11 H-R)

Plants monoecious. Stem: 10-34 cm tall. Axes: 520-1300  $\mu\text{m}$  diam., without incrustation. Internodes: 1.8-5 cm long. Branchlets: dimorphic, furcated. Sterile branchlets: 6-10, developed, 1-2.4 cm long  $\times$  354-375  $\mu\text{m}$  diam., 1-furcate; primary ray 6-10, 0.8-1 cm long, 0.5 times the branchlets length. Dactyls 2-5, 1-celled, acuminate, 1166-7500  $\mu\text{m}$  long  $\times$  167-360  $\mu\text{m}$  diam. Fertile branchlets: 7-10, forming heads, 0.2-1.7 mm long  $\times$  208  $\mu\text{m}$  diam., 1-furcate. Dactyls 3-4, elongated, curved, acuminate, 1-celled, 550-1730  $\mu\text{m}$  long  $\times$  104-250  $\mu\text{m}$  diam. Heads: stipitate, numerous, 1-3 per whorl, semi-spherical to conical, lax, 0.8-3 mm diam. Gametangia: conjoined or sejoned, at the base of dactyls, sometimes outside the heads. Oogonia: 1-3 at a node, 333-680  $\mu\text{m}$  long  $\times$  283-420  $\mu\text{m}$  diam., convolutions 8, coronula 33-50  $\mu\text{m}$  long  $\times$  50-66  $\mu\text{m}$  diam. Oospore: 250-330  $\mu\text{m}$  long  $\times$  230-371  $\mu\text{m}$  diam., striae of 6 ridges, fossa 55  $\mu\text{m}$  across, membrane granulate. Antheridia: 190-558  $\mu\text{m}$  diam., octosculate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Curitiba, 09.xi.1976, J. Rosa (SP127693). Santa Catarina, Imbituba, 17.x.1979, R.M.T. Bicudo (SP155080). RIO GRANDE DO SUL, Venâncio Aires, 18.vii.1980, R.M.T. Bicudo (SP162059); Bagé, 16.vii.1980, R.M.T. Bicudo (SP162051); Cerro Largo, 15.xi.2000, J.F. Prado (ICN91556); Itaara, 18.xii.1999, J.F. Prado (ICN91557); Muitos Capões, 20.v.2000, J.F. Prado (ICN91558); Porto Lucena, 15.xi.2000, J.F. Prado (ICN91559); Rio Grande, 06.ix.1976, J. Waechter (SP-Algae154994); Santiago, 28.xi.2000, J.F. Prado (ICN91562); São Borja, 03.VI.2001, J.F. Prado (ICN91563); Taquaruçu, 16.xi.2000, J.F. Prado (ICN91560); Torres, 23.vi.2000, J.F. Prado (ICN91561); Unistalda, 28.xi.2000, J.F. Prado (ICN91564).

**Distribution in Brazil:** Minas Gerais (Braun & Nordstedt 1883, Bicudo 1969, Bicudo & Yamaoka 1978); São Paulo (Braun & Nordstedt 1883, Wood & Imahori 1965, Bicudo & Yamaoka 1978, Picelli-Vicentim & Bicudo 1993, Necchi-Júnior *et al.* 2000, Vieira-Júnior *et al.* 2002; Picelli-Vicentim *et al.* 2004, Bicudo & Bueno 2011); Mato Grosso (Braun & Nordstedt 1883, Bicudo & Yamaoka 1978); Mato Grosso do Sul (Bicudo & Yamaoka 1978, Bueno & Bicudo 1997); Paraná, (Araújo *et al.* 2010, Bicudo & Yamaoka 1978, Meurer & Bueno 2012, Present study); Santa Catarina (Present study); Rio Grande do Sul, (Astorino 1983, Torgan *et al.* 2001, Araújo *et al.* 2010, This study).

**Comments:** This is the first record of *N. subglomerata* in Santa Catarina. *Nitella subglomerata* is a cosmopolitan and widely reported species. The species has well developed heads, 1-celled dactyls and dimorphic, 1-furcate branchlets. It is characterized by fertile branchlets reduced into heads, which distinguishes it from *N. acuminata*, and the presence of both male and female gametangia from the dioecious species *N. blankinshipii*. *Nitella subglomerata* can be found in North and South America (Wood & Imahori 1965).

**Nitella tenuissima** (Desvaux) Kützing, Phyc. Gen. 319. 1843.

(Figs. 12 A-H)

Plants monoecious. Stem: 5-11 cm tall. Axes: 160-580  $\mu\text{m}$  diam., without incrustation. Internodes: 0.7-3.7 mm long. Branchlets: monomorphic, 6-7; 2-5.5 mm, 2-3-furcate, primary ray 6-7, secondary ray 5-7, 1 of which may be central, tertiary ray 4-5, quaternary ray 3-6. Dactyls: 3-6, 2-celled, 250-1960  $\mu\text{m}$  long  $\times$  100-250  $\mu\text{m}$  diam. Heads: not formed, terminal whorls occasionally reduced. Gametangia: conjoined or sejoned, at the base of 2<sup>nd</sup>-3<sup>rd</sup> branchlet nodes, absent at the basal node, no mucus. Oogonia: 308-583  $\mu\text{m}$  long  $\times$  230-416  $\mu\text{m}$  diam., coronula 41-62  $\mu\text{m}$  long  $\times$  39-75  $\mu\text{m}$  diam., convolutions 9. Oospore: 214-416  $\mu\text{m}$  long  $\times$  205-396  $\mu\text{m}$  diam., striae of 6-7 ridges, fossa 50-62  $\mu\text{m}$  across, membrane reticulate to papilate. Antheridia: 150-267  $\mu\text{m}$  diam., octosculate, shields triangular.

**Specimens examined:** BRAZIL, PARANÁ, Guaíra, 5.x.1978, R.M.T. Bicudo (SP152547). Santa Catarina, 22.ix.1979, E.J. Paula (SP155077). RIO GRANDE DO SUL, Santa Vitória do Palmar, 12.vii.1980, R.M.T. Bicudo (SP162038); Bagé, 15.vii.1980, R.M.T. Bicudo (SP162053).

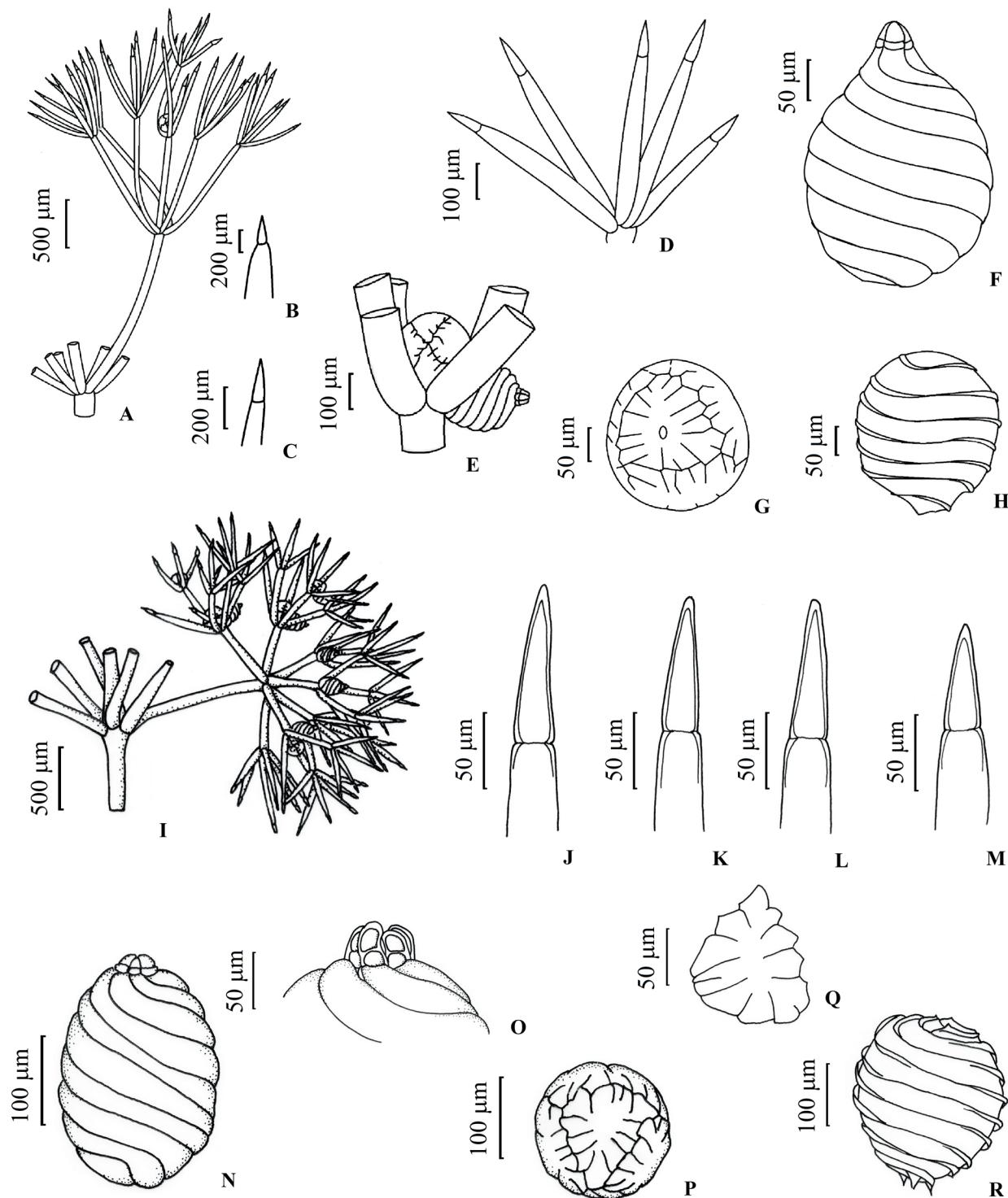
**Distribution in Brazil:** São Paulo (Vieira-Júnior *et al.* 2002, Picelli-Vicentim *et al.* 2004, Bicudo & Bueno 2011, Present study); Rio Grande do Sul (Araújo *et al.* 2010); Santa Catarina and Paraná (This study).

**Comments:** This is the first report of the species for Paraná and Santa Catarina states. The present material was identified as *N. tenuissima* due to the absence of gametangia in the branchlet's basal node, the presence of a secondary central ray, reticulated oospore membrane and 2-celled dactyls (Wood & Imahori 1965). The species is widely recorded in Europe and North and South America (Wood & Imahori 1965, Cirujano *et al.* 2007, Scribailo & Alix 2010).

**Nitella transilis** T.F. Allen, Char. Amer. 2(3): 24, pl. 23. 1896.

(Figs. 12 I-R)

Plants monoecious. Stem: 11-15 cm tall. Axes: 100-540  $\mu\text{m}$  diam., without incrustation. Internodes: 0.5-3 cm long. Branchlets: monomorphic, 6, 0.5-3 cm long  $\times$  150-320  $\mu\text{m}$  diam., 2-3-furcate, primary ray 6, 0.2-1.8 cm long  $\times$  150-320  $\mu\text{m}$  diam., secondary ray 5-7, tertiary ray 3-6, quaternary ray 3-6. Dactyls: 3-6, 2-celled, 570-2500  $\mu\text{m}$  long  $\times$  30-100  $\mu\text{m}$  diam. Heads: not formed. Gametangia: conjoined or sejoned, at the base of 2<sup>nd</sup> and 3<sup>rd</sup> furcations. Oogonia: 1 at a node, 190-470  $\mu\text{m}$  long  $\times$  110-395  $\mu\text{m}$  diam., convolutions 6-8, coronula 40-50  $\mu\text{m}$  long  $\times$  55-65  $\mu\text{m}$  diam. Oospore: 260-300  $\mu\text{m}$  long  $\times$  240-280  $\mu\text{m}$  diam., striae of 7 ridges, fossa 40-45  $\mu\text{m}$  across. Antheridia: 145-340  $\mu\text{m}$  diam., octosculate, shields triangular.



**Figs. 12 A-R.** A-H. *Nitella tenuissima*. A. Sterile and fertile whorl branchlets; B-C. Dactyls apices; D. Dactyls crown; E. Fertile node; F. Oogonium; G. Antheridium; H. Oospore. I-R *Nitella transilis*. I Whorl with two and three-furcated branchlets; J-M. Dactyls apices; N. Oogonium; O. Coronula; P. Antheridia; Q. Triangular Scute; R. Oospore.

**Specimens examined:** BRAZIL, RIO GRANDE DO SUL, Aceguá, 21.iii.2001, J.F. Prado (ICN91590); Mostardas, 04.xii.1999, J.F. Prado (ICN91591).

**Distribution in Brazil:** São Paulo (Picelli-Vicentim *et al.* 2004); Rio Grande do Sul (Astorino 1983, Torgan *et al.* 2001, This study).

**Comments:** *Nitella transilis* is a species found in North and South America (Wood & Imahori 1965).

Information on the occurrence of *Nitella* in the states of Paraná, Santa Catarina and Rio Grande do Sul is still limited. According to the present study, 22 *Nitella* species were collected from the southern region of Brazil (Paraná, Santa Catarina and Rio Grande do Sul) and eight of those are new records for the region. We were able to find new records of *Characeae* for the region, due to the large and diverse amount of material studied. However, studies of the oospore external membrane morphology have proven to be very useful for species identification and intraspecific categories of *Nitella* (John & Moore 1987, Sakayama *et al.* 2002, Urbaniak 2009). The metric variation detected among specimens in different studies is, at least in part, due to intrapopulation variation and/or phenotypic plasticity corresponding to different environmental features in which these plants are found (Asaeda *et al.* 2007, Blindow & Schütte 2007, Urbaniak 2008).

Considering their importance in the aquatic ecosystems, future research is needed and required to provide more realistic data on the diversity, morphology, distribution and ecological requirements of the species as a tool to understand the *Characeae* dynamics in the Brazilian ecosystems.

## ACKNOWLEDGMENTS

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