

Short Communication**Introduced alien ringed crayfish (*Orconectes neglectus neglectus* [Faxon, 1885]) threaten imperiled coldwater crayfish (*Orconectes eupunctus* Williams, 1952) in the Eleven Point River drainage, Missouri, USA**

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Abstract

The Eleven Point River drainage in the Ozarks ecoregion of North America contains the largest known populations of and critical habitat for the globally imperiled coldwater crayfish (*Orconectes eupunctus*). We present here the discovery of an introduced population of alien ringed crayfish (*Orconectes neglectus neglectus*) in Jolliff Spring Branch in the upper reaches of the drainage. Sampling was conducted at eight sites throughout the tributary to determine the extent of the alien species' range in the stream. *Orconectes n. neglectus* were found at four sites extending along 3.4 km of stream, with reproducing populations confirmed at two upstream sites, and few individuals found at two others. This invasion represents a threat to *O. eupunctus*, which occurs a few kilometers further downstream in the drainage and has been recorded from only the lower reaches of Barren Fork and throughout the mainstem Eleven Point River. Introduced *O. neglectus* were previously associated with localized extirpation of *O. eupunctus* in another drainage. Study results warrant monitoring of the invasion and possible reassessment of the conservation status for *O. eupunctus*.

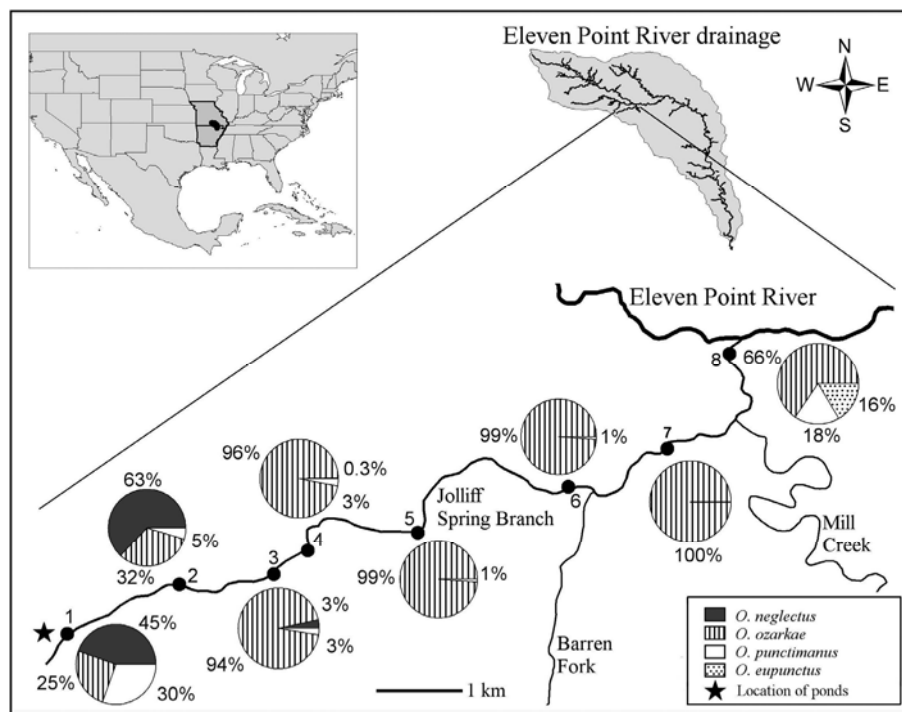
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Nearly half of the native crayfish fauna in the United States and Canada are considered imperiled or in need of conservation (Taylor et al. 2007; NatureServe 2010), and the introduction of invasive alien crayfish has been cited as the most significant threat to crayfish biodiversity in North America and worldwide (Lodge et al. 2000). Invasive crayfishes have displaced native crayfishes (Lodge et al. 1986; Light et al. 1995; Daniels 1998); adversely affected or displaced native amphibians (Gamradt and Kats 1996; Kats and Ferrer 2003), reptiles (Fernandez and Rosen 1996), and fishes degraded recreational and commercial fisheries (Hobbs et al. 1989; Guan and Wiles 1997; Dorn and Mittelbach 1999; Wilson et al. 2004; Peay et al. 2009); and altered the structure and function of stream, lake and marsh communities (Feminella and Resh 1989; Olsen et al. 1991; Charlebois and Lamberti 1996).

The ringed crayfish, *Orconectes neglectus neglectus* (Faxon, 1885; Figure 2), is native to streams in southwestern Missouri as well as

portions of Arkansas, Colorado, Kansas, Nebraska, Oklahoma and Wyoming (Taylor et al. 2007). Alien populations of this species are established in New York (Taylor et al. 2007) and in the Rogue River of Oregon, where they have displaced the native Klamath signal crayfish, *Pacifastacus leniusculus klamathensis* (Stimpson, 1857) (Bouchard 1977). An introduced population of *O. n. neglectus* was also recently documented in Little Tebo Creek, a tributary to Harry S. Truman Reservoir (Osage River) in Missouri (Jones 2009). The subspecies, the gap ringed crayfish *Orconectes neglectus chaenodactylus* Williams, 1952, was introduced and appears to be displacing two native crayfishes including the "Vulnerable" (DiStefano et al. 2010) and "Threatened" (Taylor et al. 2007) coldwater crayfish, *Orconectes eupunctus* Williams, 1952 (Figure 3), in the Spring River of Missouri and Arkansas (Magoulick and DiStefano 2007). During June 2010, sampling efforts to locate *O. eupunctus* populations found *O. n. neglectus* in the upstream reaches of Jolliff

Figure 1. Map of Jolliff Spring Branch and Barren Fork. Black circles along streams indicate sampling sites 1 through 8. Pie charts show percentage of the total catch at each site made up by each species. Location of ponds sampled by trapping is shown with a star.



Spring Branch, a small, 8.2 km, spring-fed Missouri stream located in the northern portion of the Eleven Point River drainage. The stream flows directly into Barren Fork, which harbors the imperiled *O. eupunctus*.

Our objective was to survey Jolliff Spring Branch and the downstream reaches of Barren Fork thought to harbor *O. eupunctus*, to determine the extent of the *O. n. neglectus* invasion. Results will provide managers with information pertinent to conservation status designations for native species including *O. eupunctus*, and a baseline for future monitoring of expansion and abundance of *O. n. neglectus*.

Jolliff Spring Branch averages approximately 3 m in width (1-8 m range) and the downstream reaches of Barren Fork average 6 m in width (3-9 m range); both streams are generally <1 m in depth. Land use surrounding the streams is generally livestock pasture with forested riparian corridors. Eight sampling sites were selected at regular intervals corresponding to stream access points, throughout the length of Jolliff Spring Branch and the reach of Barren Fork between its confluences with Jolliff Spring Branch and the Eleven Point River (Table 1; Figure 1). Additionally, we sampled three small ponds

located immediately north of site 1 to determine if they might be possible sources of the invasion.

Each study site was sampled once from June 24 through October 8, 2010, during low stream flows when our sampling method was most effective and consistent. Eight habitat sampling units (four riffles and four runs) were selected at each site, and upstream and downstream ends of the site were marked with a Global Positioning System unit. A random number table was used to select locations for three kick-seine subsamples within each habitat sampling unit (total $n = 4$ samples per habitat type per site). Crayfish species and densities were determined by disturbing the substrate inside a 1 m² pvc quadrat frame located directly upstream of a kick seine (1.5 m length \times 1.5 m height) with 3 mm delta mesh (Flinders and Magoulick 2005). We identified crayfish to species and returned them to the stream unharmed.

Three ponds adjacent to site 1 (Figure 1) were sampled with wire mesh minnow traps baited with canned dog food. This method has been used previously to collect *O. neglectus* and typically outperforms kick seining methods in deeper lentic environments. Traps were set in the evening, evenly spaced along the shoreline about



Figure 2. *Orconectes neglectus neglectus*. Photograph by Chris Lukhaup.



Figure 3. *Orconectes eupunctus*. Photograph by Chris Lukhaup.

Table 1. Locations of sampling sites and dates on which sampling occurred.

Site	Latitude	Longitude	Date sampled
1	36°44'37"N	91°35'59"W	24 June 2010
2	36°44'60"N	91°35'07"W	25 September 2010
3	36°45'16"N	91°34'08"W	8 October 2010
4	36°45'04"N	91°34'23"W	18 September 2010
5	36°45'24"N	91°33'17"W	18 September 2010
6	36°45'45"N	91°32'07"W	26 September 2010
7	36°46'03"N	91°31'21"W	25 September 2010
8	36°46'47"N	91°30'53"W	12 July 2010

Table 2. Total numbers and mean densities of crayfish (number of crayfish/m² ± 95% confidence interval) found in each habitat type at each site. Sites are listed numerically (1 through 8) from upstream to downstream. Crayfish species are coded as follows: NE = *O. n. neglectus*, OZ = *O. ozarkae*, PU = *O. punctimanus*, EU = *O. eupunctus*.

Site	Habitat Type	NE	OZ	PU	EU
1	Riffle	35 (2.92 ± 4.18)	9 (0.75 ± 0.91)	16 (1.33 ± 2.52)	0
	Run	63 (5.25 ± 5.45)	46 (3.83 ± 4.17)	51 (4.25 ± 4.93)	0
2	Riffle	71 (5.92 ± 2.46)	9 (0.75 ± 1.09)	1 (0.08 ± 0.27)	0
	Run	50 (4.17 ± 2.05)	53 (4.42 ± 2.14)	8 (0.67 ± 0.87)	0
3	Riffle	3 (0.25 ± 0.80)	32 (2.67 ± 2.90)	0	0
	Run	1 (0.08 ± 0.27)	79 (6.58 ± 5.00)	3 (0.25 ± 0.51)	0
4	Riffle	1 (0.08 ± 0.27)	64 (5.33 ± 1.68)	1 (0.08 ± 0.27)	0
	Run	0	188 (15.67 ± 10.35)	7 (0.58 ± 0.91)	0
5	Riffle	0	57 (4.75 ± 2.09)	1 (0.08 ± 0.27)	0
	Run	0	108 (9.00 ± 5.86)	1 (0.08 ± 0.27)	0
6	Riffle	0	47 (3.92 ± 4.27)	0	0
	Run	0	41 (3.42 ± 2.95)	1 (0.08 ± 0.27)	0
7	Riffle	0	17 (1.42 ± 1.91)	0	0
	Run	0	20 (1.67 ± 1.98)	0	0
8	Riffle	0	16 (1.33 ± 2.45)	1 (0.08 ± 0.27)	5 (0.42 ± 0.67)
	Run	0	24 (2.00 ± 1.56)	10 (0.83 ± 1.02)	5 (0.42 ± 0.51)

2-3 m from shore, and harvested the following morning. Thirty traps were placed in the largest of the ponds (~ 1400 m²), and ten and six traps were placed in two smaller ponds (< 1100 m² each).

Orconectes n. neglectus populations were established in the upstream-most reaches of Jolliff Spring Branch, but had not yet occupied the entire stream. *Orconectes n. neglectus* were found at four sites spanning the upstream 3.4 km of the stream (Table 2; Figure 1). The species appeared to be well-established only at sites 1 and 2, where it constituted 45% and 63% of the crayfish community, respectively, and occurred at higher densities than the native Ozark crayfish (*Orconectes ozarkae* Williams, 1952) and spothanded crayfish (*Orconectes punctimanus* [Creaser, 1933]). Very few *O. n. neglectus* individuals were found (four and one) at sites 3 and 4, and none were found at sites 5 through 8. *Orconectes n. neglectus* and *O. eupunctus* did not occur in sympatry; their apparent

distributions were separated by about 7.6 km of stream length. No crayfish were captured in any of the three sampled streamside ponds, and we know from previous work that *O. n. neglectus* (DiStefano et al. 2009), *O. punctimanus* (DiStefano and Westhoff 2011) and *O. ozarkae* (DiStefano RJ, personal observation) are all susceptible to baited trapping.

Our discovery of *Orconectes n. neglectus* in the upstream reaches of Jolliff Spring Branch is the first record of this species in the Eleven Point River drainage. The presence of adult and juvenile specimens indicates an established reproducing population. The downstream leading edge of the *O. n. neglectus* invasion is difficult to determine because the few individuals found at sites 3 and 4 may represent established populations or merely dispersing and colonizing individuals. It is also possible that *O. n. neglectus* were present downstream of site 4 but our sampling failed to detect them. However, the invasion may expand downstream toward

populations of imperiled *O. eupunctus* in Barren Fork and the Eleven Point River. Revisiting our study sites in the future can confirm downstream expansion of *O. n. neglectus* and evaluate changes in crayfish abundance and relative community composition over time.

The source and timing of the invasion is unknown. “Baitbucket introductions” by recreational fishers are generally considered the primary vector for alien crayfish introductions in North America (Lodge et al. 2000), but *Orconectes n. neglectus* is not on the state of Missouri’s Approved Aquatic Species List (AASL; State of Missouri 2011) for commercial trade, and thus not legally sold in fishing bait shops. However, 62% of Missouri’s known crayfish invasions involve species not on the AASL (R.J. DiStefano, unpublished data). Missouri fishing license holders are permitted to catch wild crayfish for several uses (bait, human consumption, pets, etc.; State of Missouri 2011), and we suspect that wild-caught crayfish are being transported across drainage basin boundaries and released to the wild. Our data suggest that *O. n. neglectus* were introduced in the vicinity of sites 1 and 2, possibly directly to the stream at a lone public road crossing immediately upstream of site 1 or into recreational fishing ponds at site 1, or into additional ponds (which we were unable to sample due to lack of landowner permission) at site 2.

The imperiled *O. eupunctus* is one of the world’s most geographically limited crayfishes, endemic to only portions of the Spring, Strawberry, and Eleven Point rivers of southern Missouri and northern Arkansas (Pfleiger 1996). Population declines and localized extirpations of *O. eupunctus* and native Hubbs’ crayfish, *Cambarus hubbsi* (Creaser, 1931) in many kilometers of the nearby Spring River drainage have followed a previous invasion by the *O. n. chaenodactylus* subspecies (Magoulick and DiStefano 2007). Magoulick and DiStefano (2007) concluded that *O. n. chaenodactylus* occupied similar habitats as the two native species. In 2010 the Missouri Department of Conservation, Arkansas Game and Fish Commission, and United States Geological Survey Arkansas Cooperative Fish and Wildlife Research Unit initiated an ongoing comprehensive distribution and population genetics survey for *O. eupunctus* in response to this perceived threat. Based on preliminary results of that survey, previous *O. eupunctus* collections,

and the documented *O. eupunctus* population declines in the Spring River drainage, the Eleven Point River drainage is now considered by these agencies to contain the largest populations of and important habitat for *O. eupunctus* (R. J. DiStefano, personal observation). These agencies are also concerned about the potential threat of this invasion to another “critically imperiled” (Missouri Natural Heritage Program 2011) native crayfish, the Mammoth Spring crayfish (*Orconectes marchandi* Hobbs, 1948) that occurs in the Spring River drainage, close to the Eleven Point River. Therefore, the discovery of the *O. n. neglectus* invasion in Jolliff Spring Branch, in the Eleven Point River drainage, is cause for concern.

Orconectes n. neglectus dominated the crayfish community at sites 1 and 2. Densities of the native *O. ozarkae* at these two sites were significantly lower than at several downstream sites, but we have no direct evidence that the invader has negatively affected this native species or the native *O. punctimanus*. Our study establishes a baseline for potential comparisons between future sampling results and the crayfish densities we reported.

The *O. n. neglectus* invasion in Jolliff Spring Branch poses an increased threat to the conservation status of the imperiled *O. eupunctus*. Previous studies indicate that crayfish invasions in upland streams progress fairly rapidly downstream (Peay et al. 1999; Bubb et al. 2005; Kerby et al. 2005); those studies as well as the *O. n. chaenodactylus* invasion in the nearby Spring River (Magoulick and DiStefano 2007) suggest that *O. n. neglectus* in Jolliff Spring Branch could expand its range and enter the reach of Barren Fork occupied by *O. eupunctus* within a decade. The Jolliff Spring Branch *O. n. neglectus* invasion should be monitored closely and the data reported here should be considered in future conservation status assessments for the imperiled *O. eupunctus*.

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