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First report of Amynthas carnosus (Goto & Hatai, 1899) (Oligochaeta: Megascolecidae) in the Western Hemisphere

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The earthworm fauna of North America is still greatly unknown, with undescribed native species and new records of invasive species continually being discovered. The most recent checklists are difficult to reconcile, since they vary in their geographic coverage and taxonomy. Reynolds & Wetzel (2012) state that North America (including all of Mexico, some of the Caribbean, and Hawaii) is home for 256 species of earthworms, from which 188 are considered native or endemic and 68 are peregrine, exotic, and invasive species. Blakemore (2008) listed North America north of Mexico as having 183 earthworm species, of which 59 species are exotic. One Megascolecidae genus, Amynthas, is mostly located in the warmer subtropical and tropical regions of the world, with a few species that are able to survive the winter in the temperate zones of North America (Edwards & Bohlen 1995; Görres et al. 2014). Here we report for the first time the occurrence of Amynthas carnosus (Goto & Hatai, 1899) in the United States. To our knowledge, this is not only the first report of A. carnosus in North America, but also its first report in the Western Hemisphere. The description below is intended to place our specimens in the correct taxon while avoiding future confusion with similar species, without making changes to the existing classification system.

The material analyzed included five adult earthworms, of over 30 individuals collected by Christopher Berry and Andrew Wendland near Tuttle Creek Observation Area, north of Manhattan, Riley County, Kansas, USA (39.2728°N, 96.6259°W). Collection was performed on 10 June 2015 by hand. The earthworms were found crawling on the surface of the soil after several weeks of near-record rainfall, which saturated the soil and caused a large increase in the water level of adjacent Tuttle Creek reservoir. All individuals were killed with 70% ethanol and fixed with 5% formalin for 4 days. Fixed individuals were then permanently stored in 70% ethanol and reside in Snyder's collection. External and internal characteristics of each individual were described and compared to existing literature. The specimens studied follow the redescription of A. carnosus by Blakemore (2012) within an acceptable range of variation for this species. Blakemore (2012) based his redescription on the fixation of a neotype but still allowed, following Kobayashi (1936), considerable morphological variation especially in the distribution of the genital markings. We also observed variations in the genital markings, but the general pattern in our specimens is very similar to that of the neotype of A. carnosus (see below). Subspecies are not considered here because they were erected mainly on differences in the COI barcode gene (Blakemore 2013a,b), and COI barcode sequences are not available for our specimens.

Amynthas carnosus (Goto & Hatai, 1899)

Perichaeta carnosa Goto & Hatai, 1899: 15, 24, Figs. 4, 5. Pheretima carnosa: Michaelsen 1900: 260-261. Amynthas carnosus: Sims & Easton 1972: 235. Blakemore 2012: 36-40, Fig. 1.

Description. Length 150–180 mm. Width 7 mm. Live specimens dark brown or purple dorsally, yellowish ventrally. Preserved specimens lost little pigment within the first six months of storage. Prostomium epilobic. Setae perichaetine. 16–19 setae between male pores. Four paired spermathecal pores just anterior to 5/6-8/9 in four of the earthworms. The fifth earthworm had three paired spermathecal pores from 6/7-8/9. First dorsal pore on 12/13. Clitellum at xiv-xvi, annular, cream-yellowish coloration in live and brown in preserved specimens. Two pairs of pre-clitellar genital markings pre-setal on viii and ix; pair on ix spaced further apart than the pair on viii (markings on ix more lateral, similar to the neotype as drawn in Blakemore 2012). Up to three pairs of genital markings near male pores on xviii. First pair pre-setal on xviii, slightly median to male pores; second pair post-setal and more medial than the first; third pair pre-setal

on xix, slightly more lateral than the second pair, but not as lateral as the first pair (Figure 1B). This arrangement is similar to Figure 3, Type V of post-clitellar genital markings in Blakemore (2012), but with the first and third pairs of genital marking slightly lateral to the second pair. All three pairs were present in two octothecal specimens. Two octothecal specimens had only three genital markings comprised of the post-setal xviii pair and a single marking in xix (left side in one specimen, right side in the other). The sexthecal specimen was only lacking the pre-setal xviii genital marking on the left side. Spermathecae in vi–ix, or vii–ix (in the specimen with only three pairs of spermathecal pores). Gizzard viii–ix. Intestinal origin xv. Intestinal caeca simple, beginning in xxvii and extending anteriorly for 3–4 segments. Prostate glands racemose, ducts in xviii, and extending anteriorly 1–2 segments and posteriorly 1–3 segments.

Following the keys by Reynolds (1978) or Gates (1982), specimens of *A. carnosus* might be misidentified as *A. corticis* Kinberg, 1867, *A. gracilis* Kinberg, 1867, or as *A. hupeiensis* Michaelsen, 1895. Additionally, Blakemore (2012, 2013b) reported that *A. carnosus* has been misclassified as *A. corticis* and *A. gracilis*. Figure 1 compares key external features of *A. corticis*, *A. carnosus*, and *A. gracilis*. Externally, octothecal *A. carnosus* might be differentiated from *A. corticis*, which is also octothecal, by the two pairs of genital markings usually median and anterior to the setal line at viii and ix and by the genital markings median to the male pores and sometimes extending to anterior xix (Blakemore 2012); *A. corticis* normally has three small paired genital markings on vii–ix adjacent to the spermathecal pores, and rarely median to the male pores (*cf.* Fig. 1A and 1B) (Gates 1972; Borges & Moreno 1990; Blakemore 2013b).

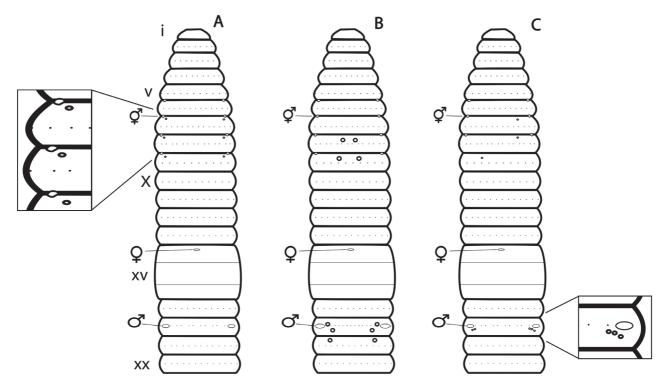


FIGURE 1. Diagram of pores and genital markings in a ventral view of A) *Amynthas corticis*, B) *A. carnosus*, and C) *A. gracilis*. $\mathcal{J} =$ male pores, $\mathcal{Q} =$ female pores, $\mathcal{Q} =$ female pores.

The sexthecal morph of *A. carnosus* has also been misidentified as *A. gracilis* in the past, due to similar genital markings median to the male pores and because *A. gracilis* is sexthecal. The spermathecal pores on *A. gracilis* are located on 5/6–7/8 (Gates 1972), while in the sexthecal morph of *A. carnosus* spermathecal pores are located at 6/7–8/9 (Blakemore 2012; our specimens). *Amynthas gracilis* has been reported to also have genital markings near the spermathecal pores, but not commonly paired and usually extending from vi–ix (Gates 1972, 1982). The genital markings on *A. gracilis* median to the male pores are arranged as clusters of 1–11 markings posterior to the setae (Gates 1972), while other authors reported fewer: one to three genital marks at each male pore (Borges & Moreno 1990). The genital markings in *A. gracilis* are smaller than those of *A. carnosus* (*cf.* Fig. 1B and 1C). Male pore-associated genital markings on *A. carnosus* are highly variable ranging from no genital markings up to 6 individual markings, which might not always be paired and might extend to be on the anterior part of xix (Blakemore 2012). The first dorsal pore on *A. gracilis* is usually at 10/11 (Gates 1972, 1982; Borges & Moreno 1990) and at 11/12 on *A. corticis* (Gates 1972; Borges & Moreno 1990), while in *A. carnosus* it is on 12/13 (Blakemore 2012). Specimens from North America previously classified as *A. corticis* or *A. gracilis* should be carefully reexamined to confirm their identity.

Amynthas hupeiensis is sexthecal with spermathecal pores in the same locations as *A. carnosus*, but is easily differentiated by the color of live specimens. *Amynthas hupeiensis* is usually green, while *A. carnosus* is brown-purple colored (Blakemore 2012). In addition, *A. hupeiensis* is usually smaller than *A. carnosus*.

Amynthas carnosus is known to have a wide range in Asia: it has been reported in Korea, Japan, and China (Blakemore 2003; 2012; 2013a; Blakemore *et al.* 2006; Blakemore & Lee 2013). Approximately 10–20 species of this genus are known to be peregrine species (Edwards & Bohlen 1995; Hendrix *et al.* 2008). As the vector for introduction of this species can only be speculated, it might have been used as fishing bait, as the area of collection is popular among anglers (C. Berry, pers. comm.). It also appears that *A. carnosus* might be well established in the area, as it was collected in a relatively high number. The potential of *A. carnosus* as a peregrine species is yet to be determined, and effects of its colonization are unknown. Close attention should be given to the ecological effects after the establishment of this species, due to the known effects of related species (e.g., Snyder *et al.* 2011; Greiner *et al.* 2012).

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