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Geographic Range and Morphological and Chromosomal Variability of *Carex molestiformis* (Cyperaceae) East of the Mississippi River

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Abstract

Carex molestiformis, described in 1997 as an endemic to the Ozark and Ouachita Mountain regions, is newly collected from Georgia, Mississippi, and Ohio. Herbarium records also have confirmed this species from North Carolina, Virginia, and West Virginia bringing the overall range of this species to 11 states. A morphological comparison of *C. molestiformis* from east of the Mississippi River with western populations did not reveal regional differences. Some eastern populations occupied ruderal habitat, namely drier grassy roadsides and hay meadows rather than the river bottom openings typical of western populations. New chromosome counts for this species from the eastern portion of its range confirmed existing reports of $n = 37$ and also found agmatoploidy with $n = 35$.

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INTRODUCTION

Carex molestiformis Reznicek & P. Rothr. is a member of section Ovales with close morphological similarity to the widespread species *C. brevior* (Dewey) Mack. *ex* Lunell and *C. molesta* Mack. *ex* Bright. All three species have gynaecandrous spikes, medium size, broadly winged perigynia, cespitose habit, and leaf sheaths with a conspicuous hyaline zone on the ventral face. Although these species are only subtly different, *C. molestiformis* is diagnosed by its few spikes per inflorescence, the terminal one having a prominent staminate base, and its relatively wide achenes. When first described (Reznicek and Rothrock 1997), *C. molestiformis* was thought to be endemic to the Ozark and Ouachita Mountain regions of Arkansas, Missouri, and Oklahoma. However, before publication of the Cyperaceae for the "Flora of North America" (Mastrogiuseppe et al. 2002), scattered collections indicated that the species, although rare, was nonetheless a floristic element in the central Appalachians and parts of the Cumberland Plateau in Kentucky and Tennessee. Further herbarium review and new collections have enlarged the range of this species from five to eleven states. Here we review the biogeography of *C. molestiformis*, examine the morphological variability of Appalachian versus Ozark-Ouachita populations, and report additional chromosome counts.

MATERIALS AND METHODS

Three of the new state records were the result of our ongoing field work. An exhaustive search of herbarium materials was deemed beyond the scope of this study. However, in our effort to elucidate state-level ranges we have examined materials from DUKE, NCU, PAC, PENN, US, and WVA.

Non-metric Multidimensional Scaling (NMS) was performed, using PC-ORD5 software (McCune and Mefford 2006), to test whether a sample of 18 accessions of *Carex molestiformis* from regions east of the Mississippi River differed meaningfully from 15 Ozark-Ouchita accessions. Also, 16 *C. brevior* accessions were selected from throughout its geographic range to permit comparisons of intra- and inter-specific variation. The data matrix included 13 potentially diagnostic characters measured from inflorescences (overall length, length of lowest internode, and number of spikes), spike bases (length of male portion of terminal spike), pistillate scales (length, width, pointedness, and scale length/perigynium length), and perigynia (overall length, body length, width, length/width ratio, and number of ventral nerves). Pointedness of the pistillate scales is the distance from the apex of the scale to the perpendicular line equal to 0.5 mm in length. Sørenson (Bray-Curtis) served as the distance measure. A random starting configuration was employed in the first run of 250 real data and 250 randomized data iterations. Scree plots recommended that two dimensions be analyzed in the final run. The final run had 53 iterations and had achieved a stable solution. The resulting Kruskal stress was moderate (12.3).

Chromosomes counts were made of accessions from Kentucky and Tennessee following a previously described protocol (Rothrock and Reznicek 1998). Briefly, field transplants were grown in the greenhouse. Cold treatment was used to stimulate the production of new inflorescences. Immature spikes were preserved in methanol, chloroform, and propionic acid (6 3 2). Within the subsequent 72 hours, anthers were dissected from the spikes and squashed in 2% lactic-acetic-orcein (Cooperrider and Morrison 1967). Meiosis I chromosome figures were examined from five or more pollen mother cells. Photographs and drawings were made with a Nikon Labophot-2 microscope using phase contrast at 1,000× magnification. Voucher specimens, prepared from fruiting greenhouse material and deposited at the University of Michigan Herbarium (MICH), include photomicrographs and drawings of countable figures.

RESULTS AND DISCUSSION

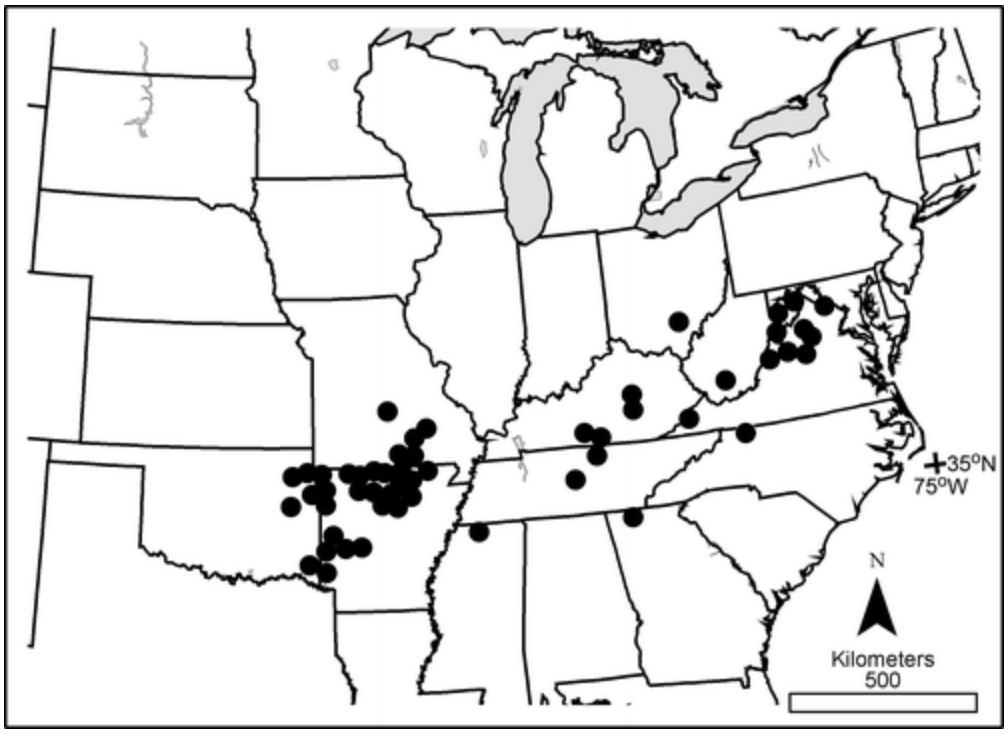
New State Records

Georgia, Murray County

Narrow unmowed stream alluvium bank, Cohutta Springs Road at jct. with Summerour Road, ca. 1 mi E of US 441 (34°53.326• *P.E. Rothrock 4817* (GA, MICH, VSC, Bryson herbarium).

Mississippi, Marshall County

Open wet roadside ditch and meadow along bypass Street, Potts Camp, W of Bypass Street; just N of Hwy US 178; between Hwy US 78 and US 178 (34°39.175



In the western portion of its range, the native habitat of *C. molestiformis* appears to be openings in river bottoms (Reznicek and Rothrock 1997). Further east, populations tend to be more ruderal but are usually not far from rivers or streams and other bottomland sedges (e.g., *Carex squarrosa* L., *C. typhina* Michx., *C. grisea* Wahlenb.). The habitat at the Georgia and Mississippi localities compares favorably with those previously observed. The Georgia site, along Sumac Creek, supported bottomland species such as *Carex aureolensis* Steud. and *C. caroliniana* Schwein., and woody species *Betula nigra* L., *Celtis occidentalis* L., *Sambucus canadensis* L., and *Ulmus americana* L. The Mississippi site had *Carex longii* Mack. and *C. oklahomensis* Mack. Likewise among the herbarium records, with exceptions, habitat descriptions are often alluvial, e.g., "short-vegetation meadow edge … near river" (Alleghany County, North Carolina, *C. J. Rothfels 2917*, DUKE, MICH), "low wet ground" (Rutherford Co., Tennessee, K. *Rogers 44773*, NCU), "flat meadow of thin grass near stream" (Shenandoah National Park, Augusta Co., Virginia, *F.R. Fosber and E.H. Walker 36407*, US), and "head of Blackthorn Run" (Pendleton Co., West Virginia, *E.E. Hutten s.n.*, WVA).

The recently discovered Ohio (Hocking Co.) site was somewhat unusual in being relatively dry and elevated. It raises the possibility that some populations of *C. molestiformis* may not be native but rather are incidences of introduction in contaminated hay or grass seeds planted for erosion control and roadside maintenance or for pastures and hay meadows. Two collections within Shenandoah National Park (Stony

Man Mountain overlook, *F.R. Fosber and E.H. Walker 36480*, US; Hogswallow Flat overlook, *Fosber 48449*, US) are also from dry grassy roadsides and *C. molestiformis* was observed by one of us (CTB) along open roadsides and in high hay meadows and pastures for livestock off the Blue Ridge Parkway southeast of Waynesboro, Virginia in 2006. However, some native populations may be located on drier habitat, for example several early 1940s collections from Grant County, West Virginia from "shale barrens near Petersburg" (*WVa. U. Biol. Expedition s.n.*, WVA). The probable existence of native as well as human influenced populations of *C. molestiformis* poses an interesting conservation problem. Even within its presumed native habitat, *C. molestiformis* depends upon a disturbance regime for sustaining populations (Reznicek and Rothrock 1997). In ruderal habitat it is difficult to differentiate between natural populations of an early successional species versus those perhaps infrequent populations that were inadvertently aided by human activity.

Regardless of nativity, it is important to consider whether these eastern populations of *C. molestiformis* retain clear species delimitation from other species in *Carex* section Ovales and whether they exhibit any apparent differentiation from populations occurring in the distant Ozark Mountain center of distribution. As expected, the NMS analysis sorted all *C. brevior* from *C. molestiformis* (Figure 2). The two species exhibited a similar degree of infraspecific variation (axis 1). The 18 specimens of *C. molestiformis* from east of the Mississippi River had a range of variation that overlapped fully with specimens from further west. Although the eastern populations are often more ruderal as well as less frequent, they did not exhibit any apparent morphological differences.

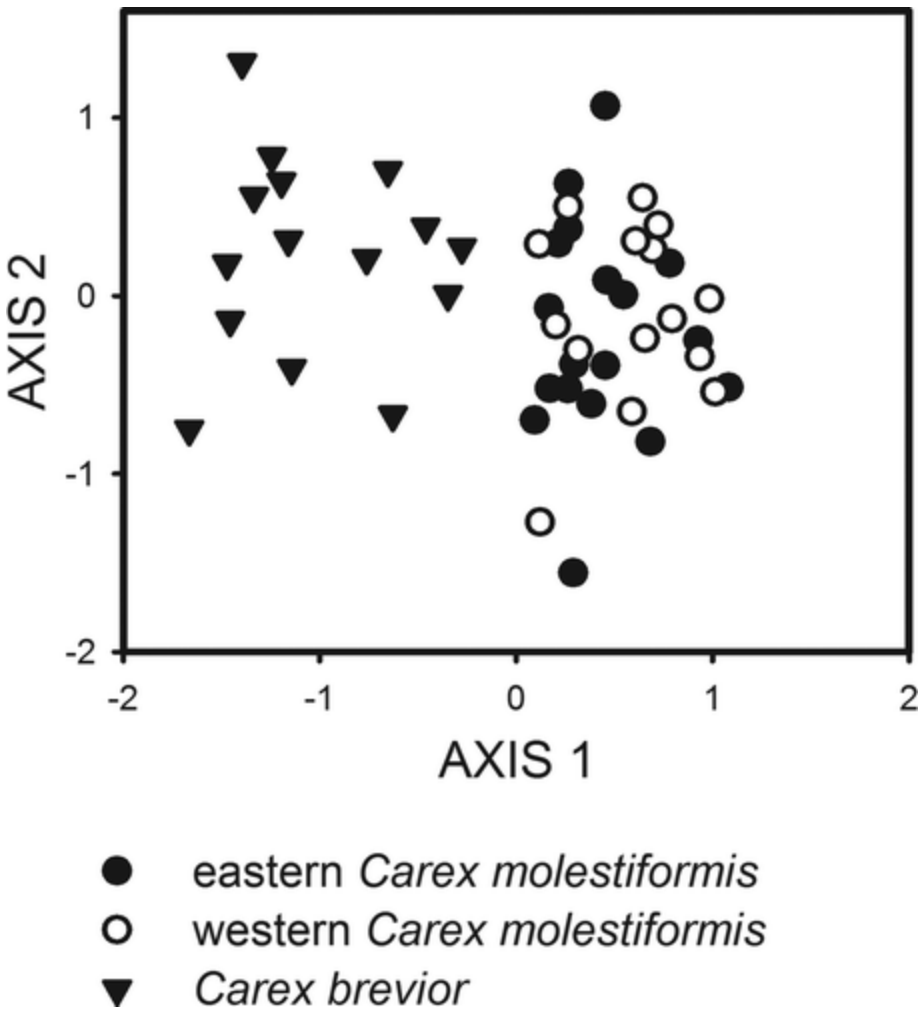


Figure 2 NMS plot for *Carex molestiformis* from east (18 specimens) and from west (15 specimens) of the Mississippi River and for *C. brevior* (16 specimens).

Previously the chromosome number for *C. molestiformis* was reported as $n = 37$ based upon plants from three localities in Arkansas and Oklahoma (Table 1). In this study, five additional plants from three localities east of the Mississippi were counted. The individual from Cumberland County, Kentucky, had an $n = 37$, identical to plants from farther west. On the other hand, the pairs of plants from Rockcastle County, Kentucky, and Jackson County, Tennessee, had $n = 35$ suggesting some karyotypic differentiation with geographical distribution. No individuals with $n = 36$ were encountered. By contrast, plants of the sister species *C. brevior* have a long agmatoploid series of $n = 24, 26, 28, 30, 32, 34$ (Rothrock and Reznicek 1998) but do not overlap with chromosome numbers for *C. molestiformis*. Another related species, *C. molesta*, has $n = 34$ or 35 (Reznicek and Rothrock 1997). Although these new counts demonstrate that *C. molesta* and *C. molestiformis* have some overlap in chromosome number, the

differences in external morphology, especially of the achene (Reznicek and Rothrock 1997), remain consistently different.

Table 1 Chromosome counts for *Carex molestiformis*

n	State, County	Greenhouse Voucher/Reference
35	KY, Rockcastle	SE of Mount Vernon, in median of I-75 just S of mile 58 (ca. mile 57.8), 37°19.667′N and 84°18.217′W (Reznicek 10617). 15 June 1998. *P.E. Rothrock 3728* (MICH)
35	TN, Jackson	Jackson County Airport, N of Gainsboro, along E side of Big Bottom Road, 36°23.783′N and 85°38.617′W (Reznicek 10632). 16 June 1998. *P.E. Rothrock 3729* (MICH)
37	KY, Cumberland	Ca. 5.5 mi W of Burkesville, along W side of North Hollow Road, 2.6 road miles N of its jct. with route 1205, 36°47.717′N and 85°28.867′W (Reznicek 10639). 16 June 1998. *P.E. Rothrock 3730* (MICH)
37	AR, Garland	Reznicek and Rothrock (1997)
37	AR, Pulaski	Reznicek and Rothrock (1997)
37	OK, Mayes	Reznicek and Rothrock (1997)

Acknowledgments

We are grateful to the curators of all the herbaria that we scoured for specimens of *Carex molestiformis* and especially to Alan Weakley (NCU) and Tom Wieboldt (VPI), who helped with data requests. Rob Naczi accompanied A.A.R. on field work in Kentucky and Tennessee, and Carl Rothfels kindly allowed us to cite his collection of *C. molestiformis* from North Carolina.

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Appendix. Specimens Used in Morphometric Analysis

Carex brevior (Dewey) Mack.: Ontario (Hastings) *Oldham 15353* (MICH); Ontario (Lambton) *Dodge s.n. in 1899* (MICH); Ontario (Thunder Bay) *Oldham 18906* (MICH); Connecticut (Litchfield) *Hermann 11879* (MICH); Illinois (Fayette) *Reznicek 10198* (MICH); Illinois (Winnebago) *Fell 52–347* (MICH); Iowa (Page) *Wilson 1300* (MICH); Kansas (Coffey) *Morse 3052* (MICH); Kansas (Stafford) *Morse 254* (MICH); Missouri (Boone) *McKenzie 1600* (MICH); North Carolina (Durham) *Blomquist 13467* (DUKE); Ohio (Ottawa) *McCormac 2547* (MICH); Texas (Rusk) *Reznicek 10400* (MICH); Vermont (Chittenden) *Gilman 92129* (MICH); Virginia (Goochland) *Fleming 9093* (VPI); Wisconsin (Grant) *Hermann 8777*(MICH).

Carex molestiformis Reznicek & P. Rothr.: Arkansas (Baxter) *Hyatt 5342* (MICH); Arkansas (Boone) *Hyatt 3848.05* (MICH); Arkansas (Fulton) *Hyatt 5489* (MICH); Arkansas (Marion) *Hyatt 5602* (MICH); Arkansas (Marion) *Demaree 64832* (MICH); Arkansas (Polk) *Reznicek 9368* (MICH); Arkansas (Pulaski) *Rothrock 2901* (MICH); Arkansas (Scott) *Reznicek 9371* (MICH); Arkansas (Searcy) *Hyatt 6463* (MICH); Arkansas (Sharp) *Hyatt 7149* (MICH); Arkansas (Stone) *Hyatt 5454* (MICH); Arkansas (Washington) *Reznicek 9793* (MICH); Georgia (Murray) *Rothrock 4817* (MICH); Kentucky (Cumberland) *Reznicek 10639* (MICH); Kentucky (Madison) *Mears s.n. in 1995* (MICH); Kentucky (Rockcastle) *Reznicek 10639* (MICH); Mississippi (Marshall) *Bryson 23214* (MICH); North Carolina (Alleghany) *Rothfels 2917* (MICH); Ohio (Hocking) *Reznicek 11572* (MICH); Oklahoma (Adair) *Reznicek 9785* (MICH); Oklahoma (Delaware) *Reznicek 9770* (MICH); Oklahoma (McCurtain) *Reznicek 9357* (MICH); Tennessee (Jackson) *McNeilus 92.421* (MICH); Tennessee (Jackson) *Reznicek 10632* (MICH); Virginia (Albemarle) *Stevens 3766* (VPI); Virginia (Albemarle) *Stevens 7072* (VPI); Virginia (Albemarle) *Stevens 25,354* (MICH); Virginia (Augusta) *DeGormo & Massey s.n. in 1939* (VPI); Virginia (Bath) *Wieboldt 6406* (VPI); Virginia (Cumberland) *Fleming 7935* (VPI); Virginia (Madison) *Fleming 5515* (VPI); Virginia (Page) *Bradley 19088* (NCU); Virginia (Wise) *Wieboldt 10198* (VPI).