## Helodrilus hachiojii sp. nov. (Oligochaeta: Lumbricidae) from Japan

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**Abstract** A small (60-70mm) semi-aquatic or limnic lumbricid earthworm from a creek in Hachioji, western Tokyo appears new to science and is named after the locality. It is tentatively attributed to the genus *Helodrilus* Hoffmeister, 1845 – characterized by lack of nephridial bladders – rather than *Eisenia* Malm, 1877 that holds the only other endemic lumbricid, *E. japonica* (Michaelsen, 1891). The current species is close to type *Helodrilus oculatus* Hoffmeister, 1845 but differs on its clitellum in segments 23-31 with tubercula pubertatis in 29, seminal vesicles in 9-12, and intramural calciferous glands in 11-12 extending into 13-14. Further lack of spermathecae is distinctive, whereas *He. oculatus* is not known for parthenogenesis in its European headquarters. Restricted distribution suggests endemicity, however discovery of a specimen in a rice paddy at Lake Biwa indicates at least partial transportation. Alternatively, its apparent rarity and restriction may relate to common sampling omission of small species.

Key words: Earthworms, systematics, lumbricids, Eisenia, Eiseniella, Asia, biodiversity.

## Introduction

Studies on Japanese earthworms by the author (Blakemore, 2003; 2005; 2006a) involved attempts to recollect and neotypify historical species without types, e.g. those by Goto & Hatai (1889, 1890). Incidental to these surveys, several other species were unearthed and the specimens described below were obtained from Hachioji in 2004. As no similar description can be found, they are believed to be of a new species that is named herein. This brings the total Japanese fauna to 82 species (with a further ten *species incertae sedis*), of which about 50% are exotic and just a dozen are holarctic Lumbricidae and two of these, cosmopolitans *Dendrobaena pygmaea* (Savigny, 1826) and *Eiseniella tetraedra* (Savigny, 1826), are recent new records (Blakemore, 2004; Blakemore *et al.*, 2007).

## **Materials and Methods**

Specimens were transported to the laboratory in Yokohama where they were anaesthetized and preserved in 80% ethanol; formalin was avoided to facilitate possible DNA extraction at a later time. Dissection and sketching with camera lucida in the author's usual style follows taxonomic conventions given in (Blakemore, 2002, 2006b). Types are deposited in the Shinjuku Branch of the National Museum of Nature and Science (formerly the National Science Museum).

# Systematic results

## Helodrilus hachiojii sp. nov.

**Diagnosis:** Length 60-70mm. Lightly pigmented brown dorsum. Anterior body cylindrical, posterior flattened rectangular. Prostomium epilobous. First dorsal pore in furrow 4/5. Setae closely paired, in anterior at least. Spermathecal pores not found. Male pores confined to segment 15. Clitellum saddle-shaped on segments 23-31. Tubercula pubertatis in form of paired papillae on segment 29 lateral of setae b. Calciferous glands in segments 11-12 (and 13-14). Nephridia holoic, bladders (mostly) lacking. Spermathecae lacking.

**Material examined:** NSMT-An 371 – Holotype, H, clitellate mature - dissected and sketched (Fig. 1), NSMT-An 372-3 – Paratype Nos. P1-2 two sub-adults - dissected, NSMT-An 374-6 – P3-5 an aclitellate juvenile and two immatures all lacking tubercula pubertatis - inspected. A single specimen (non-type) from north of the lake deposited in Lake Biwa Museum (LBM), mature - undissected but externally it complies with *H. hachiojii*, LBM: 1380000005 ex Accession Batch Number FY2007-11.

**Locality:** from Komiya Park, Hachioji, western Tokyo ca. 35°40'N.139°19'E (collected by R.J.B. 22.x.2004). An additional specimen from moist mud of harvested rice paddy opposite convenience store at Shiotsuhama, Nishiazai-cho, northern Lake Biwa, Shiga-ken ca. N 35°22'.136°02' (collected by R.J.B. on survey with Dr Eiso Inoue 4.x.2007).

**Habitat:** in mud beside creek draining spring-fed moat of Otanibenzaiten shrine. [Location photos <u>www2.kankyo.metro.tokyo.jp/sizen/yuusui57sen/photo/21/21.htm</u> March, 2007]. Also moist soil of harvested rice paddy (northern Lake Biwa).

**Species associations:** from original collection site, small microdriles (tubificids?) and large pheretimoid megadriles – members of the *Amynthas corticis* (Kinberg, 1867)

species-complex *sensu* Blakemore, 2003. At Lake Biwa site several pheretimoids also present including *Metaphire hilgendorfi* (Michaelsen, 1892)/ *Amynthas tokioensis* (Beddard, 1892) species-complex.

Behaviour: limicolous (mud-dwelling); unresponsive or docile to touch (no thrashing).

**Lengths mm:** 69 (H), 60 (P1), 50 (P2), 55 (P3), 40 (P4), 30 (P5); range for adults 60-70mm. **Width:** 2.5-3.0mm.

**Body:** body cylindrical in anterior and dorso-ventrally flattened in final 30 or so segments of posterior to become almost rectangular in section with setal pairs at corners; segmental counts 105 (H), 116 (P1), 125 (P2), 108 (P3), 99+ (P5 posterior amputee?); range 100-125.

**Colour:** unpigmented pink with yellow clitellum and blood vessels and intestinal digesta visible through cuticle in life; anterior lightly pigmented brown, clitellum pale buff otherwise bleached white in ethanol.

Prostomium: closed epilobous.

First dorsal pore: rudimentary in furrow 3/4 (H), wide open from 4/5 (H, P1-2).

**Setae:** eight per segment from segment 2; small, black and closely paired in anterior with ratio aa:ab:cd:dd:U (U= circumference) approximately 4:1:3:1:14:0.5 (see figure); in posterior, gaps widen and in last 30 or so segments setae larger and opaque with ratio d:d less than half circumference; no setal tumescences noted.

Nephropores: inconspicuous (internally nephridia duct near ventral setal couples).

**Clitellum:** saddle-shaped to flanks below setal c-lines in segments 23-31 (H), not fully developed but some dorsal modification in (21,)22-30,31 in (P1), in Lake Biwa specimen, (23?)24-31; typical range thus 22,23-30,31.

Male pores: small, equatorial, non-tumid and confined to segment 15 in mid-bc lines.

**Female pores:** minute in segment 14; determined to be lateral of b setae from internal ducting.

Spermathecal pores: not found, absent.

**Genital markings:** tubercula pubertatis in form of a pair of large, discrete raised papillae ventrally on segment 29 just lateral to b setae (no glands correspond internally); no other banding found.

**Septa:** with some thickening in anterior to intersegment 13/14 thereafter thin; septum 17/18 to mid-gizzard reduced or aborted.

**Vascularization:** hearts paired in segments 7-11; dorsal blood vessel single; some vessels ventrally on calciferous glands, but not notably well supplied dorsally. Sub-neural vessel or oesophageal vessels not found.

**Calciferous glands:** large annular with deep intramural striations in segments 11-12 (striations continue to a lesser degree in 13-14); no extramural sacs nor diverticula.

**Intestines:** oesophageal valve in intersegment 14/15, large thin walled and dilated crop in segments 15-16, muscular gizzard large in 17-18 and just encroaching into 19, intestinal origin in 19, typhlosole simple lamellar commencing from about 22-23; caeca absent.

**Nephridia and vesicles/bladders:** holoic (i.e., two per segment) noted from segments 6 or 7 onwards (possibly small and concealed in oesophageal mass further forwards?); bladders generally absent but a few nephridia have simple flask-shaped bladders (at least in ca. 8rhs and 20lhs in H only, and some similar bladders also in one or two other segments in gregarine infested P2, but none seen in P1); pre-septal funnels with terminal ducts near ventral setal couples and no canalization to intestine. Nephridia are classed as mostly lacking bladders, i.e., avesiculate.

**Male organs:** vasa deferentia traced to non-iridescent funnels in segments 10 and 11, small testis possibly free in 11 (not clearly distinguished in 10); seminal vesicles paired: small and vestigial posteriorly in 9 and anteriorly in 11; larger posteriorly in 10 and anteriorly in 12; these characteristics imply infertile holandry with incipient metandry.

**Ovaries:** small paired saccular with funnels in segment 13; small or vestigial ovisacs paired anteriorly in 14.

Prostates: none.

Spermathecae: none found.

Longitudinal muscuature: not determined.

Gut contents: fine organic material and dark soil (mud) consistent with habitat.

Cocoons: none found.

**Parasites:** heavy infestation with gregarine *Monocystis* sp. cysts in body cavity of dissected (P2) and seen through body wall of (P3).

#### **Remarks:**

1. The specimens' location (mud), species-association with microdriles, nephridia reduced in anterior and lacking bladders, and the gut contents are all indicative of a semi-aquatic or

limicoline habitat; but presence of dorsal pores and the gizzard muscular rather than atrophied, as would be expected, suggest adoption of this habitat is relatively recent.

Non-iridescent testis/male funnels, absence of spermathecae or spermatophores, minute male pores and parasitism of reproductive organ segments are indicative of parthenogenesis.
 Small size, parthenogenicity with associated tolerance of higher parasite burden, high proportion of juveniles, and species association (e.g. cosmopolitan *Amynthas corticis*) in a locality disturbed by human activity (the moat was dug as an emergency water supply according to a local information board) and a rice paddy, all suggest exotic origin.

4. Its wide but narrow distribution (just two localities in Japan) and no other similar species on record suggest this species is new to the Japanese faunal list by transportation.5. Alternatively, its rarity and restriction may relate to sampling omission of small species.

#### Discussion

Although this species has several characteristics of a cosmopolitan, no description matching the current specimens was found from the literature despite lengthy searches. Certainly, it is not one of the 33-34 Lumbricidae that are common around the world (Blakemore, 2002, 2006b), neither does it correspond to any species yet known from Japan or adjacent countries (e.g. Easton, 1981; Perel, 1997; Blakemore, 2003, 2005). Thus it is given a name on the premise that it is a new species and in order to facilitate further eco-taxonomic study.

Generic placement is difficult. Some similarity is with aquatic *Eiseniella* Michaelsen, 1900 that has rectangular or quadrangular posterior segments [as also seen sometimes in *Bimastos parvus* (Eisen, 1874), *Dendrobaena octaedra* (Savigny, 1826) and *Eisenia balatonica* (Pop, 1943)], although having a gizzard restricted to segment 17 only is characteristic for *Eiseniella* genus (Michaelsen, 1900: 471; Gates, 1972). *Dendrobaena* Eisen, 1873 also has calciferous glands in 11-12 but it has widely paired setae and ocarina-shaped nephridial bladders. Another possibility is *Eisenia* Malm, 1877 that provides the only two other lumbricid species thought endemic to Japan and/or Korea, and although this genus allows similar calciferous glands in 11-12 and seminal vesicles in 9-12, the nephridial bladders are saccular or more extensive sausage-shaped (sometimes J-shaped?).

Only one lumbricid, *Eisenia japonica* (Michaelsen, 1891) originally obtained from Enoshima, Hakodate and "Fusi-jama" (Mt Fuji), was previously considered a Japanese endemic, but this is now also known from Korea, northern China and Sakhalin Island, Siberia (Perel, 1979: 78) plus a single (dubious?) record from Germany by Graff (1954), as reported by Easton (1981: 40) and an unconfirmed report from Slovakia on the "Databank of Slovak Fauna" (http://zoology.fns.uniba.sk/checklist/interface/results\_page.asp?binomen=Eisenia March, 2007) (cf. Gates, 1975; Blakemore, 2006b). A European Union website (EDDA, 2007) lists *Eisenia japonica* (Michaelsen, 1891) as the "Japanese Red Worm" in improvements of soil and stock feeding but does not necessarily imply its origin from Europe.

Table 1 compares the current species to regional species: *Eisenia japonica* (Michaelsen, 1891) (syn. *gigantica* Oishi: 1934, *minuta* Oishi: 1934) and *E. nordenskioldi* (Eisen, 1873) that were both originally in *Allolobophora* Eisen, 1874, and Korean *Eisenia koreana* (Zicsi, 1972) that was originally described in *Eiseniella* with transfer to *Eisenia by* Easton (1983). Siberian *Eisenia balatonica* (Pop, 1943), *Eisenia sibirica* Perel & Graphodatsky, 1984 and *Eisenia augusta* Perel, 1994 have clitella on 24,25-30, 27-32,33 and 27,28-33; and tubercula pubertatis on 26-29, 29-30 and 29-31, respectively (Perel, 1997: 75-76, figs. 27.2, 28; Csuzdi & Zicsi, 2003: 141). The current species differs in many characters from these *Eisenia* spp. but lack of consistent nephridial bladders appears most crucial.

Table 1. Characters of <i>Helodrilus hachiojii sp. nov.</i> , <i>Eisenia japonica</i> (morphs), <i>E.</i>					
koreana and E. nordenskioldi sub-species (from Blakemore, 2003, 2006b)					

	He. hachiojii	E. japonica	E. koreana	E. nordenskioldi
Length mm	60-70	(24)42-102(175)	30-35	25-150
Segments	100-125	(85)96-140(151)	?	80-176
Colour	Unpigmented/	Anterior	Dark brown	Puce or pallid
	light brown	pink/brown		
Clitellum	22,23-30,31	23,24-31	25-31	26,27-33,34
ТР	29	27 or 27 and 29	27-28	29-31,32
Shape of	Round	Round-triangular	Elongate	?
TP		_	-	
Nephridial	Absent (mostly)	Large, sausage-	Present	Sausage-shaped
bladders	· · · ·	shaped throughout		
Habitat	Semi-aquatic	Terrestrial	Amphibious	Terrestrial
TP tubercula pubertatic				

TP – tubercula pubertatis.

Absence of nephridial bladders (and these not replaced by canals) is unusual in Lumbricidae yet characterizes European *Helodrilus* Hoffmeister, 1845, moreover, the current specimens key out closest to type *Helodrilus oculatus* Hoffmeister, 1845 from Michaelsen (1900: 471, 480, 496), Sims and Gerard (1985, 1999: 49, 93) and Blakemore (2006b). *He. oculatus oculatus* is also semi-aquatic and widespread ranging from Ireland to regions of the Russian federation or former USSR; although rather poorly defined and lacking types, it is not yet reported from the Orient nor known with parthenogenesis. Easton (1983: 483) lists its synonyms as: syn. *hermanni* Michaelsen, 1890; *fontinalis* Friend, 1921 and its sub-species as *He. oculatus samniticus* Cognetti, 1914 [syn. *oculatus dudichi* Pop, 1943:14 (and possible homonym *Allolobophora samnitica* Pop, 1943: 14); *massiliensis* Bartoli, 1962]. Other similar (sub?)species are *He. deficiens* Zicsi, 1985 from the Danube which also possesses paired papillae but on 30 and 31, and *He. phillipei* Qiu & Bouché, 1998 that has calciferous glands in 11-14 (with diverticula in 11), however it has tubercula pubertatis occupying 28-29 and its type-locality is either in South of France or North of Morocco.

The current specimens differ from *He. oculatus*, not least, because this species has spermathecal pores (in 9/10/11 in setal c lines), large male pores within a tumid area extending from mid-14 to mid-16, and tubercula pubertatis as ovoid ridges lateral of setae b on 29-30 (Michaelsen, 1900: 497; Perel, 1979: 180; Sims & Gerard, 1999: 94). Variance from *He. oculatus* in all these characters may relate to parthenogenesis, however that such a degradation would leave evidence of an increased number of seminal vesicles or affect the calciferous glands would be most unexpected and so, on the balance of all similarities and differences (as shown in Table 2), the current specimens are considered a separate, albeit parthenogenetic, entity.

Characters	He. hachiojii	He. oculatus	
Length (mm)	60-70	35-80	
Segments	100-125	90-150	
Colour	Light brown	Unpigmented with some black dots	
Setae	Closely paired; no tumescences	Very closely paired, black; tumescences around ab on (10)11(12)	

Table 2. Characters of Helodrilus hachiojii sp. nov. and type species He. oculatus.

11-14 (annular)	10 (saccular, Sims & Gerard, 1999)
22,23-30,31	21,22-32
29	29-30 (or 30-31, Perel, 1997: 55)
Round	Elongate, continuous
Four pairs in 9-12	Two pairs in 11 & 12
Rectangular	Circular
None (parthenogenetic)	In 9/10/11(/12) in setal c lines
?	"fasciculated"
Absent (mostly)	Absent (some replaced by canals?)
	22,23-30,31 29 Round Four pairs in 9-12 Rectangular None (parthenogenetic) ?

TP – tubercula pubertatis.

The genus *Helodrilus* has taxonomic priority over contending genera noted above, it currently has 30 (sub-)species (Blakemore, 2006a), none previously reported outside Europe or the Mediterranean region. *Helodrilus hachiojii* appears close to *He. oculatus* and, although unlikely to be native, since it is not proven to be exotic we must at this time assume it an endemic lumbricid from Japan and anticipate discovery of its amphimictic form (perhaps in Europe or Siberia?) that would nevertheless retain the earlier name.

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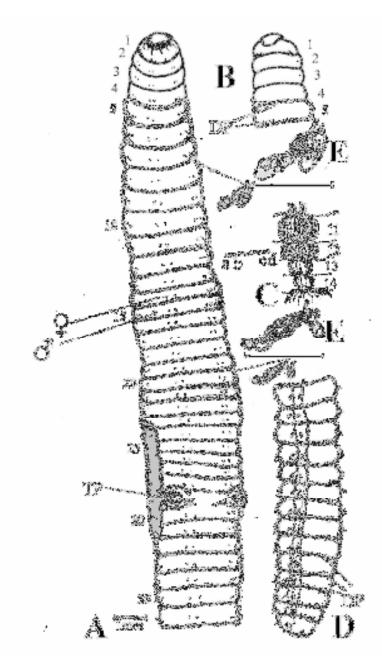
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**Fig. 1.** *Helodrilus hachiojii sp. nov.* - Holotype from Komiya-koen, Hachioji, western Tokyo, Japan. A, ventral view of anterior showing setal ratio on segment 12; B, dorsal view of prostomium; C, diagramatic sections of calciferous glands; D, dorsolateral posterior segments; E, *in situ* and enlargements of nephridia from segments 8 and 20 showing the only possible nephridial bladders found - see text for details. Abbreviations: a-b-c-d are setae on segment 12; DP – dorsal pores, TP – tubercula pubertatis. The clitellum is shaded and all scale bars are 1mm.