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## Karyotype designation and habitat description of the northern short-tailed shrew (*Blarina brevicauda*, Say) from the type locality

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### ABSTRACT

The karyotype of the northern short-tailed shrew (*Blarina brevicauda*) has been studied extensively across its range, revealing polymorphisms as a result of Robertsonian fissions/fusions. However, no karyological data have been reported at the type locality due to disagreement on its location. This information is vital given the proximity of a contact zone with the Elliot's short-tailed shrew (*B. hylophaga*). Recent evidence has indicated that the type locality of *B. brevicauda* is situated at the western edge of this species' distribution in eastern Nebraska. Therefore, it is now possible to establish a karyotype of specimens found at this location. Specimens were collected in 2006 at the type locality and field karyotyped. Chromosome morphology was consistent with previous reports. Two specimens collected from the type locality had a diploid number of 50 and a fundamental number of 48. Further investigation may reveal additional karyotypes at this location.

### INTRODUCTION

The northern short-tailed shrew (*Blarina brevicauda*) is a common and widespread inhabitant of the eastern United States and southern Canada. At its southern borders, it comes into contact with its congeners, the southern short-tailed shrew (*B. carolinensis*) and the Elliot's short-tailed shrew (*B. hylophaga*), which are similar to *B. brevicauda* in external and cranial morphology (Benedict 1999a, 1999b; Bowles 1975, 1977; Brant and Ortí 2002; Braun and Kennedy 1983; Ellis et al. 1978; Genoways and Choate 1972; Graham and Semken 1976; Jones et al. 1984; Jones 1964; Moncrief et al. 1982; Tate et al. 1980; Thompson 2008). However, chromosome morphology (karyology) is one of the most reliable methods for distinguishing species of short-tailed shrews (*Blarina*).

The first karyotype for *B. brevicauda* was reported by Meylan (1967) based on 21 specimens of the subspecies *B. b. talpoides* from Ontario, Canada. Those specimens formed two distinct karyotypic groups with the larger group ( $n = 16$ ) possessing a diploid number ( $2n$ ) of 50, which consisted of 48 acrocentric autosomes and two metacentric sex chromosomes (i.e., a large X and a very small Y). The second group ( $n = 5$ ) had a  $2n$  of 49 with one large, submetacentric autosome and 46 acrocentric autosomes. This polymorphism appeared to be the

result of Robertsonian fission/fusion of two acrocentric autosomes, with both groups having a fundamental number (FN) of 48.

Lee and Zimmerman (1969) also reported the karyotype of *B. brevicauda*. They examined specimens ( $n = 53$ ) of the subspecies *B. b. kirtlandi* from central Illinois and confirmed the diploid numbers of 50 ( $n = 46$ ) and 49 ( $n = 6$ ), as reported by Meylan (1967). In addition, one specimen was found with a  $2n$  of 48. By combining their results with those of Meylan (1967), a ratio of 84:15:1 for the diploid numbers of 50, 49, and 48 was found, respectively.

Genoways et al. (1977) found similar results in specimens of the subspecies *B. b. brevicauda* ( $2n = 50$  or 49, FN = 48) from Nebraska. However, the sex chromosomes consisted of a large metacentric X-chromosome and a small acrocentric Y-chromosome. In addition, the Robertsonian polymorphism in diploid number was the result of a fission/fusion of two small acrocentric autosomes as seen by Meylan (1967), as well as a fusion of two large acrocentric autosomes. Although a  $2n$  of 48 was not observed directly (Lee and Zimmerman 1969), a combination of both fission/fusion events would constitute this result (Genoways et al. 1977).

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A comprehensive examination of the karyology of the genus *Blarina* was completed by George et al. (1982), with specimens being karyotyped from throughout the distribution of *B. brevicauda*. However, because the exact site of the type locality of *B. brevicauda* has been disputed (see, for example, Jackson 1961; James 1823; and Jones 1964), George et al. (1982) were not able to report the karyotype of animals from the type locality. Recent archeological research (Carlson et al. 2004; Genoways and Ratcliffe 2008) has established the type locality at the site of Engineer Cantonment (Nebraska: Washington Co.; 3 miles S and 4 miles E of Fort Calhoun 41.71194° N, 95.95017° W). During 1819-1820, Engineer Cantonment was the winter camp of U.S. Army Major Stephen H. Long's expedition (Say 1823). This expedition initially was charged with documenting the natural resources, physical geography, and Native American tribes of the upper Missouri River region (Woodman 2009).

Engineer Cantonment is notable as the type locality of several species of plants, invertebrates, and vertebrates.

The type locality of *B. brevicauda* lies about 67 km north of a contact zone with another member of the genus, *B. hylophaga* (Bowles 1975, 1977; Thompson 2008), which has been shown to shift in geographic position (Benedict 1999b). Thus, it is important to establish the characteristics of northern short-tailed shrews occurring at the type locality, because if future shifts in distribution occur, the true identity of *B. brevicauda* will have been established without question. The goal of this paper is to present the karyology of *B. brevicauda* collected from the type locality of this species and to compare these karyotypic data with those of other populations of *Blarina*. In addition, we describe the current habitat at the type locality of *B. brevicauda* and compare our observations to historic descriptions of this site.

## MATERIALS AND METHODS

*Specimen collection.*— On 1 October 2006, 120 Sherman live traps were set in a roadside borrow ditch at the type locality for *B. brevicauda*. Eighty traps were set in the west ditch, and 40 were set in the east ditch. All traps were baited with sunflower seeds and checked in the morning. Individuals that were collected alive were used for karyotyping. All specimens of *B. brevicauda* were retained as voucher specimens in the Museum of the High Plains (MHP) at Fort Hays State University in Hays, Kansas. The trapping protocol followed the guidelines of the American Society of Mammalogists

(Gannon et al. 2007).

*Field karyotyping.*— Karyotypes were processed as described by Baker et al. (2003) and detailed by Thompson (2008). Cells were located with a Nikon: Alphaphot YS microscope (Nikon Inc., Melville, New York). Diploid and fundamental numbers of chromosomes were counted for each cell found. Cells were photographed with a Coolpix 950 digital camera (Nikon Inc.). Five to 10 cells were photographed for each slide, except when fewer than 5 cells were found or in the event that the cells were not consistent (Baker et al. 1989; Sullivan et al. 1986).

## RESULTS

*Current habitat at type locality.*— Three *B. brevicauda* were trapped in the western roadside ditch of North River Drive road that now exists at the type locality. The elevation of this site is 305 m. The predominant vegetation found in the ditch was prairie cordgrass (*Spartina pectinata*), reed canary grass (*Phalaris arundinacea*), smooth brome grass (*Bromis intermis*), and several species of forbs. To the west were steep hills covered with trees and other woody vegetation. To the east, the habitat changed to a low-lying grassland, which included native, warm season grasses such as big bluestem (*Andropogon gerardii*), Indian grass (*Sorghastrum nutans*), reed canary grass, and switch grass (*Panicum virgatum*). This grassland is part of the Boyer Chute National Wildlife Refuge.



2N = 50, FN = 48

**Fig. 1.** Karyotype of *Blarina brevicauda* (MHP 37241) collected from type locality at the site of the Engineer Cantonment (Nebraska: Washington Co., 3 miles S and 4 miles E of Fort Calhoun, 41.71194° N, 95.95017° W).

*Description of karyotypes.*— All individuals collected were female; therefore, no information on the morphology of Y-chromosome can be presented. Only two specimens had scorable karyotypes (MHP 37240-

37241). Each karyotype had a  $2n$  of 50 and a FN of 48 (see Fig. 1, MHP 37241). Specifically, both of these animals had 48 acrocentric autosomes and two large, metacentric X-chromosomes.

## DISCUSSION

*Habitat comparison at type locality.*—The landscape surrounding the type locality has changed considerably since the holotype was obtained at the winter camp (Engineer Cantonment) of Stephen H. Long's expedition. Genoways and Ratcliffe (2008) provided a detailed description of the surrounding landscape, but in general, the camp was located at the eastern base of a series of hills and cliffs. The location also marked the edge of the Missouri River. The type locality was only a few meters from the edge of an oxbow off the main river channel.

Since the holotype specimen was obtained, the cabins used by the cantonment have disappeared. Today, a grain bin and a few small storage buildings are present at the site. The Missouri River has been channelized, and the oxbow that was adjacent to the campsite no longer is present. In its place is a low-lying grassland. A paved blacktop road (i.e., North River Drive) and bike trail run north-south between the hills and grassland. The steep hills to the west, which historically were covered in grass, remain but now are covered in woody vegetation. Much of the surrounding lowland habitat, which once was wetland, has been drained and converted to farmland. Finally, urban development is evident in the surrounding landscape. This development includes residential housing of the metropolitan city of Omaha, Nebraska, which is encroaching from the west and south.

*Comparison of karyotypes.*—Shrews (Soricidae) are known for considerable interspecific and intraspecific karyotypic variability (Ford et al. 1957; Sharman 1991). The genus *Blarina* follows this trend, exhibiting a high number of Robertsonian rearrangements and non-Robertsonian additions and deletions. Species within the genus tend to have higher diploid numbers than fundamental numbers (Zima et al. 1998). These karyotypic patterns within *Blarina* are similar to those of its tribal relative, the small-eared shrews (*Cryptotis*, Zima et al. 1998). The most similar *Cryptotis*, the least shrew (*C. parva*), has a similar distribution to members of the genus *Blarina* and occurs sympatrically at the type locality of *B. brevicauda*. Genoways et al. (1977) reported a  $2n$  of 52 and a FN of 50 for a specimen of

*C. parva*. This karyotype has acrocentric autosomes with a large, metacentric X-chromosome and a small, subtelocentric Y-chromosome.

George et al. (1982) identified 4 chromosomal groups within the genus *Blarina*. The first group corresponded to *B. carolinensis*, which had a  $2n$  of 46 and FN of 44 throughout most of its geographic distribution. However, the second group consisted of two subspecies of *B. carolinensis*: *B. c. minima* in southwestern Tennessee ( $2n = 40, 39, 38, 37, 36, 35$ , or  $34$ , FN = 45, 44, 43, 42, or 41; Beck et al. 1991; Elrod 1992; Elrod et al. 1996; George et al. 1982; Qumsiyeh et al. 1997) and *B. c. peninsulae* in Florida ( $2n = 52, 51$ , or  $50$ , FN = 52; George et al. 1982). The karyotype of the first group had all acrocentric autosomes with a large, metacentric X-chromosome and a small, acrocentric Y-chromosome. *Blarina carolinensis peninsulae* was similar to other *B. carolinensis*, except for the presence of a small pair of metacentric autosomes. On the other hand, *B. c. minima* had variable autosomes. In total, *B. c. minima* had 10 banded autosomes, which was much different than its congeners (George et al. 1982).

The third karyotypic group represented *B. hylophaga* (George et al. 1982). This species forms a contact zone with *B. brevicauda* (Benedict 1999a, 1999b; Bowles 1975, 1977; Genoways and Choate 1972; Thompson 2008) about 67 km south of the type locality of *B. brevicauda*. *Blarina hylophaga* has a  $2n$  of 52 and FN of 62, 61, or 60 (Genoways et al. 1977; George et al. 1982). There are 19 pairs of acrocentric autosomes, 5 pairs of metacentric autosomes, and a polymorphic pair, which are completely metacentric, completely acrocentric, or both (George et al. 1982).

The fourth group reported by George et al. (1982) was *B. brevicauda*. The karyotype throughout the range of *B. brevicauda* has a  $2n$  of 50, 49, or 48 and FN of 48. The morphology of the karyotypes described herein is typical for *B. brevicauda* (Genoways et al. 1977; George et al. 1982; Lee and Zimmerman 1969; Meylan 1967). As presented by earlier authors (Genoways et al. 1977; George et al. 1982; Lee and Zimmerman 1969; Meylan 1967), these specimens had the most prevalent diploid number ( $2n = 50$ ) found within this species. In addition,

they both had the diagnostic FN of 48 established by previous research (Genoways et al. 1977; George et al. 1982; Lee and Zimmerman 1969).

Although we found no variation between the two karyotypes for *B. brevicauda* from this species' type locality, the Robertsonian polymorphism scenarios that have been presented by others (Genoways et al. 1977; Lee and Zimmerman 1969; Meylan 1967; Oshida

et al. 2003) seem possible given the simplicity of the karyotype. In addition, across the Missouri River, Thompson (2008) found karyotypic polymorphism within the species in Harrison County, Iowa. Therefore, we predict that further fieldwork in the vicinity of the type locality of *B. brevicauda* will reveal additional variability among karyotypes of animals collected in this area.

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