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Mallomonas multiunca var. *pocosinensis* var. nov. (Synurophyceae) from freshwater localities along coastal sections of North Carolina, U.S.A.

by

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With 2 tables and 6 figures

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Abstract: A new variety of *Mallomonas multiunca*, *M. multiunca* var. *pocosinensis* Siver var. nov., is described from very acidic, humic waterbodies situated on the Atlantic Coastal Plain of North Carolina, U.S.A. The new variety is similar to the type in scale structure, but differs in possessing a unique type of bristle. Bristles of *M. multiunca* var. *pocosinensis* have a double cleft-like expanded distal end somewhat similar to the plume-like bristles of *Mallomonas plumosa*. The concave region of the distal hooked end is interrupted by the presence of a long pointed tooth that essentially divides the cleft into two sections. Based on initial findings it appears that this taxon is an important member of the phytoplankton flora in waterbodies along coastal regions of North Carolina.

Introduction

The genus *Mallomonas* Perty, in the class Synurophyceae (Andersen 1987), consists of unicellular motile cells with one or two emergent flagella, one bilobed or two separate elongated chloroplasts, and a highly organized covering of siliceous scales and bristles. The fine detail of scale and bristle structure, determined by electron microscopy (Asmund & Kristiansen 1986; Siver 1991), is used to identify species and subspecific taxa. Differences in bristle structure are most often used in conjunction with differences in scale structure to delineate between species. However, taxa with similar scale structure, but distinctly different bristles are typically recognized at the variety or form level. There are 163 species, varieties and forms of *Mallomonas* that have been described with electron microscopy (Kristiansen 2001).

Asmund (1956) originally described *Mallomonas multiunca* Asmund from an acidic, humus-stained bog lake in Denmark. In a later publication Asmund (1959) reported

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this species from additional acidic bog localities, as well as from several eutrophic and apparently more alkaline waterbodies, and concluded that this species was "adaptable to different environments". Since that time *Mallomonas multiunca* has been observed in both of these types of habitats (Siver 1991). Athough Asmund (1959) documented some variation in scale morphology, possibly related to temperature and trophic status, it was not considered of taxonomic significance (Asmund & Kristiansen 1986). In addition, no previous reports of *M. multiunca* have included differences in bristle structure. The purpose of this paper is to describe a new variety of *Mallomonas multiunca* based on a uniquely different bristle structure from several waterbodies situated along the coastal plain of North Carolina, U.S.A. In addition, the ecological conditions under which the type organism has been observed are reviewed.

Methods

Water, plankton net (10 µm) and surface sediment samples were made from the study locations in May and June, 2001. The surface sediment sample (one cm) was taken with a Glew gravity corer (Glew 1988). An aliquot of each plankton sample was dried onto a piece of aluminum foil, attached onto an SEM stub with Apiezon wax, coated with a mixture of palladium and gold for one minute with a Polaron model E 5100 sputter coater and observed with a Leo 982 SEM. The surface sediment samples were prepared according to the procedure used by Marsicano and Siver (1993) and also observed with a Leo 982 SEM.

Water samples were taken from each locality at the time of collection and used to measure a suite of chemical characteristics, including pH, total phosphorus and total nitrogen using the methods of Ahrens and Siver (2000), and water color using the platinum-cobalt method (APHA, 1985). Specific conductance and Secchi disk depth were measured in the field using a Hydrolab DataSonde 4A and a standard 20 cm black and white disc, respectively.

Results

Description:

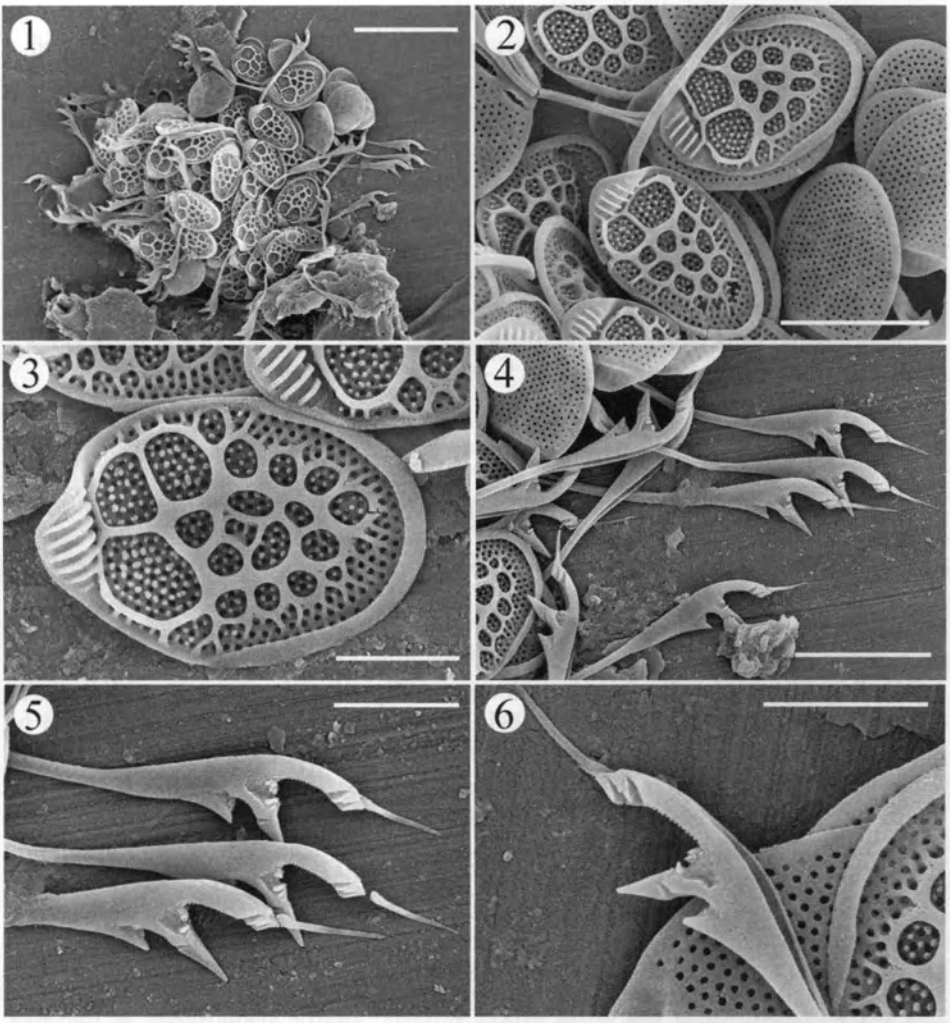
Mallomonas multiunca var. **pocosinensis** Siver nov. var.

Latin Diagnosis: A var. *multiunca* apicibus setarum differt: Sinus inter binos dentes apicis dente tertiore valde conspicuo aequaliter divisus est.

The bristles of var. *pocosinensis* differ from the type in having a distal end with a large pointed tooth that divides the hook into two concave sections.

Type Locality, Allen Road Canal, Pocosin Lakes Wildlife Refuge, North Carolina, U.S.A. Iconotype Fig 1. The epithet is named after the Pocosin vegetation that dominants the region.

Cells of *Mallomonas multiunca* var. *pocosinensis*, in the Series Heterospinae, are relatively small, spherical to slightly ovoid, range in size from 11-18 µm × 10-16 µm, and are covered with hooked bristles (Figs 1-6). As is true of the type, both the scales and bristles of var. *pocosinensis* are distinctive. The structure of the scales of var. *pocosinensis* is similar to that found in the type. Scales have a secondary layer consisting of a reticulum of thick ribs forming a series of large meshes (Figs 2-



Figs 1-6. *Mallomonas multiunca* var. *pocosinensis* Siver var. nov. Fig. 1) Siliceous remains of a whole cell. Scale bar = 5 µm. Figs 2-3) Close-ups of scales depicting both the ventral and dorsal surfaces. Note the series of secondary ribs forming the large meshes, two larger distally-placed meshes, dome with parallel ribs and papillae. Scale bar = 5 µm for Fig. 2 and 1 µm for Fig. 3. Fig. 4) The remains of a group of bristles showing the double-clefted distal end with elongated tip. Scale bar = 2 µm. Fig. 5) Close-up of Fig. 4 showing details of the distal ends of bristles. Note the long pointed tooth that divides the cleft into two sections and the long needle-like tip. Scale bar = 1 µm. Fig. 6) Close-up of a bristle tip. Note the notches, the minute teeth lining the concave margin and the longitudinal slit along the convex surface. Scale bar = 1 µm.

3). The two (Fig. 2) and sometimes three (Fig. 3) anterior-most meshes are much larger in diameter than the more posterior meshes. The base plate consists of relatively large, evenly-spaced pores (Fig. 2) and a series of papillae that are typically found

within the meshes (Fig. 3). The dome is fairly shallow, but broad, and consists of a series of parallel ribs (Figs 1-3). Scales range in size from 2.8-3.3 × 1.6-2.2 μm.

The bristles of *M. multiunca* var. *pocosinensis* consist of a thin and delicate shaft, a double cleft-like expanded distal end with a needle-like tip (Figs 4-6). The concave region of the distal hooked end is interrupted by the presence of a long pointed tooth that essentially divides the cleft into two separate concave regions. The concave portion below the large tooth is about half the size of the portion above the tooth (Figs 5-6). The edge of each concave section, especially the more distal one, is lined with a series of tiny teeth (Fig. 6). The distal-most portion of the concave region that bears the extended needle tip, and the large tooth, are both notched (Figs 5-6). A longitudinal slit is characteristic of the dorsal convex margin of the hook (Figs 4 & 6). Bristles range in length from 4.5 - 6.5 μm.

Ecological Findings:

To date, *Mallomonas multiunca* var. *pocosinensis* has been found in four different waterways in the Pocosin National Wildlife Refuge, one site in the Croatan National Forest and one locality in the Bladen Lakes State Park region (Table 1). All of these regions are situated on the Atlantic Coastal Plain in the State of North Carolina. The Pocosin National Wildlife Refuge and the Croatan National Forest are located along the coast and are largely dominated with vegetation known as pocosins (Richardson 1981). Pocosins are wetland regions dominated by evergreen shrubs and scattered emergent trees (Christensen et al. 1981). The shrub layer is often dominated by plants such as *Cyrilla racemiflora*, *Lyonia lucida*, *Ilex glabra* and *Zenobia pulverulenta*, and *Pinus serotina* is the most common tree species (Christensen et al. 1981). Drainage from the Pocosin National Wildlife Refuge is now accomplished by a series of canals and the refuge itself has been effectively isolated from the surrounding region through the construction of a series of earthen dams (Daniel 1981). The Bladen Lakes region consists of vegetation associated with carolina bays which is similar in composition to pocosin vegetation (Richardson 1981). Because the soils are comprised largely of woody peat and muck the wetlands, ponds and waterways associated with pocosin and carolina bay vegetation usually consists of acidic, poorly buffered and humic water.

Mallomonas multiunca var. *pocosinensis* was especially abundant in samples taken from four canals surveyed in the Pocosin National Wildlife Refuge (Table 1). In addition, abundant concentrations of scales were also found in surface sediments from each of the four canals. The water in each canal had a pH of only 3.5, was very humic with color values ranging from 1250 to 1500 Pt-Co units, high in both total phosphorus and total nitrogen and with specific conductance values ranging from 119-128 μS (Table 1). The high concentrations of both TP and TN were presumably due to the high level of suspended particulate matter that is found throughout the waterways of the Pocosin National Wildlife Refuge. *Mallomonas multiunca* var. *pocosinensis* was also found in pools of water in a locality in the Croatan National Forest that are actively managed in order to attract waterfowl (Table 1). Like the localities where this taxon was found in the Pocosin National Wildlife Refuge, this

Table 1. Characteristics of the waterbodies harbouring *Mallomonas multiunca* var. *pocosinensis* along the Coastal Plain of North Carolina, U.S.A.

Name	Latitude, Longitude	pH	Spec. Cond (µS)	TP (µg L^{-1})	TN (µg L^{-1})	Secchi Disc (m)	Color (Pt-Co units)
Conman's Corner[1]	N 35° 44' 43" W 76° 29' 36"	3.5	120	46	2,036	0.19	1,500
Clayton Canal[1]	N 35° 44' 21" W 76° 28' 39"	3.5	119	28	1,743	0.22	1,250
Allen Road Canal[1]	N 35° 44' 14" W 76° 30' 51"	3.5	128	49	2,120	0.14	1,665
Boerma Canal[1]	N 35° 44' 35" W 76° 29' 41"	3.5	121	38	2,008	0.2	1,415
Middle Catfish Waterfowl Im.[2]	N 35° 57' 41" W 76° 08' 32"	4.4	44	12	544	0.92	150
Jessup[3]	N 35° 51' 46" W 76° 43' 47"	3.7	93	10	850	0.56	875

[1] Pocosin Lakes National Wildlife Refuge
[2] Croatan National Forest
[3] Bladen Lakes State Forest Region

site, known as Middle Catfish Waterfowl Impoundment, was also very acidic (pH of 4.4), but was less humus-stained, contained lower concentrations of dissolved salts (sp. cond. = 44 µS) and had lower total phosphorus and total nitrogen concentrations (Table 1). Lastly, abundant numbers of the new taxon were also found in Jessup Pond, a very acidic (pH of 3.7), humic pond in the Bladen Lakes region (Table 1).

Discussion

Mallomonas multiunca var. *pocosinensis* differs from the type in the morphology of the bristles. Bristles of var. *pocosinensis* have a unique large tooth that divides the concave portion of the distal hook into two unevenly-sized sections. Bristles of the type lack the large tooth and have a single large concave portion. The dimensions of cells, scales and bristles for var. *pocosinensis* are all on the smaller ends of the ranges reported for the type (Asmund & Kristiansen 1986). Although larger domeless scales with a less developed secondary reticulum have been reported for the type (see Asmund & Kristiansen 1986), similar scales have not been observed for var. *pocosinensis*. Because the taxon appears to differ significantly from the type in bristle structure, but not in scale design, it is described as a new variety.

Interestingly, bristles of *M. multiunca* var. *pocosinensis* are similar in morphology to the plume-like bristles of *Mallomonas plumosa* Croome & Tyler (Croome & Tyler 1983). The plume bristles found on the anterior of the cells of *M. plumosa* have either one (like var. *pocosinensis*) or two large teeth that typically divide the concave region into roughly equal-sized sections. Despite the similarity in the distal ends of the bristles, *M. plumosa* and *M. multiunca* var. *pocosinensis* are very different species in all other respects (Asmund & Kristiansen 1986).

Based on a review of the literature it appears that *Mallomonas multiunca* is most often reported from two very different types of habitats (Table 2). Asmund (1956) originally described *M. multiunca* from a humic peat bog with a pH near 4. In a later

Table 2. Literature records indicating the ecological conditions of waterbodies where *Mallomonas multiunca* has been reported.

Reference	Locality	pH and Related Factors	Trophic Conditions	Spec. Cond. (µS)	Temperature (°C)	Seasonality	Other
1. Asmund 1956[1]	Denmark	Low pH, near 4	Bog, darkly stained water	----	18	August	----
2. Asmund 1959[1]	Denmark	Low pH, near 4 3.7 - 4.6	Bogs, darkly stained, oligotrophic	----	ca. 3 to warmer	Partial ice cover in March to June	----
3. Asmund 1959[1]	Denmark	Slightly alkaline, 7.5	Eutrophic	----	wide range from 3 to 25	March to June	Very adaptable; scale size related to temp. and trophic status
4. Harris & Bradley 1960[1]	England	Acidic peatlands	Humic stained	----	cool	Autumn	Data for type species
5. Harris & Bradley 1960[1]	England	----	Eutrophic, dung contaminated	----	cold	Winter to spring	Data for a eutrophic form
6. Bradley 1964[1]	Iceland	----	----	----	cold	Winter to spring, autumn	----
7. Wujek & van der Veer 1976[2]	Netherlands	Peat bog	----	----	----	----	----
7. Takahashi 1978[2]	Japan	Alkaline, 7 - 8	----	----	Cold stenotherm, 9 - 12	----	----
8. Wee et al. 1982[2]	Michigan, USA	bog	----	----	----	----	----
9. Dürrschmidt 1984[1]	Germany	----	----	----	----	----	Widespread but rare; Giessen region
10. Kristiansen 1985[2]	Denmark	Alkaline, 8.2 - 9.1	Highly eutrophic	----	Cold	Spring	----
11. Kristiansen 1988[2]	Denmark	Alkaline	Eutrophic	----	----	Spring	----
11. Hällfors & Hällfors 1988[1]	Finland	----	----	----	----	----	Common in Europe and NA

12. Hickel & Maass 1989[2]	Germany	----	----	----	----	----	Common in Europe and NA
13. Cronberg 1989[2]	Tropics, several countries	----	----	----	----	----	Not found in survey of tropics
14. Wee & Gabel 1989[2]	Iowa, USA	6.4 - 7.3	----	90; 400; 500	4 - 21	----	----
15. Wujek & Bland 1991[2]	Florida, USA, subtropics	----	----	----	----	----	Four sites, central/south FL
16. Siver 1991[1]	Connecticut, USA	5.6 - 6.2, acidic woodland pond	----	<100	----	Autumn	----
17. Santos & Leedale 1993[2]	Portugal, subtropics	----	----	----	----	----	----
18. Wee et al. 1993[2]	Louisiana, USA subtropics	7.0 - 7.2	----	----	17.6; 24.5	----	----
19. Wujek & Swinehart 1995[2]	Indiana, USA	6.4	----	----	----	----	----
20. Calado & Craveiro 1995[1]	Portugal	----	Eutrophic	----	----	Spring	Three of 25 sites
20. Santos et al. 1996[1]	Portugal, subtropics	7.3 - 8.2	----	470 - 1040	11.8 - 24.7	----	----
21. Gutowski 1997[1]	Germany, Berlin region	7.4 - 7.8	----	660 - 990	cold, 3.4 - 11.5	----	----
22. Voloshko & Gavrilova 2001[2]	Russia	----	----	----	----	----	Reviews other records from Russia
23. Wei & Yuan 2001[1]	China, Jiangxi Province	7.3	----	----	20	----	New record for China

[1] Reference includes information (an image or comment) on bristle morphology and represents the type variety.
[2] Reference does not include information on bristle morphology.

communication Asmund (1959) again reported this taxon from bog localities with pH between 3.7 and 4.6. Similarly, Harris and Bradley (1960) reported *M. multiunca* from small pools on acid peatlands. In North America, Wee et al. (1982) also described this organism from a bog and Siver (1991) reported it from a small acidic wooded lake. In this study var. *pocosinensis* was found as a dominant organism in only very humic and acidic habitats associated with pocosin or carolina bay vegetation.

In addition to being found in habitats characterized by acidic and humic conditions, *M. multiunca* is perhaps more often reported from eutrophic waterbodies with a much higher pH (Table 2). Asmund (1959) reported this organism from a eutrophic farm pond with a pH near 7.5. Harris and Bradley (1960) found scales of this taxon in eutrophic, dung-contaminated pools and Kristiansen (1985) reported *M. multiunca* from a highly eutrophc, alkaline lake with a pH ranging between 8.2 and 9.1. *Mallomonas multiunca* has also been found in alkaline localities in Japan (Takahashi 1978), Iowa, USA (Wee & Gabel 1989), Germany (Hickel & Maass 1989; Gutowski 1997), Louisiana, USA (Wee et al. 1993), Portugal (Santos et al. 1996) and China (Wei & Yuan 2001).

Given the two distinctly different types of localities in which *M. multiunca* has been found the question arises as to whether the totality of all records represent two distinct taxa that differ on a physiological, but not morphological, basis. Unfortunately, there is insufficient data to support or refute such a claim. Culture studies would most likely be necessary to make such a determination. The unique type of bristle described here for var. *pocosinensis* has not been previously reported, regardless of habitat type.

It is also of interest to note that although Durrschmidt (1984) reported *M. multiunca* as a rare but widespread taxon, Hickel and Maass (1989) noted that it was mostly found in North America and Europe. Since these publications *M. multiunca* has continued to be reported from localities on both of these two continents (Table 2), as well as one finding from China (Wei & Yuan 2001), but has not been reported from the tropics (Cronberg 1989). Thus, based on all available reports at this time, *M. multiunca* appears to be primarily restricted to temperate and subtropical localities in the northern hemisphere.

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