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Stauroneis kingstonii sp. nov. (Bacillariophyta: Naviculales), a new diatom species from the Black Swamp, Arkansas, USA

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Abstract

A freshwater diatom species, *Stauroneis kingstonii sp. nov.,* is described from cypress-tupelo wetlands of the Cache River, Arkansas, USA. *Stauroneis kingstonii* can be distinguished from other *Stauroneis* species by its narrow lanceolate shape, high length:breadth ratio, coarse areolae and striae, and lateral raphe bounded by a broad axial area and straight proximal raphe ends. The diatom is currently known only from the Cache River Watershed and found living benthic or epiphytic on submerged bald cypress and water tupelo tree bark, in slightly acidic, and fresh to slightly brackish waters.

Introduction

Stauroneis Ehrenberg (1843: 311), a freshwater diatom genus erected by Ehrenberg (1843a) and later typified by Boyer (1927), is characterized by uniseriate striae, long narrow valves, and a thickened transverse central area referred to as a stauros. *Stauroneis* taxa are commonly found in epipelic, mossy, or aerophilic habitats (Round *et al.* 1990). Traditionally considered to have low diversity, much recent work has expanded the richness of this genus to include 17 new species in Sardinia (Lange-Bertalot *et al.* 2003), 6 new species in Germany (Werum & Lange-Bertalot 2004), 40 new species in Antarctica (Van de Vijver *et al.* 2004), and 21 new species in western North America (Bahls 2010, 2012). These findings contrast with typical reports from the USA such as the U.S. Geological Survey's National Water Quality Assessment (NAWQA) Program that record less species richness. For example, NAWQA (U.S. Geological Survey 2014) found the greatest *Stauroneis* richness for Arkansas within the Cache River, but these only included six known taxa (*S. anceps* Ehrenberg (1843b: 306), *S. obtusa* N.Lagerstedt (1873: 36), *S. producta* Grunow (in van Heurck 1880: pl. 4), *S. smithii* Grunow (1860: 564), *S. smithii* f. *incisa* Pantocsek (1902: 21), and *S. thermicola* (J.B.Petersen) Lund (1946: 61)) and one unknown species (*Stauroneis* sp. 0A UL NAWQA KM).

The Cache River, in Arkansas, flows from north to south within the Mississippi River Alluvial Plain (MAP). The MAP ecoregion is the large floodplain stretching from southern Illinois to the mouth of the Mississippi River, comprising 141,958 km^2 of glacial-fluvial outwash (Omernik 1987). The humid, sub-tropical climate of the MAP is advantageous for wetland vascular plant communities dominated by bottomland hardwood forests (Hodges 1997). As part of the MAP, the Cache River Watershed (CRW) was formed from the deposition of Pleistocene glacial-fluvial outwash (Saucier 1994), and millennia of river meanderings have resulted in numerous wetlands and oxbow lakes (Fig. 1). Based on their hydrology and geomorphic setting within the floodplains, connected depressions are hydrogeomorphically classified wetlands that are common throughout the CRW and MAP (Klimas *et al.* 2004). Forests of bald cypress, *Taxodium distichum* (L.) Rich. (1810: 298) and water tupelo, *Nyssa aquatica* L. (1753: 1058), dominate connected depressions, which are subject to regular inundation (Dale & Ware 2004). In addition to beneficial nutrient sequestration and floodwater retention, cypress-tupelo wetlands have been shown to be vital wildlife habitat for a variety of fish (Killgore & Baker 1996) and song birds (Wakeley & Roberts 1996).

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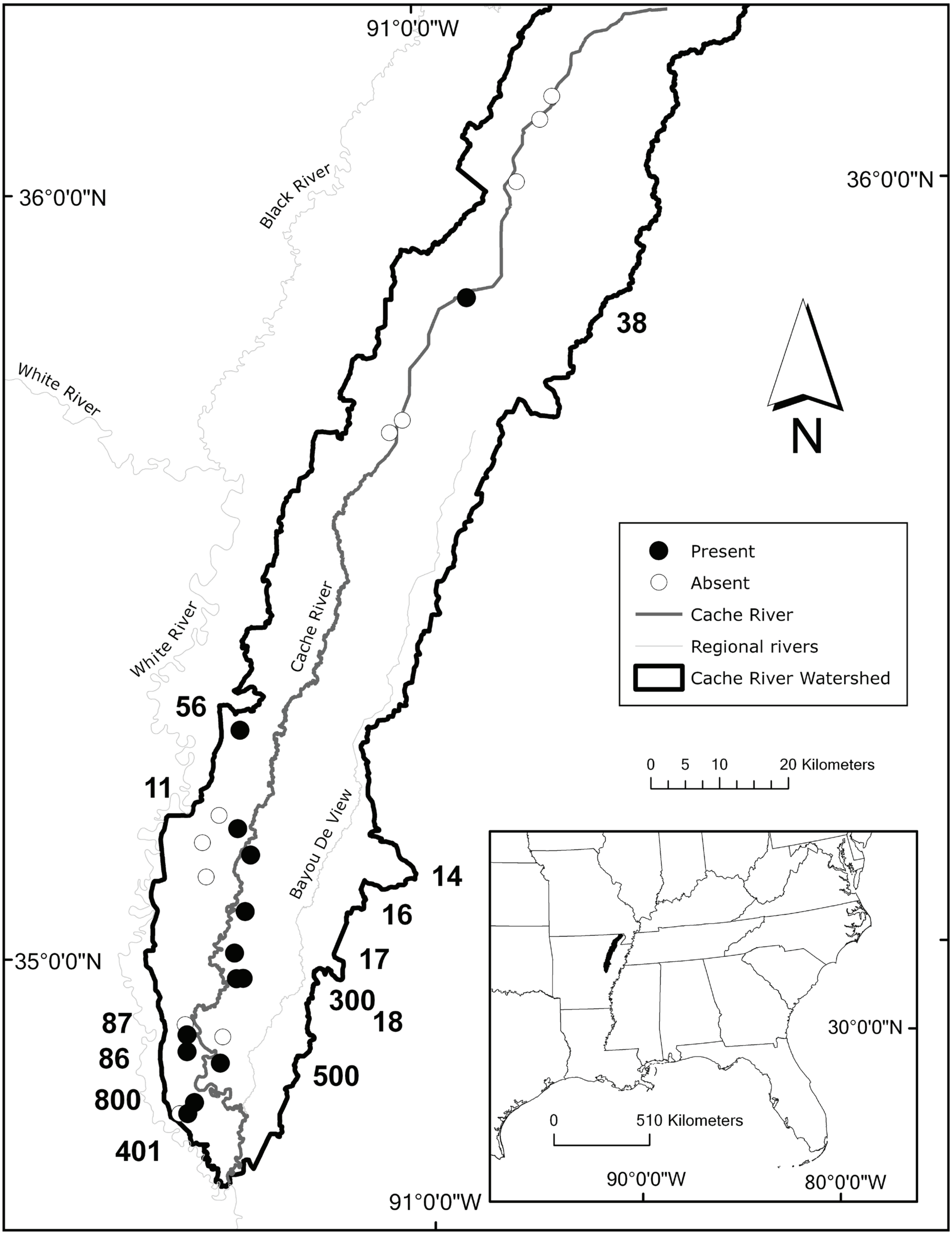


FIGURE 1. The Cache River watershed, Arkansas, USA. Station 14 is the *Stauroneis kingstonii* type locality. Filled circles indicate numbered sampling stations where *Stauroneis kingstonii* occurred and unfilled circles indicate where valves did not occur in samples.

The MAP and the CRW have experienced land-use changes from being predominantly forested to becoming agricultural; the watershed is now dominated by 85% agriculture land-use (Layher *et al.* 2004, Sayler 2012). Despite regional alterations, many areas along the southern extent of the Cache River are under conservation management by U.S. Fish and Wildlife Service, Arkansas Game and Fish Commission, Arkansas Natural Heritage Commission, and The Nature Conservancy. The wetland forests of the Rex Hancock Black Swamp Wildlife Management Area, the Cache River National Wildlife Refuge, and the White River National Wildlife Refuge compose one of the largest remaining tracts of bottomland hardwood forests in the MAP, and the area is recognized as a wetland of international importance (Ramsar 1987).

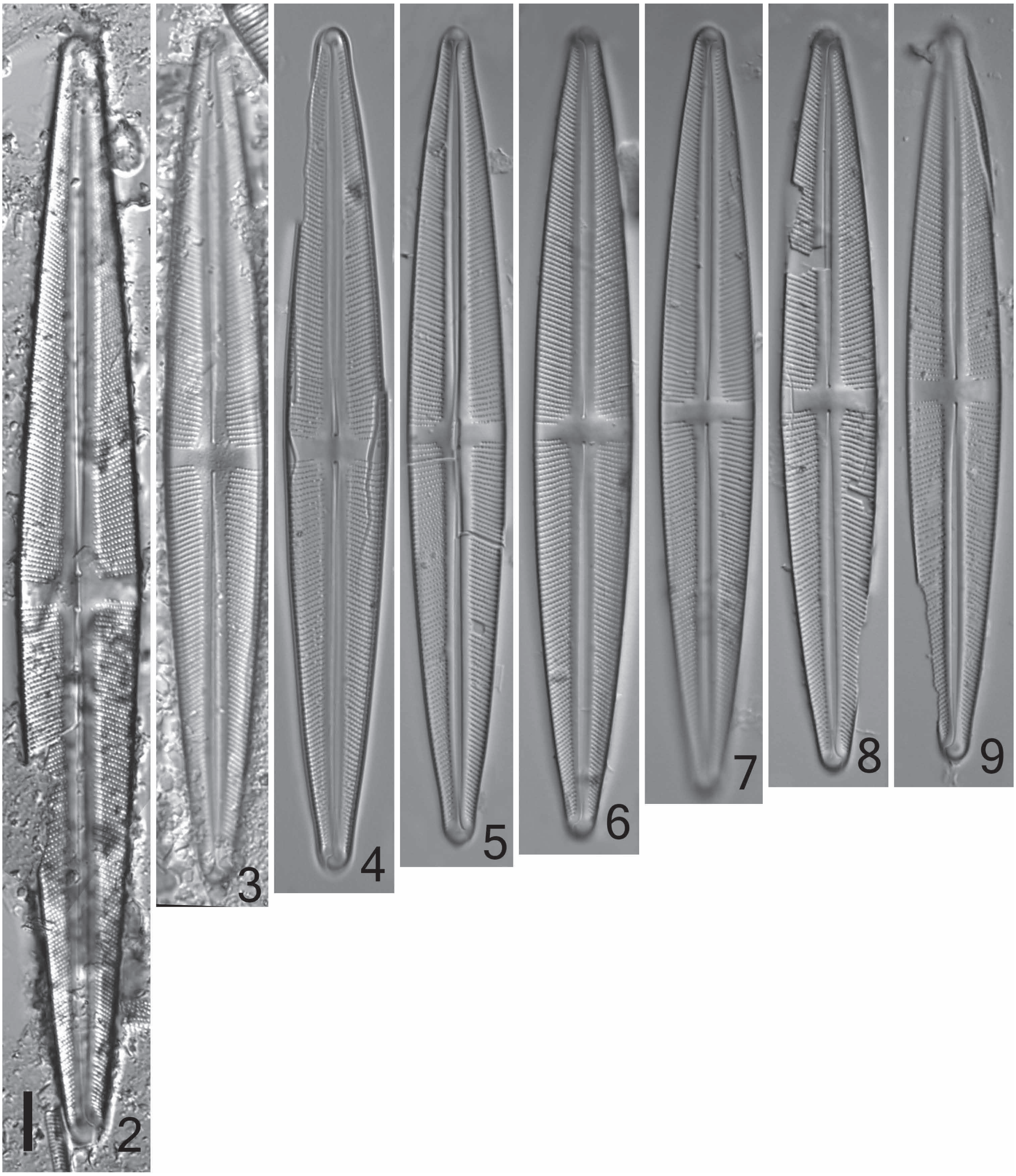
While there has been exploration of the diatom communities of this region, there have been no investigations of the epidendric and benthic diatom communities of the cypress-tupelo wetlands (Sears & Couch 1932, Czarnecki 1990, Pan and Stevenson 1996, Kleiss *et al.* 2000, Smith 2008, Enache & Potapova 2009, Bhattacharya 2012). As part of a study to develop biological indicators of wetland water quality, samples were collected from epidendric and benthic habitats to assess differences in diatom community structure. During this investigation a new diatom species was observed. We describe *Stauroneis kingstonii sp. nov.*, based on morphological characteristics distinguished using light and scanning electron microscopy. The morphological features are compared with similar *Stauroneis* species: *S. acuta* W. Smith (1853: 59), *S. angustilancea* Lange-Bertalot and Metzeltin (2004: 175), *S. baconiana* Stodder (1859: 26), *S. beeskovea* L.Bahls (2010: 55), *S. gremmenii* Van de Vijver and Lange-Bertalot (2004: 39), and *S. indianopsis* L.Bahls (2010: 85) (Table 1).

Materials and Methods

Twenty-four wetlands along the Cache River were sampled for diatom community composition in June 2012. Epidendric samples were collected from cypress-tupelo communities by scraping the bark of trees with a razor blade 2–4 cm below the waterline. Benthic samples were obtained using a spatula to lift an inverted half of a petri dish that was depressed into the sediment. All samples were placed in 50 ml centrifuge tubes and preserved in a 3% glutaraldehyde solution. Diatom samples were cleaned using 30% hydrogen peroxide and nitric acid and rinsed with deionized water (Stoermer *et al.* 1995). Some cleaned material was filtered through 65 μm-mesh netting to remove clays (compare Figs 3 and 4). Aliquots of cleaned samples were dried to coverslips and then mounted on microslides using Naphrax® (Brunel Microscopes Ltd., Wiltshire, UK). A Leica DMLB II microscope (Leica Microsystems, Wetzlar, Germany) with differential interference contrast optics and operating at 1000x magnification under full oil immersion was used in conjunction with a Qimaging 3.3 M camera for micrographic documentation. For scanning electron microscopy, cleaned type material was dried onto coverslip fragments that were mounted on aluminum stubs. Stubs were then sputter coated with Au-Pd and examined in a JEOL JSM-6060LV (JEOL Ltd., Tokyo, Japan) scanning electron microscope operated at 15 kV at St. Cloud State University (Minnesota, USA). For each sampling station, 600 diatom valves were counted along random transects, and species data were converted to percent abundance relative to total diatom count at each station (Burge 2014).

Forty-four specimens of the new species were examined in the light microscope and the length, breadth, striae density, areolae density, and the length:breadth ratios of each were recorded. Terminology for valve morphology follows Barber & Haworth (1981), Round *et al.* (1990), and Cox (2012).

Physicochemical parameters were sampled in May, June, and July 2012. Field parameters including pH, dissolved oxygen (mg l^{-1}), specific conductance (μS cm^{-2}), and temperature (^{o}C) were measured on a Thermo Scientific Orion Star A329 pH/ISE/Conductivity/Dissolved Oxygen Portable Multiparameter Meter (Thermo Fisher Scientific Inc., Waltham, Massachusetts). Turbidity was analyzed using a Hach 2100P Turbidity Meter (Hach Company, Loveland, Colorado) in the field or in the lab at the end of each field day. Water samples collected included 1.0 l of water for ammonia, total nitrogen, total phosphorus, and chlorophyll-a, and two 15 ml filtered (0.45 μm) water samples for dissolved nutrients (U.S. Geological Survey 2006). Water quality samples were frozen upon collection/processing until analysis at Arkansas State University Ecotoxicology Research Facility (Cobb 2013).



FIGURES 2–9. Size diminution series of *Stauroneis kingstonii*, LM, DIC. Fig. 3. Holotype specimen (ANSP GC-65192). All images in valve view and at same scale; scale bar (Fig. 2) = 10 µm.

New Species Description

Stauroneis kingstonii Burge, Marsico & Edlund *sp. nov.* (Figs 2–13)

Valves linear-lanceolate and relatively narrow with acutely rounded ends. Length 118.0–186.3 µm, breadth 14.7–20.1 µm. Axial area narrow at the apices, widening toward the stauros (Fig. 10). Central area with a rectangular stauros, slightly widening near the valve margins. Shortened striae not observed in the central area. Raphe fissures lateral, proximal endings nearly straight, and distinctly inflated (Fig. 10). Distal raphe ends hooked in a sickle shape toward the secondary side of the valve (Fig. 10). Striae radiate throughout, 11–13 in 10 µm. Areolae within striae distinct, 16–20 in 10 µm. For measurements, n = 44 valves. Pseudosepta were not observed.

Type:—UNITED STATES. Rex Hancock Black Swamp, Cache River, near Howell, Arkansas, USA. Elevation 58 m, 35°7’55.31”N, 91°16’59.27”W, collected 21 June 2012, D.R.L. Burge (ANSP GC–65192, circled specimen, **Type! designated here** (= Fig. 3), prepared from material ANSP GCM—5694; CANA–108109, **isotype! designated here**).

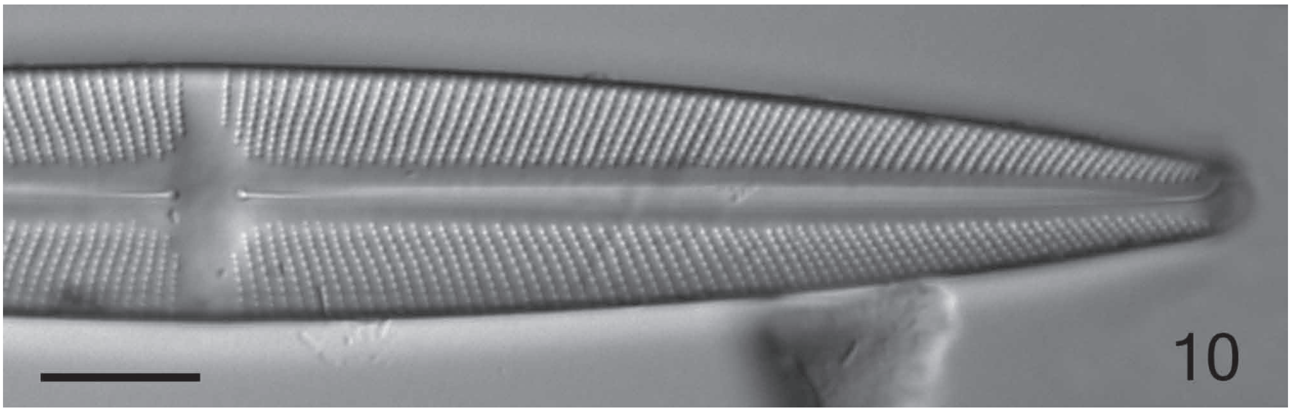


FIGURE 10. Raphe features of *Stauroneis kingstonii*; lateral raphe branch and broad axial area, proximal raphe ends nearly straight, and distal raphe ends sickle-shaped. Scale bar = 10 µm.

Observations:—*Stauroneis kingstonii* is distinguished among *Stauroneis* taxa by its large relatively narrow valves, acutely rounded apices, relatively course striae, lack of pseudosepta, and broad axial area. Characteristic of *Stauroneis* taxa, the internal distal raphe ends are slightly deflected to one side, and single rows of areolae are internally occluded by hymenes (Fig. 13). The internal proximal raphe ends terminate with a slight curve into the stauros, which is continuous across the central area. The siliceous thickenings of the axial area continue along the raphe to the apices (Fig. 12).

Stauroneis kingstonii is distinct by its linear valve shape characterized by the high length:breadth ratio of 7.6 to 10.8 (Table 1). *Stauroneis acuta* also has a high length:breadth ratio; however, the valve outline of *S. acuta* is distinctly rhombic, the striae are more radiate, proximal raphe ends are curved, and the areolae are less dense when compared to *S. kingstonii. Stauroneis beeskova* also has a similar valve outline with a high length:breadth ratio; however, *S. beeskova* differs from *S. kingstonii* by the more radiate striae pattern occurring in greater density, and the length:breadth ratio is typically less. While *S. baconiana* and *S. indianopsis* have similar length ranges to *S. kingstonii*, both species have higher breadth ranges and striae densities in addition to more rostrate valve apices. *Stauroneis indianopsis* has distinctly curved proximal raphe endings, whereas in *S. kingstonii* they are only sometimes slightly bent. *Stauroneis gremmenii* is similar in length to *S. kingstonii*; however, the valves in *S. gremmenii* are wider, with rostrate apices, and greater striae density. With breadth and narrowly lanceolate valve outline similar to *S. kingstonii, S. angustilancea* differs by having curved raphe endings, shorter lengths, and lower length:breadth ratio, and protracted apices.

Ecology and biogeography:—Habitat is epidendric or epipelic. *Stauroneis kingstonii* was found occurring at less than 1% abundance at over half of the bio-indicator sampling stations, predominantly in the downstream portion of the CRW (13 out of 24 stations; Fig. 1, Table 2). Recognizing that our sample size is too small to draw robust ecological tolerances, we note specimens were found on both epidendric substrates and in benthic samples, with seven occurrences each on *Taxodium* and in the benthic samples, but only two observations on *Nyssa*. Across the multiple substrates, other diatom taxa found at 5% or greater abundance in communities with *S. kingstonii* include: *Achnanthes inflata* (Kützing) Grunow (1867: 7), *Diadesmis confervacea* Kützing (1844: 109), *Eolimna tantula* (Hustedt) Lange-Bertalot (2004: 147), *Eunotia bilunaris* (Ehrenberg) Schaarschmidt (1880: 159), *E. metamonodon* Lange-Bertalot (2011: 153), *Frustulia vulgaris* (Thwaites) De Toni (1891: 280), *Gomphonema gracile* Ehrenberg (1838: 217), *Humidophila contenta* (Grunow) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová (2014: 357), *Lemnicola hungarica* (Grunow) F.E.Round & P.W.Basson (1997: 77), *Nitzschia amphibia* Grunow (1862: 574), and *N. palea* (Kützing) W.Smith (1856: 89) (Burge 2014).

Wetlands were slightly acidic to circumneutral, and *S. kingstonii* tolerated both low dissolved oxygen conditions and elevated nutrients (Table 3). The lack of forest buffer and increased turbidity in the wetlands suggests some tolerance of *S. kingstonii* to moderate disturbance. Consistent with most *Stauroneis* taxa, *S. kingstonii* occurred predominantly in freshwater wetlands, with a few observations in fresh-brackish wetlands (range: 71.2–537.0 µS cm^{-1}; Table 3). As this is the first description of the species, knowledge about its full ecological niche and biogeographical distribution is preliminary.

TABLE 1. Morphometric comparisons among *Stauroneis kingstonii* (n=44), *S. acuta*, *S. angustilancea*, *S. baconiana*, *S. beeskova*, *S. gremmenii*, and *S. indianopsis*.

	S. kingstonii	*S. acuta*[1]	*S. angustilancea*[2]	*S. baconiana*[2]	*S. beeskova*[2]	*S. gremmenii*[1]	*S. indianopsis*[2]
Valve length (µm)	118 –186.3	130–180	85–115	115–140	93–133	119–152	105–163
Valve breadth (µm)	14.7–20.1	25–32	14–17	15–22	14–20	22–24	20–27
L:B Ratio	7.59–10.8	5.3–6.7	5.2–6.6	7.6–9.3	6.5–7.7	5.1–6.3	5.0–6.4
Striae in 10 µm	11–13	15–16	17–20	16–22	21–24	16–18	16–17
Areola in 10 µm	16–20	14–16	16–18	16–22	18–22	17–18	16–18
Valve outline	linear-lanceolate	rhombic-lanceolate	narrowly lanceolate	rhombic-lanceolate	narrowly lanceolate	lanceolate to linear-lanceolate, undulate margins	linear-lanceolate
Apices	acutely-rounded	broadly rounded non-protracted	rounded, protracted	rostrate to sub-capitate	rounded, subtly protracted,	rostrate, strongly non-abruptly protracted	pinched, barely protracted
Proximal Raphe	nearly straight	curved	curved	nearly straight	nearly straight	straight, clearly expanded	curved
Striation	radiate, strongly near the apices	radiate to strongly radiate	radiate	radiate	steeply radiate	moderately radiate in the middle, strongly towards the apices	radiate, strongly near the apices

[1]Van de Vijver et al. 2004, [2]Bahls 2010

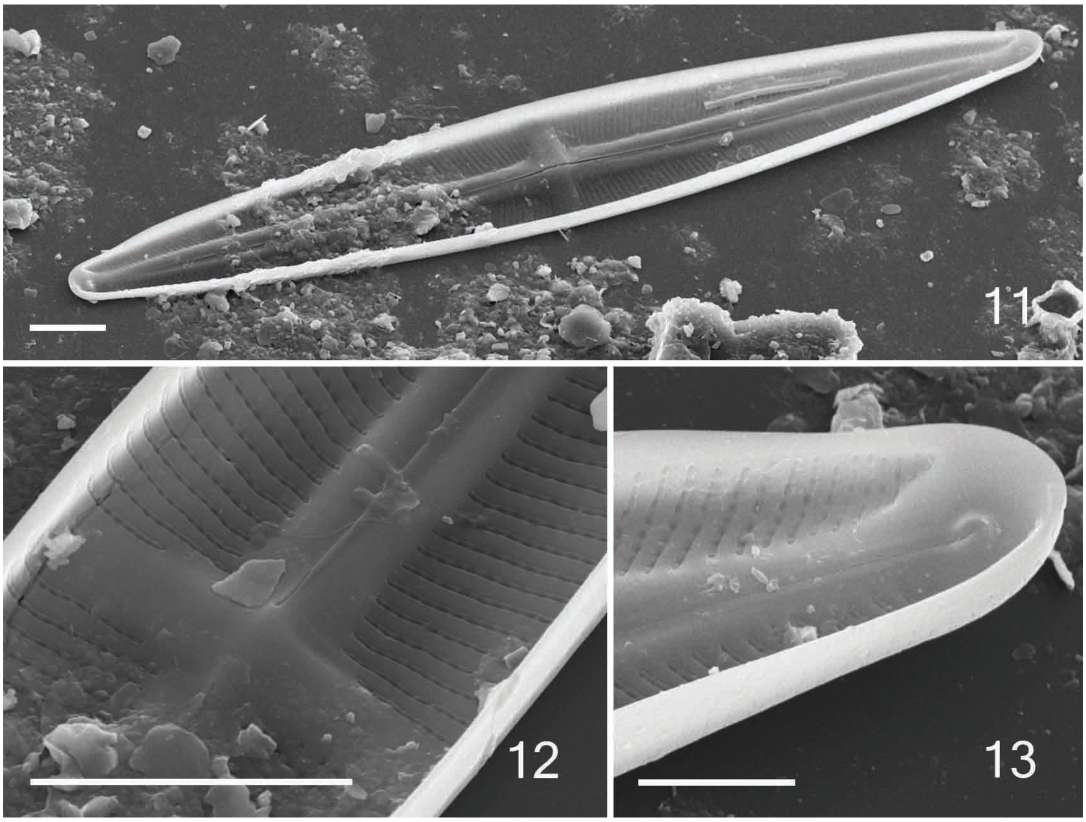
TABLE 2. Location, date, and percent abundance of *Stauroneis kingstonii* on *Taxodium distichum* (bald cypress), *Nyssa aquatica* (water tupelo), and benthic substrates.

Station	Date	Latitude	Longitude	Bald cypress	Water tupelo	Benthic
11	28 Jun 2012	35.16688	-91.30278	-	0.3	0.2
14*	21 Jun 2012	35.13203	-91.2831	0.5	0.2	-
16	25 Jun 2012	35.05811	-91.2932	0.2	-	-
17	25 Jun 2012	35.00391	-91.31139	0.2	-	-
18	22 Jun 2012	34.97034	-91.30892	-	-	0.5
38	20 Jun 2012	35.85646	-90.92	-	-	0.2
56	22 Jun 2012	35.29561	-91.29657	0.2	-	-
86	26 Jun 2012	34.87577	-91.38957	0.2	-	-
87	25 Jun 2012	34.89798	-91.38904	-	-	0.5
300	28 Jun 2012	34.97073	-91.2982	-	-	0.7
401	25 Jun 2012	34.794638	-91.389822	0.2	-	0.2
500	27 Jun 2012	34.8601	-91.337	<0.1	-	-
800	28 Jun 2012	34.80901	-91.37873	-	-	0.5

*: Type locality

-: Absent

Etymology:—*Stauroneis kingstonii* honors the diatomist John C. Kingston (1949–2004). Initially a student and then a regular visitor to the Iowa Lakeside Laboratory *Diatom Ecology and Systematics* course, Dr. Kingston is honored for his lifetime of contributions to diatom research by a teaching assistant fellowship, under which this work was conducted. As a diatom distinct within the genus, *S. kingstonii* is appropriately named in honor a scientist who stood out among his peers (Stoermer & Smol 2004).



FIGURES 11–13. SEM images of *Stauroneis kingstonii*. Fig. 11. Internal view of an entire valve. Scale bar = 10 µm. Fig. 12. Internal valve view showing thickened stauros, longitudinal ribs of raphe sternum, and slightly deflected proximal raphe ends. Scale bar = 10 µm. Fig. 13. Internal valve view showing distal raphe end deflected into a helictoglossum, striae comprising single rows of areolae that are each occluded by a hymene. Scale bar = 5 µm.

TABLE 3: Physiochemical parameters and percent forested land-use within the 100m buffer for 13 wetlands with *Stauroneis kingstonii.*

Parameter	Mean	Minimum	Maximum
pH	6.47	5.78	7.12
DO (mg l^{-1})	2.22	0.08	6.01
Conductivity (µS cm^{-1})	192.77	71.17	537.00
Turbidity (NTU)	36.01	8.61	176.93
NO_3-N (mg l^{-1})	0.05	0.02	0.12
NO_2-N (mg l^{-1})	0.01	0.00	0.05
TN (mg l^{-1})	1.59	0.76	7.18
TP (mg l^{-1})	0.49	0.10	3.93
PO_4-P (mg l^{-1})	0.21	0.03	2.08
Forest (%)	51.75	14.06	79.88

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