

Survey of Busan Oligochaeta earthworms supported by DNA barcodes

Robert J. Blakemore* and Seunghan Lee

¹Biodiversity Laboratory, College of Natural Science, Hanyang University, Seoul 133-791, Korea

*Correspondent: rob.blakemore@gmail.com

An earthworm survey of Busan metropolitan area unearthed a dozen taxa in four families (including Enchytraeidae). Members of mostly common, cosmopolitan earthworm species-complexes were: *Drawida* cf. *koreana* Kobayashi, 1938, *Amyntas* cf. *corticis* (Kinberg, 1867), *Aporrectodea trapezoides* (Dugès, 1828) and *Eisenia fetida* (Savigny, 1826). Also found were *Amyntas hupeiensis* (Michaelson, 1895), *A. masatakai* (Beddard, 1892) and *Metaphire ryunome* Blakemore, 2012 - the latter a new Korean record. New taxa are: moniligastrid *Drawida songae yeongdo* subsp. n.; megascolecid *Amyntas carnosus roki* subsp. n. which is compared to nominal taxon *A. carnosus* (Goto and Hatai, 1899) from Japan, to *A. carnosus monstriferus* (Kobayashi, 1936) stat. n. from Korea and to *A. lichuanensis* Wang and Qiu, 2005 stat. n. from China; plus lumbricid *Eisenia japonica vaga* subsp. n. deemed an objectively-based molecular taxon on its unique DNA COI gene barcode. Restoration of *Eisenia xanthurus* (Templeton, 1836) for *E. andrei* is mooted (in Appendix).

Keywords: biodiversity, coastal soil fauna, exotic species-complex, genetics, land sampling

©2013 National Institute of Biological Resources
DOI: 10.12651/JSR.2013.2.2.127

INTRODUCTION

Busan (부산 35° 10'46''N 129° 04'32''E) (formerly Pusan) is the major port and South Korea's second largest city (population ~3.6 million). Situated in the southeast coast of the Korean peninsula its climate is cool, humid subtropical. It may be expected to have a diverse biota of both native and introduced species due to an extensive coastline with several metropolitan mountains combined with its long and continuous history of trade, especially with Japan. Despite being homeland of several universities, most Busan institutes seem to favour marine biology and no systematic survey of its soil fauna has been undertaken to date. In the spirit of Kobayashi (1936; 1937; 1938; 1941) and Easton (1981), the current work contributes to the first author's current Japanese and Korean studies, e.g. Blakemore (2003; 2008; 2012a-e, 2013a, b), and attempts to use genetic barcodes of primary types or neotypes for definitive species identity (Hebert *et al.*, 2003; Blakemore, 2013c).

MATERIALS AND METHODS

A brief survey (by RJB) was conducted in the Spring

of 2013 by digging and hand-sorting soils at likely earthworm 'hot spots' (e.g. moist and vegetated microhabitats) at sites:

April 24th - Yeongdo Island along the southwest coast.

April 25th - Geumjeongsanseong Fortress and Geumgang Park north of the city (but because the forest is mainly pine plantation with acidic soil it had a low abundance of earthworms).

April 26th - Gwangalli Beach and walking towards Nam-gu peninsula just east of the city; plus Jungang Park opposite the central station that had a high diversity of earthworm species.

A minimum number of worms were collected for identification with any immatures or easily known taxa returned to the soil. Taxonomic determinations follow the classifications in Sims and Easton (1972) and Blakemore (2002; 2010; 2012f). Specimens in 80% EtOH lodged in the NIBR facility had small tissue samples taken for mtDNA COI barcode analysis (courtesy of Seunghan Lee at Hanyang University) using methods similar to Blakemore *et al.* (2010) with data in an Appendix. Genetic analyses and phylogram via defaults from 'MEGA 5' (www.megasoftware.net) and BLAST programs (www.blast.ncbi.nlm.nih.gov/BLAST.cgi) are compared to Genbank (genbank.com) and ENA (www.ebi.ac.uk/ena) entries or the author's iBOL (www.boldsystems.com)

‘Japanese Earthworm Types’ project data (e.g. Blakemore, 2012e) as well as to ongoing Korean studies.

Discussion is confined to remarks after each species’ description.

Abbreviations are: GMs - genital markings, lhs - left hand side; rhs - right hand side; TP - tubercula pubertates; TS - tumid setae; DPs - dorsal pores; neph. - nephridium; “?” indicates some taxonomic uncertainty. Scale bars in all figures are 1 mm.

GENETIC/TAXONOMIC RESULTS AND DISCUSSION

Ten or more earthworm species were collected that constitute first records for Busan and provide preliminary DNA data for taxon identification and phylogenetic relationships (Fig. 1).

Annelida Lamarck, 1802

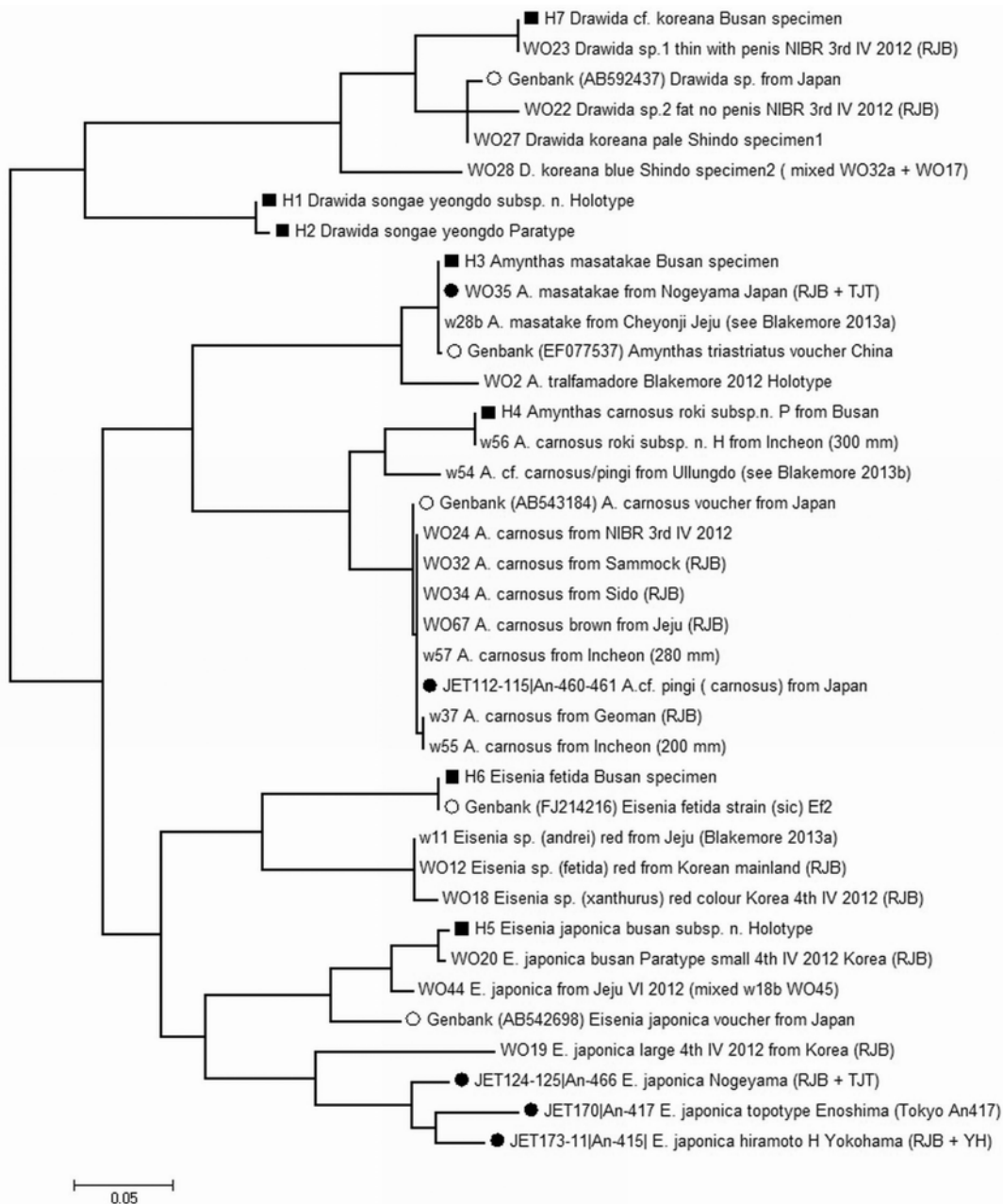


Fig. 1. MEGA5 Maximum Likelihood phylogram of DNA code results for Busan specimens compared to similar taxa. Solid squares (■) are Busan samples; open circles (○) are unconfirmed GenBank names (from China, Korea or Japan); closed circles (●) are from the author’s Japanese studies. (Sample codes and provisional or new species names are explained in the Appendix legend).

Microdrilacea Benham, 1890

ENCHYTRAEIDAE

1. *Enchytraeus*? spp. several specimens were noted but not collected as they are ecologically less significant compared to megadriles (Megadrilacea) and not particularly diverse. Only 676 enchytraeid taxa are known globally compared to ~7,000 earthworms (Cs. Csuzdi, 2012 and pers. comm. June, 2013; Blakemore, 2012f), i.e., less than about one tenth of terrestrial megadriles.

Megadrilacea Benham, 1890

MONILIGASTRIDAE

2. *Drawida* cf. *koreana* Kobayashi, 1938: 102

Material. NIBR-IV0000261283 (DNA sample H7), mature specimen, 26th April from Jungang Pk.

Description. Slight blue-grey colour. Length 47 mm. Clitellum 10-13 at least. Dorsal pores absent. Spermathecal pores in atria in 7/8. Male pores on 10/11 protrude on penes. Genital markings are present on 8rhs above setae ab and 10rhs on male pore.

Remarks. The DNA sample H7 (Appendix) agrees 100% with WO23, a specimen from NIBR, Incheon (IV0000249929) identified as "*Drawida* sp." and it is possible that both are *D. koreana* proper. Originally described on 150 or so specimens ranging from "Kyôjô" in North Korea to Gyeonggi-do South Korea, *D. koreana* is a variable species. Several other specimens in current studies are superficially similar but appear to differ molecularly (Fig. 1). Types of *D. koreana* are missing - if they ever existed - and there is an urgent need for neotypification, albeit previous and contemporary Korean workers have avoided this for the last 75 yrs! Further study is ongoing to determine intraspecific variation permissible for *D. koreana* identification using both its morphology and genetics (Blakemore in prep.).

3. *Drawida songae yeongdo* Blakemore subsp. n. (Fig. 2).

Material. NIBR-IV0000261284 (DNA sample H1), H, holotype, a complete specimen 24th April, 2013 from Yeongdo Island in woodland just below Temple on south-east coastal walk. In same sample: IV0000261285 (DNA sample H2), P, paratype, a smaller (37 mm) and paler immature; and IV0000261293, three immature *Amyntas* sp. that were not studied further.

Etymology. Noun, after type-locality.

Description. Colour copper green in life looking like *Amyntas hupeiensis* but without its characteristic smell and coiling behaviour: this specimen was sluggish and

remained elongated. Prostomium prolobous. Clitellum not noticeable. Length 135(+?) mm. Segments 202 with the last seven segments paler, possibly regenerated or a zone of growth. No DPs. Nephropores near intersegments and appear mainly in cd but are above d in the anterior. Spermathecal pores in 7/8 in ab lines. Male pores in 10/11 in ab lines. Female pores in anterior of 12 in ab lines. Genital markings in 8-10: paired in 8, 9 and 11 with analogue in 10lhs (faint remnant in 10rhs?); sessile glands correspond internally.

Septum "9/10" displaced to mid-segment 10; 11/12 also displaced and combined with 12/13. Dorsal blood vessel single; hearts large in 6-9. Spermathecal ampulla on 7/8 with long duct to body wall; no atrium present in 8. Testis sacs in 10/11 with long coiled vasa deferentia to flat, sessile prostates; no penes. Ovaries as several sheets enclosed in septa with small and empty egg sac posteriorly from 12/13. Gizzards in 14-17; gut filled with organic soil supporting classification as a topsoil species. Nephridial bladders elongate.

Remarks. The current species appears particularly close to *Drawida songae* Hong, 2002 from Daegu that is somewhat smaller at 70-93 mm with fewer segments (viz. 172). Both taxa have simple, superficial spermathecal and male pores, and four gizzards in 14-17. However *D. songae* was described with a pink colour (but possibly due to preservation?) rather than green as in the current specimen. The genital markings were twice described as from 7-11,12 (although it is ambiguous since only some were figured - see Fig. 2B), whereas the current specimen lacks markings in both 7 and 12 but has them clearly in 8-11. The markings in *D. songae* were said to have "muscular areas" that probably correspond to the sessile glands observed in the present species. Other differences are that the egg-sacs in *D. songae* were larger (more mature or seasonal?) from 12-14 and were said to cover the gizzards. Perhaps most distinctive, the prostates were said to be absent whereas the current specimen has low, flat glands. The accumulation of these differences possibly merits separate species status although the gross similarities suggest a close relationship and thus it is proposed as a new sub-species. Attached DNA data should help resolve its ultimate status and relationships, should further material of nominal *D. songae songae* be uncovered.

MEGASCOLECIDAE

4. *Amyntas carnosus roki* Blakemore subsp. n. (Fig. 3).

Material. Incheon specimen NIBR-IV0000261264 (DNA w56) H, holotype, mature broken in two parts, anterior dissected; collected by RJB 23rd Oct., 2012 from Incheon Great Park from infilled ditch just above the park bound-

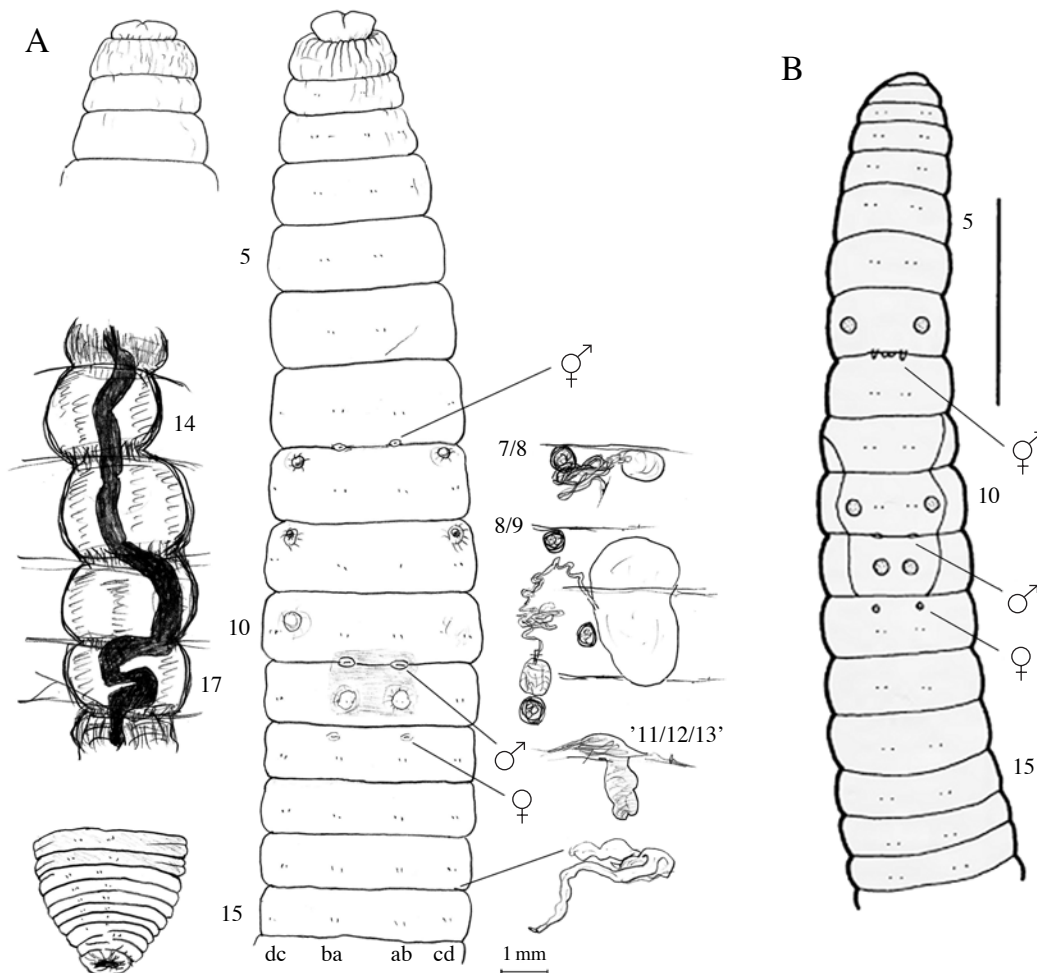


Fig. 2. A. *Drawida songae yeongdo* subsp. n. holotype (H, DNA H1) showing prostomium, gizzards, pygidium, habitus with reproductive organs *in situ* and all four genital marking glands in 8-11hs plus a nephridium from 15rhs. B. *D. songae songae* after Hong (2002: fig. 1) with GMs in 7, 10-11 (but not 12?) for fair use comparison.

ary. Busan specimen IV0000261286 (DNA H4) P, paratype, mature 175 mm long, undissected but broken in two parts, 24th April, 2013 from Yeongdo Island, just down from Temple in woodland on walk to the beach.

Etymology. From acronym of Republic of Korea (ROK), latinized to “roki”, due to its wide distribution (Incheon ↔ Busan) even though endemism just in South Korea is not implied.

Description. Lengths H, 270+30 mm (=300 mm); P, 125+50 mm (=175 mm). Segments H, 112+24 (=136). Colour dark brown dorsum, clitellum buff. Prostomium epilobous open. First dorsal pore 12/13. Setae ca. 60 on 12. Spermathecal pores post-intersegmental in U-shaped hemispheres on dorsal aspects of 5/6/7/8/9. Clitellum 14-16. Female pore mid-ventral on 14. Male pores superficial on small mounds within concentric rings ca. 0.3 C apart on 18 with ca. 14 setae intervening. Genital markings absent (from both specimens).

Internally (H) septa 8/9/10 aborted. Spermathecae in 6-9 as figured. Holandric with seminal vesicles in 11 and 12. Last hearts in 13. Prostates racemose in 18. Intestinal caecae simple from 27. Other characteristics comply with nominal taxon (see Blakemore, 2012a).

Remarks. Distinctive characters are the tendency to large size (175-300 mm), U-shaped post-intersegmental spermathecal pores (called “spermathecal papillae” by Kobayashi, 1936: 130) plus lack of genital markings thereby complying with Kobayashi’s (1936, text-figs types II & I) also coded by Blakemore (2012a, fig. 3) [as already noted, segment counts for XVIIrh are out by one in this latter figure]. On these characters the present subspecies appears to differ from the nominal taxon’s neotype and from other synonyms in Blakemore (2012a: 36), also supported with definitive DNA barcode data from its primary type (w56 in Appendix). It shows only 91% similarity to other *A. carnosus* specimens and was just

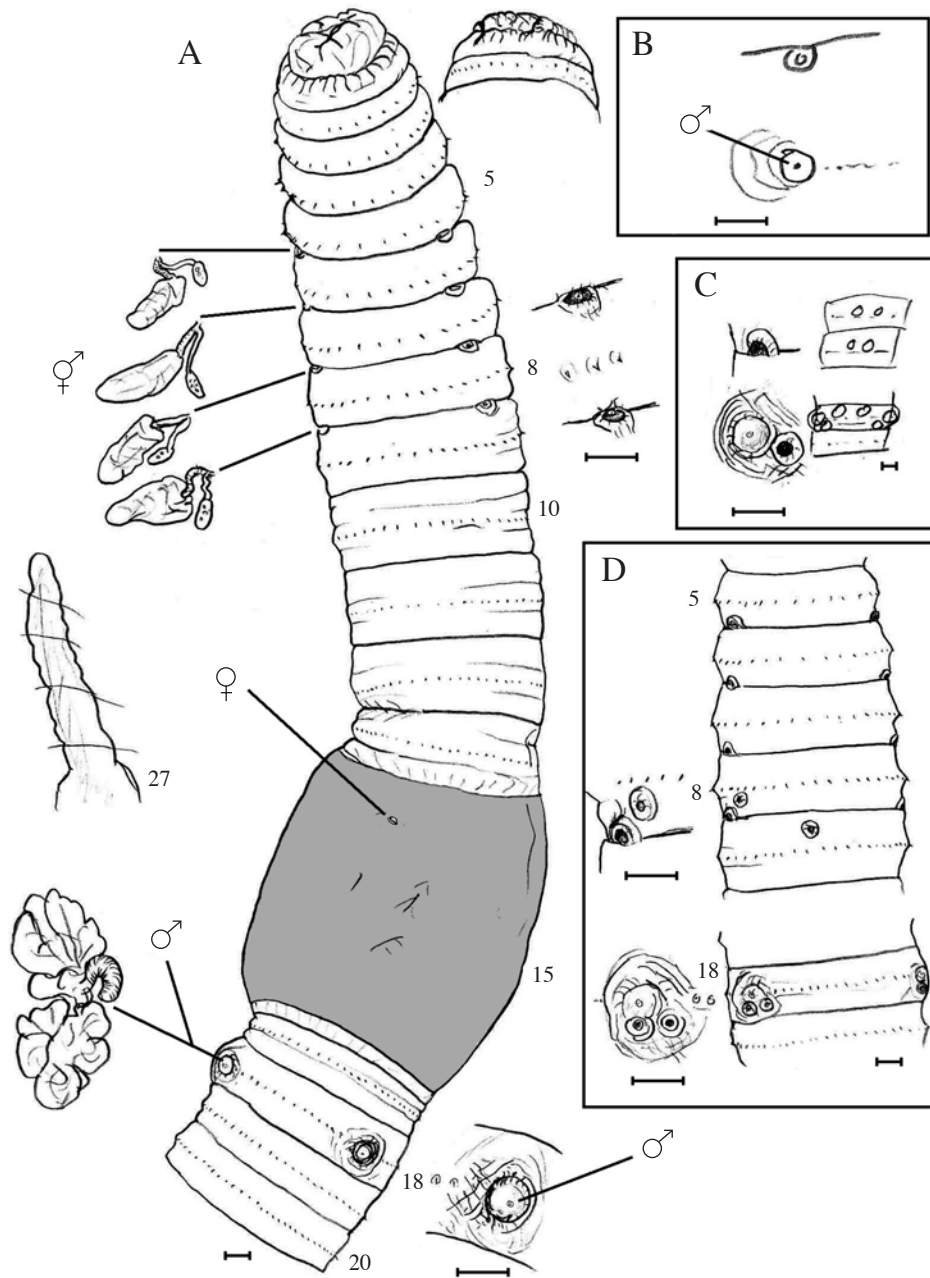


Fig. 3. A. *Amynthus carnosus roki* subsp. n. holotype (H, DNA w56) showing habitus, prostomium, spermathecae, caecum and prostate of 18lhs *in situ* with X2 enlargements of post-intersegmental spermathecal pores of 8rhs and simple male pore of 18rhs. B. Sketch of these from Busan paratype (P, DNA H4 agreeing 100%). 3C and 3D are comparison sketches with similar X2 enlargements for sympatric *A. carnosus carnosus* (Goto and Hatai, 1899) specimens from Incheon (200 and 280 mm long) with pre-intersegmental spermathecal pores that provided DNA samples w55 and w57, respectively, agreeing >99% with each other (see Fig. 1 and Appendix).

93% compliant with w54 DNA data from an Ulleungdo specimen. The DNA of sample H4 from Busan has 100% agreement with w56 from Incheon designated as paratype and holotype, respectively.

This taxon is particularly similar to *A. monstiferus* (Kobayashi, 1936), differing not least on the lack of markings on segment 8, and possibly they should be

merged. Pending further research, *A. carnosus monstiferus* is reduced to sub-species status as it complies with Kobayashi's (1936) types XII & I of *A. carnosus*!

Blakemore (2012a: 41) stated, “the genital marking variation in *A. carnosus* allowed for by Kobayashi in his detailed and most thorough account is excessive, rather representing a congeries of morphs, if not separate spec-

ies". This seems to be borne out by the preliminary DNA results presented here as justified with the new taxon. In particular, sample IV0000261266 (providing DNA w57) was collected at the same time as *A. carnosus roki* Holotype (w56) yet it complies with Kobayashi (1936) text-fig. types IX & III of *A. carnosus carnosus*. Whereas IV0000261263 (providing DNA w55) also from Incheon Great Park but found crawling on the surface at dusk, complies with Kobayashi's types VI & VIII and is almost exactly the same as *A. carnosus* neotype (Tokyo NMST An435). Both these latter specimens are closer morphologically to materials from elsewhere in Korea and, significantly, from Japan that clearly separate out on their genetics from the current taxon.

From the results in the DNA phylogram (Fig. 1) it appears that the specimen from Ulleungdo (providing DNA sample w54) labeled as "*Amyntas pingi* (Stephenson, 1925)" by Blakemore (2013b: 60) may also merit separate sub-specific status, which is investigated further in an accompanying paper based on *A. pingi* types (Blakemore, 2013d this issue).

Regarding other putative sub-species of *A. carnosus*, it was already noted by Blakemore (2012a: 36) that *A. pingi chungkingensis* (Chen, 1936) probably merits elevation to separate species level as *A. chungkingensis*. This taxon was ascribed to "*Am. carnosus chungkingensis*" by Wang and Qiu (2005: 25, tab. 2) when comparing it to their *Amyntas carnosus lichuanensis* Wang and Qiu, 2005 from Mt. Xingdou, Hubei, China. Herein this is also raised to separate species level as *A. lichuanensis* stat. nov. since it has no characters in any way similar to revised concepts of *A. carnosus*. For example, this 300-400 mm species has rows of 7-8 papillae median to male pores on 18, and corresponding rows of three or four papillae on segment 8 median to spermathecal pores that number only three pairs in 6/7/8/9 with its spermathecae nubby stubs. These authors also record *A. carnosus carnosus* (Goto and Hatai, 1899) from this location, but their identification may be suspect since they claim subsumed *A. fornicatus* (Gates, 1935) (and *A. diffringens*!) too (see Blakemore, 2013d).

Similar specimens from any region may now be compared genetically to an increasingly refined formulation of *A. carnosus*, including the present new sub-species. Moreover, as coded by Blakemore (2012a, fig. 3), Kobayashi's (1936) text-figs types II & I (lacking markings) and possibly types XII & I (with markings in 8), having U-shaped post-intersegmental spermathecal papillae should now be removed as characteristic of *A. c. carnosus* proper. Kobayashi (1936: 130) tabulated 14 such specimens from 204 from Korea and this may represent an approximate proportion (~7%) of the *A. carnosus roki* (and *A. c. monstiferus*) sub-species, but its area of prevalence is currently indeterminate.

5. *Amyntas cf. corticis* (Kinberg, 1867)

Material. NIBR-IV0000261287, a mature specimen collected 25th April, 2013 from Dongnae Botanic Gardens, Geumgang Park; IV0000261288 second and third complete specimen (160 and 100 mm long) plus a possible sub-adult (80 mm) from Jungang Park, 26th April.

Remarks. The Dongnae specimen had spermathecae only in 7-9 that had diverticula reduced or wanting and lacked GMs. Approximately 130 pheretimoids are described with spermathecal pores in 6/7/8/9 (Blakemore unpublished), of these about 69 are in *Amyntas* plus *A. carnosus* and *A. corticis* that sometimes (rarely?) have specimens lacking their anterior pair of spermathecae, as in *A. chungkingensis* (Chen, 1936) that was at one time associated with *A. carnosus* as noted above (it differs further in having only one pair of GMs on 18 just below the male pores). Other than these two taxa, the only previously known Korean representative is *Amyntas moakensis* Hong and Kim, 2002 that, however, is smaller (32 mm) and does appear to have a pair of GMs posterior-ventral to male pores (cf. Hong and Kim, 2002: fig. 3) despite its description of having none, and long tightly coiled diverticula on the spermathecae.

In Blakemore (2002; 2010; 2012f) it keys to *Amyntas asiaticus* Michaelsen, 1900. Michaelsen (1900: 527) described Chinese *Pheretima asiatica* (Michaelsen) with "Intestinal caeca long, with wart-like outgrowths underneath. Spermathecal diverticulum about as long as the ampulla, its proximal third tapered somewhat, slightly tortuous and narrow" - conditions that do not apply to the current specimen. The tentative conclusion is a parthenogenetically degraded *A. corticis* sixthelcal morph lacking GMs but retaining its prostate glands. DNA data from this otherwise unremarkable and common species is pending; nevertheless, it is reviewed further in an accompanying paper (Blakemore, 2013d).

6. *Amyntas hupeiensis* (Michaelsen, 1895)

Material. Specimens collected from Jungang Park, 26th April but returned to the earth there.

Remarks. This is a distinctive and fairly common cosmopolitan species already well known from Korea (e.g. Kobayashi, 1938: 152) thus not unexpected in Busan.

7. *Amyntas masatacae* (Beddard, 1892)

(Fig. 4A).

Material. NIBR- IV0000261289 (DNA sample H3; Fig. 4A) from Jungang Park, 26th April. Natural History Museum, London types inspected for confirmation (see Blakemore, 2012d).

Description. Length 180 mm. Clitellum 14-16. Sperma-

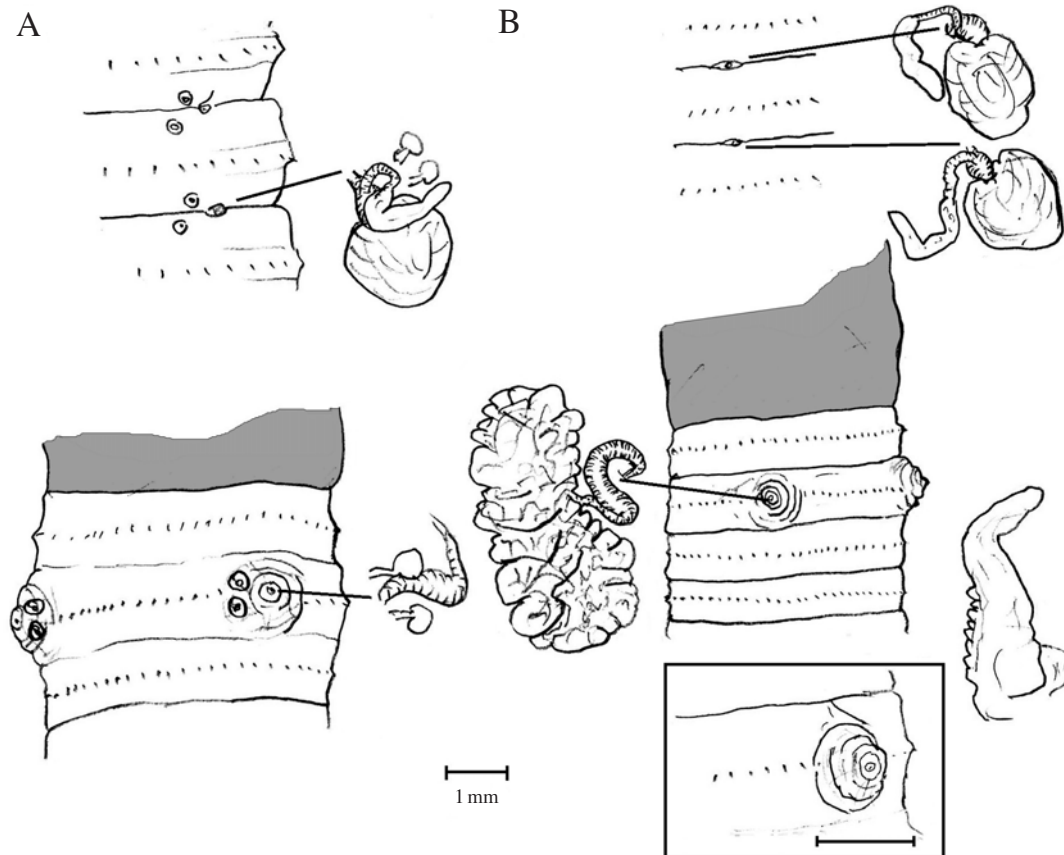


Fig. 4. A. *Amynthus masatakae* (Beddard, 1892) Busan specimen showing spermathecal and male regions. B. *Metaphire ryunome* Blakemore, 2012 Busan specimen [boxed is X2 enlargement of its 18th male pore that appears elevated, non-superficial, i.e., = *Metaphire*].

thecal pores 7/8/9. Male pores on 18, superficial. GMs typically present, paired near spermathecal and male pores. Spermathecae with elongated, paprika-shaped diverticular bulbs on long stalks. Prostates (always?) reduced to muscular duct. Glands associated with GMs, where present. Caeca ventrally incised.

Remarks. DNA data in the appendix helps to confirm this identity based on the redescriptions of *A. masatakae* by Blakemore (2012d: 134; 2013a: 29). Kobayashi (1937: 337, 1938: 140) observed complete prostates, in some of his Korean specimens, although such specimens should now be compared to *Metaphire ryunome* as next described. Whereas this latter taxon has male pores slightly elevated (resembling those in the type-species of *Metaphire* Sims and Easton, 1972 - see Blakemore, 2012b) and thus strictly non-superficial qualifying for membership of genus *Metaphire*, such a condition has not been confirmed in amphimixic forms of *A. masatakae* although accidentally indicated in Blakemore (2012a: 31) where it says "*M. masatakae*", again possibly alluding to specimens resembling *M. ryunome*.

Note that although Kobayashi (1938: 138) said the caeca were simple "without indentations" (lapsus?), his

earlier account (Kobayashi, 1937: 338) had "each with several serriformed outgrowths on ventral margin only", as were found in the current specimen. Kobayashi (1937: 340) also notes that Goto and Hatai (1899) were incorrect to cite the present species as having manicate caeca.

8. *Metaphire ryunome* Blakemore, 2012 (Fig. 4B).

Material. IV0000261294 (DNA HY2, this data not included in phylogram Fig. 1) collected from Jungang Park, 26th April, 2013.

Description. Length 116 mm. Clitellum 14-16. Spermathecal pores 7/8/9. Male pores on 18, slightly protruded. GMs absent (cf. Shiga types). Spermathecae with elongated, paprika-shaped diverticular bulbs on long stalks. Prostates present but copulatory pouches absent. Caeca ventrally incised (27rhs removed to check for resident Rotifera that, however, were not found).

Remarks. Slight differences from the original description of Japanese *M. ryunome* are that the GMs were not recorded and that the caeca were serrate. Nevertheless, DNA shows 100% conformity with barcode results in

Blakemore (2012d: fig. 4). Finding *M. ryunome* in Shiga, Japan and now Busan, Korea indicates that this species has wide distribution suggesting it is peregrine and is to be expected from other countries. The BLAST results (Appendix) show it closest to Chinese *A. incongruus* (Chen, 1933) on GenBank; however, that taxon typically has spermathecal pores in 5/6/7 rather than 6/7/8 and a different arrangement of GMs, at least, compared to those discussed by Blakemore (2012d) for *M. ryunome*. Most likely the identity of the Chinese specimen was mistaken, just as the DNA data for *A. masatakae* appears misidentified with Chinese *A. triastriatus* (Chen, 1946) on Gen Bank (see Appendix).

LUMBRICIDAE

9. *Aporrectodea trapezoides* (Dugès, 1828)

Material. NIBR-IV0000261290, four representative specimens from many collected 26th April, 2013 on walk from Gwangalli Beach towards Nam-gu.

Description. Male pores on 15. Clitella 26-34; tubercula pubertates 31-33 continuously.

Remarks. Abundant locally and previously recorded from Korea (e.g. Kobayashi, 1941: 150).

10. *Eisenia fetida* (Savigny, 1826)

Material. NIBR-IV0000261291 (DNA sample H6), a mature specimen collected 26th April, 2013 from Jungang Park beside a walking trail.

Description. Body has 'classic' transversely coloured stripes alternating with pale intersegments; also pale laterally in 8,9-11,12. Clitellum 26-31,32; TP on 28-30.

Remarks. Part of the species-complex (Fig. 1 and Appendix), agreeing 100% with GenBank sample (FJ214216) from "Denmark: Svendborg". Obviously this specimen differs from some Korean samples variously named "*E. fetida*" or "*E. andrei*" - see Blakemore (2013a: 42; 2013b: 64) and recent Mongolian work (Blakemore, 2013c). True identity and status of specimens/taxa remains unclear, requiring further work (Blakemore in prep.).

11. *Eisenia japonica vaga* Blakemore sub-sp. n. (Fig. 5).

Material. NIBR-IV0000261292 (DNA H5) H, holotype, a mature specimen collected 26th April, from Jungang Park under (Japanese?) sakura cherry trees. IV0000250889 (DNA WO20) P, paratype, an undissected mature specimen collected 5th April, 2012 by RJB from near Hgae Bridge, Young River central South Korea; (a second sympatric specimen from there, IV0000250890, not tested genetically, may be similar).

Etymology. Latin for "wandering" from its distribution that may also include New Zealand.

Description. Grey unpigmented in alcohol with white clitellum and dark dorsal vessel visible. Length 58 mm. Segments 120. First dorsal pore 4/5. Clitellum $\frac{1}{2}$ 23-31. TP distinctively raised on 27 and 29. Setae ab on 11, 17 and 18 faintly marked; ab on 25 and 26 clearly tumid. Spermathecal pores in 9/10/11 in c lines. Holandric: testes iridescent in 10 and 11; seminal vesicles in 9-12, the latter elongate. Last hearts in 11. Calciferous glands in 11 and 12. Intestine from 15. Gizzard in 17-18. Typhlosole large T-shaped from about 19 onwards. Nephridial bladders sausage-shaped. Gut contains mainly mineral soil (i.e., a topsoil species that is at least partly geophagous).

Remarks. Superficial agreement supported by its DNA data shows this to be part of the *Eisenia japonica* Michaelsen, 1892 species-complex. It is not clear which part is closest, however it is remarkably similar to a New Zealand specimen described by Blakemore (2012c: fig. 3, 2013a: 40, 45). The DNA data is unequivocal that the Busan specimen objectively represents an as yet unknown taxon, and ditto sample WO20 having 99% agreement (Fig. 1 and Appendix). The current specimen sequence is only 92-93% similar to GenBank specimens unidentified as "Lumbricidae sp." (AB607078) or as *Eisenia japonica* (AB542698) from Japan. And it is just 82-83% similar to the author's *Eisenia japonica japonica* (Michaelsen, 1892) topotype from Enoshima, Japan or to *E. j. hiramoto* Blakemore, 2012 from near Yokohama. Such genetic dissimilarity (~20%) is normally associated with separate species/genera or even separate families (e.g. see sample H2 megaBLAST results in Appendix).

The scant difference from previous descriptions (e.g. by Kobayashi, 1941; Gates, 1975; Blakemore and Grygier, 2011; Blakemore, 2012c) of *E. japonica* are perhaps the tumid setae ab only on 25 and 26 (cf. on 22 and 26 in a syntype - Fig. 5), and the TP on 27 and 29 maybe less triangular. Differences from *E. japonica minuta* (Oishi, 1934) supposedly from Morioka and Asamushi, Japan that is, however, more usually combined under the nominal taxon, are that the TP are not so round and the size is slightly more than 24-55 mm as allowed for by Oishi (1934) and later by Kobayashi (1941: 153) and Easton (1981, tab. 2).

Kobayashi (1941) had specimens from "Keiki-d6" identified as *A. japonica minuta* Oishi, 1934 - they also had setae ab of 25 and 26 on oval papillae - although it is now certain neither that this complies with the Japanese originals nor that his identification is correct in light of the new information presented here. Moreover, that the specimen providing DNA sample WO19 (Fig. 1 and Appendix) represents *E. japonica gigantica* (Oishi, 1934) originally from Tomakomai, Japan reported from "Kwaizan" in Chŭngchŏng-bukto, South Korea by Kobayashi (1941:

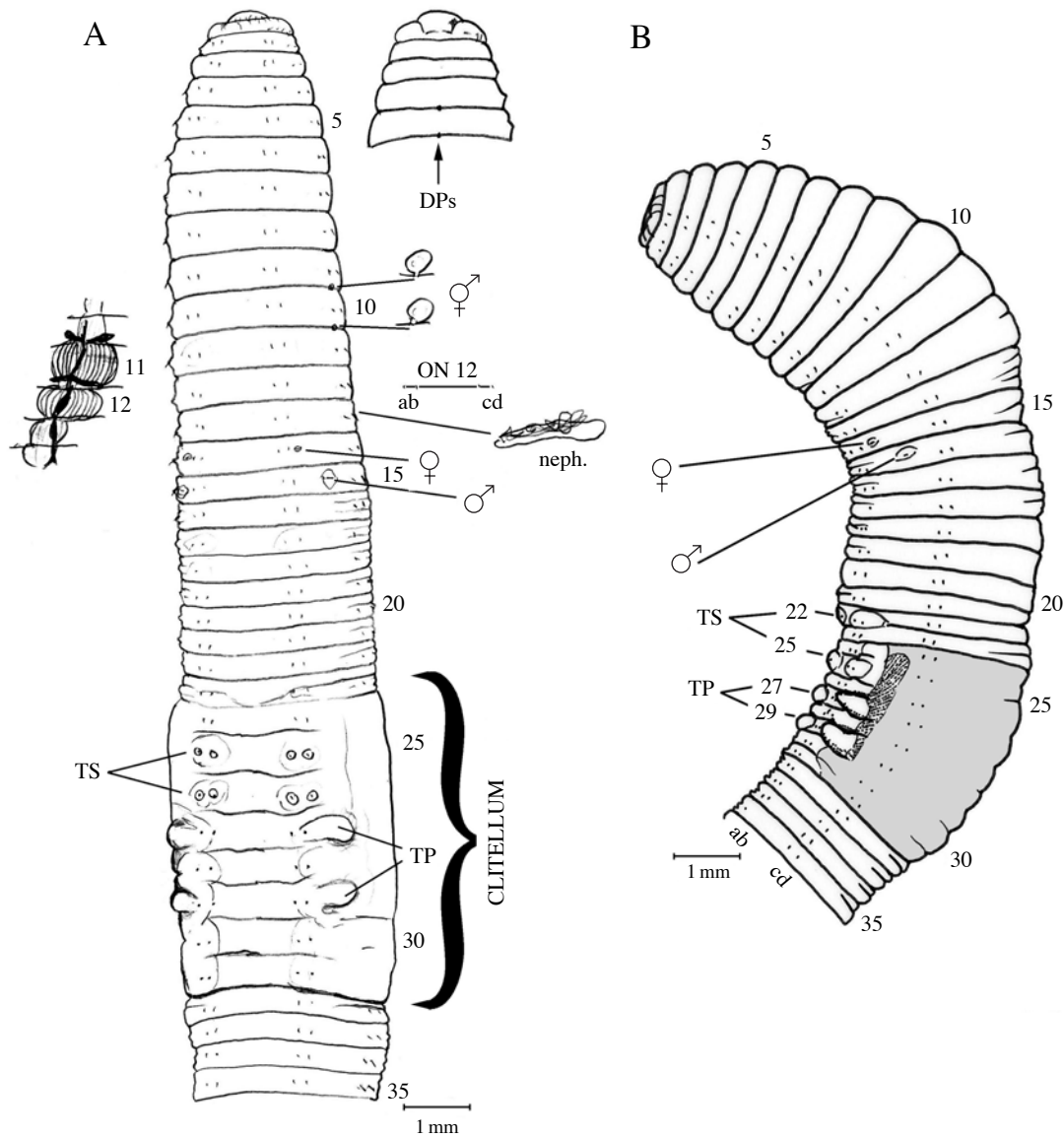


Fig. 5. A. *Eisenia japonica vago* sub-sp. n. Busan holotype (H, DNA H5). B. *Eisenia j. japonica* (Michaelsen, 1892) Berlin syntype Nr. 2117 (as anticipated, DNA was not extractable from this older material), author’s sketch after Blakemore and Grygier (2011, fig. 2) that compares to Hamburg syntypes V119-V121 (see Blakemore and Grygier, 2011, fig. 3).

152) is also unclear. Neither *minuta* nor *gigantica* have since been found and reported sufficiently for comparison by subsequent Japanese workers.

Eisenia japonica vago is a molecular taxon objectively defined on its definitive DNA.

ACKNOWLEDGEMENTS

The collection trip was part of a Korean biodiversity project under auspices of Hanyang University courtesy of Prof. Wonchoel Lee supported by the National Institute of Biological Resources (NIBR), funded by the Min-

istry of Environment (MOE) of the Republic of Korea. Sean Prosser and Natalia Ivanova of Canadian Guelph University Centre for Barcoding provided the ‘JET’ sample DNA results through courtesy of the director Dr Paul Hebert. Thanks for loans and curation of museum types plus access to library resources are extended to staff at NIBR (Incheon), Dr Andreas Schmidt-Rhaesa (Hamburg), Dr Birger Neuhaus (Berlin), Dr Toshiaki Kuramochi (Tokyo), Emma Sherlock (London) and to Dr Csaba Csuzdi (Budapest). Anonymous referees and JSR editors are thanked for constructive comments, yet all taxonomic work/decisions are the primary author’s responsibility.

REFERENCES

[For brevity, not all taxonomic authorities appended to species binomials are cited here].

- Blakemore, R.J. 2002. Cosmopolitan earthworms - an ecotaxonomic guide to the peregrine species of the World. VermEcology, Kippax, Australia. Pp. 506 [CD publication].
- Blakemore, R.J. 2003. Japanese earthworms (Annelida: Oligochaeta): a review and checklist of species. Organisms, Diversity and Evolution 3(3):241-244 [Available from: <http://www.urbanfischer.de/journals/ode/>; www.senckenberg.de/odes/03-11.htm].
- Blakemore, R.J. 2008. Korean earthworm species - updated checklist. In: A series of searchable texts on earthworms - Supplemental [CD publication compliant with ICZN, provided online as a courtesy from: <http://www.annelida.net/earthworm/Korean.pdf>].
- Blakemore, R.J. 2010. Cosmopolitan earthworms. 4th Edition. VermEcology, Yokohama, Japan. Pp. 750 [CD publication].
- Blakemore, R.J. 2012a. *Amyntas carnosus* (Goto and Hatai, 1899) redescribed on its neotype (Oligochaeta: Megadrilacea: Megascolecidae). Journal of Species Research 1(1):35-43 [Available from: www.nibr.go.kr/download?attach_path=journalandattach=2012vol1no1_4.pdf].
- Blakemore, R.J. 2012b. Restating scope of genus *Metaphire* Sims and Easton, 1972: 40 years on (Oligochaeta: Megascolecidae). Zoology in the Middle East, Supplementum 4:5-14.
- Blakemore, R.J. 2012c. On Schmarida's lost worm and some newly found New Zealand species. Journal of Species Research 1(2):105-132 [Available from: http://www.nibr.go.kr/english/event/journal_spe_3.jsp].
- Blakemore, R.J. 2012d. Earthworms from NIBR Jeju-do biosphere compared to historical and new Japanese types (Oligochaeta: Megadrilacea: Megascolecidae). Journal of Species Research 1(2):133-115 [Available from: http://www.nibr.go.kr/english/event/journal_spe_3.jsp].
- Blakemore, R.J. 2012e. On opening a box of worms (Oligochaeta, Megascolecidae) - Historical earthworm specimens transferred to Tokyo from the Saito Ho-on Museum of Natural History in Sendai. Bulletin of the National Science Museum, Tokyo Series A 38(3):95-124.
- Blakemore, R.J. 2012f. Cosmopolitan earthworms. 5th Edition. VermEcology, Yokohama, Japan. Pp. 950 [CD publication].
- Blakemore, R.J. 2013a. Jeju-do earthworms (Oligochaeta: Megadrilacea)-Quelpart Island revisited. Journal of Species Research 2(1):15-54 [Available from: www.nibr.go.kr/eng/event/journal_spe_3.jsp].
- Blakemore, R.J. 2013b. Ulleung-do earthworms - Dagelet Island revisited. Journal of Species Research 2(1):55-69 [Available from: www.nibr.go.kr/eng/event/journal_spe_3.jsp].
- Blakemore, R.J. 2013c. Earthworms newly from Mongolia (Oligochaeta, Lumbricidae, *Eisenia*). ZooKeys 285:1-21 [Available from: <http://www.pensoft.net/journals/zookeys/article/4502/abstract/earthworms-newly-from-mongolia-oligochaeta->, doi: 10.3897/zookeys.285.4502].
- Blakemore, R.J. 2013d. *Megascolex (Perichaeta) diffringens* Baird, 1869 and *Pheretima pingi* Stephenson, 1925 types compared to the *Amyntas corticis* (Kinberg, 1867) and *A. carnosus* (Goto & Hatai, 1899) species-groups (Oligochaeta: Megadrilacea: Megascolecidae). Journal of Species Research 2(2):99-126.
- Blakemore, R.J. and M.J. Grygier. 2011. Unravelling some Kinki worms (Annelida: Oligochaeta: Megadrili: Lumbricidae) Part III. Journal of Soil Organisms 83(2):231-244 [Available from: www.senckenberg.de/files/content/...83_2/06_artikel_blakemore.pdf].
- Blakemore, R.J., E. Kupriyanova and M.J. Grygier. 2010. Neotypification of *Drawida hattamimizu* Hatai, 1930 (Oligochaeta: Megadrili: Moniligastridae) as a model linking mtDNA (COI) sequences to an earthworm type, with a response to the 'Can of Worms' theory of cryptic species. ZooKeys 41:1-29 [Available from: doi: 10.3897/zookeys.41.374, <http://pensoftonline.net/zookeys/index.php/journal/article/view/374/401>].
- Chen, Y. 1936. On the terrestrial Oligochaeta from Szechuan II with notes on Gates' types. Contribution of the Biological Laboratory of Science Society of China (Zoology) 11:269-306.
- Csuzdi, Cs. 2012. Earthworm species, a searchable database. Opuscula Zoologica, Budapest 43(1):97-99.
- Easton, E.G. 1981. Japanese earthworms: a synopsis of the Megadrile species. - Bulletin of the British Museum (Natural History) Zoology 40(2):33-65 [Available from: <http://www.archive.org/stream/bulletinofbritis40zoollond#page/n43/mode/2up>].
- Gates, G.E. 1975. Contributions to a revision of the earthworm family Lumbricidae, XVIII. *Eisenia japonica* (Michaelsen, 1891). Megadrilologica 2(4):1-3.
- Goto, S. and S. Hatai. 1899. New or imperfectly known species of earthworms. No. 2. Annotations Zoologicae Japonensis 3(1):13-24.
- Hebert, P.D.N., A. Cywinska, S.L. Ball and J.R. deWaard. 2003. Biological identifications through DNA bar-codes. Proceedings of the Royal Society, B 270:313-321.
- Hong, Y. 2002. New earthworms of the genus *Drawida* Michaelsen, 1900 (Oligochaeta: Moniligastridae) from Korea. Revue Suisse de Zoologie 109(3):475-482 [Available from: <http://biostor.org/reference/115384>].
- Hong, Y. and T.-H. Kim. 2002. Four New Earthworms of the genus *Amyntas* (Oligochaeta: Megascolecidae) from Korea. Korean Journal of Biological Science 6:195-199.
- Kobayashi, S. 1936. Distribution and some external charac-

- teristics of *Pheretima (Ph.) carnosus* (Goto et Hatai) from Korea. Science Report of the Tohoku Imperial University 11(1):115-138.
- Kobayashi, S. 1937. Preliminary survey of the earthworms of Quelpart Island. Science Report of the Tohoku Imperial University (B) 11(3):333-351.
- Kobayashi, S. 1938. Earthworms of Korea I. Science Report of the Tohoku Imperial University 13(2):89-170.
- Kobayashi, S. 1941. Earthworms of Korea. II. Science Report of the Tohoku Imperial University (B) 16(2):147-156.
- Michaelsen, W. 1900. Das Tierreich 10: Vermes. Friedländer and Sohn, Berlin. Pp 757.
- Oishi, M. 1934. Earthworms. Zoological Magazine, Tokyo 46:133-134 [in Japanese].
- Sims, R.W. and E.G. Easton. 1972. A numerical revision of the earthworm genus *Pheretima* auct. (Megascolecidae: Oligochaeta) with the recognition of new genera and an appendix on the earthworms collected by the Royal Society North Borneo Expedition. Biological Journal of the Linnean Society 4:169-268.
- Wang, H.-J. and J.-P. Qiu. 2005. A Preliminary Report on Pheretimoid Earthworms from the Xingdou Mountain Nature Reserve-With Description of Two New Species and a New Subspecies of the Genera *Amyntas* and *Metaphire*. Journal of Shanghai Jiaotong University (Agricultural Science) 23(1):23-30 [in Chinese with English summary, Available from: http://file.lw23.com/0/00/00a/00a16e20-280a-4e0a-b495-c0b857_805d8b.pdf].

Submitted: June 7, 2013, Accepted: August 19, 2013

Appendix - mtDNA CO1 barcode gene results with analysis via BLAST and MEGA-5*

[JETxx samples are the author's iBOL (www.boldsystems.com) project on Japanese Earthworms at Guelph Uni., Canada; WOyy samples were attempted by a student at Ewha Womens' Uni. but were mixed in their lab (as lamented in Blakemore, 2013a) requiring resampling with prefix "w" at Seoul National Uni.; Hzz and HYzz are Hanyang Uni. data of current Busan material. Arranged chronologically, some provisional/superseded taxa names are retained for reason of cross-reference, other names - of primary types - are definitive (cf. Fig. 1). Most specimens were collected by author (RJB) some with help of TJ Tansy (TJT) or Yuko Hiramoto (YH)].

*MEGA5 alignments of samples WO2, WO12, WO18, WO22 were reversed for comparison.

> JET112-115|An-460-461|Hikone, Japan, initial id as *Amyntas cf. pingi* (= *A. carnosus*) (RJB)

AACTTTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTCGAATCGAGCTCAGGCCAA
CCGGGCTCCTTTCTGGGAAGGACCAACTATATAACACAATTGTAACAGCACACGCATTCCTTATAATTTTCTTTCTAGTTAT
ACCAGTATTCATTGGGGGATTGGAAAATTGGCTACTACCCTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAAC
AACATGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGAGTAGAGAAAGGAGCAGGTACAGGAT
GAACAGTATATCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGC
TGGGGCATCCTCAATTTGGGGGCCATTAACCTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGA
ATCCCGCTATTTGTATGAGCAGTAGTAATTACTGTCGTAATACTACTATTACTATCATTACCAGTTCTTGCCGGAGCTATTACAAT
ACTACTAACAGACCGAAACCTAAATACATCATTCTTCGACCCAGCGGGAGGGGGGACCCAATTCTGTACCAACACCTATTT

> JET124|Tokyo An-466.1|Nogeyama, Japan *Eisenia japonica* |COI-5P (RJB+TJT)

AACTTTATACTTTTATCTCGGAGTTTGAGCCGGAATAGTGGGAGCTGGAATAAGACTTCTCATTTCGAATTGAACTGAGTCAG
CCGGGAGCCTTCCTAGGAAGAGATCAACTATATAACACAATTGTAACAGCCCACGCATTTGTAATAATTTTCTTCTTAGTTA
TACCTGTATTTATTGGGGGTTTCGAAACTGGTTACTTCTTTAATACTAGGAGCCCCGATATAGCCTTCCCACGACTCAAC
AATATAAGATTCTGACTTCTACCTCCATCCCTCATCTACTTGTATCTCCGCTGCAGTAGAAAAAGGTGCAGGTACAGGAT
GAACAGTATACCCTCCCTATCAAGAAATCTAGCACACGCAGGTCTTCAGTAGATCTAGCCATCTTTTCTTCACTTAGCA
GGAGCTTCTCAATTTGGAGCTATTAATTTTATCACTACAGTTATTAACATACGCTGAAGAGGACTACGATTAGAACGAA
TTCCCTTATTCGTATGAGCCGTAGTGATTACAGTAATTCTATTACTTCTATCTTCCAGTACTTGCAGGAGCTATCACCATA
CTACTAACAGATCGAACTTAAATACTTCACTTCTTCGACCCTGCAGGTGGTGGAGATCCAATTCTATATCAGCATCTCTTC

> JET170|An-417|Enoshima, Japan topotype *Eisenia japonica japonica* |COI-5P (RJB)

AACTTTATACTTTATCTCGGAGTCTGAGCCGGGATAGTGGGTGCTGGTATAAGACTTCTCATTTCGAATTGAACTGAGCCAG
CCGGGAGCCTTCCTAGGAAGAGATCAACTATATAACACAATTGTAACAGCCCATGCATTTGTAATAATTTTCTTCTTAGTTAT
ACCTGTATTCATTGGGGGTTTCGAAACTGGCTACTTCCCTTAAATACTAGGAGCCCCGATATAGCCTTCCCACGACTTAAAC
ATATAAGATTCTGACTACTGCCCCATCCCTTATCTACTAGTATCTCCGCCGAGTAGAAAAAGGTGCAGGTACAGGATG
AACGGTATATCCCTCTATCAAGAAATCTAGCACACGCAGGTCTTCAGTAGATCTAGCCATTTTCTCACTTCACTTATGCGG
GAGCTTCTCAATTTGGGGCTATTAATTTTATCACTACAGTTATTAATATACGCTGAAGAGGACTACGATTAGAACGAAT
CCCTTATTCGTATGAGCTGTAGTAATTACAGTAGTTCTTTTACTTTTATCCCTCCCAGTACTTGCAGGAGCCATTACCATACT
ACTAACAGATCGAACTTAAATACTTCACTTCTTCGATCCCGCAGGTGGTGGAGATCCAATTTTATACCAACATCTTTTC

> JET173|An-415|Hodogaya-ku, Japan holotype *Eisenia japonica hiramoto* |COI-5P (RJB+YH)

GGGGTTTCGAAACTGGTTACTTCTTTAATACTAGGTGCCCCGATATGGCCTTTCACGACTCAACAATATAAGATTCTG
GCTACTACCCCATCCCTCATCTACTCGTATCTCCGCTGCAGTAGAAAAAGGGGCAGGTACGGGATGAACAGTATACCCT
CCCCTATCAAGAAATCTAGCACACGCAGGTCTTCAGTAGATCTAGCCATCTTTTCACTTCACTTAGCAGGAGCTTCTCAAT
TCTTGGAGCTATTAATTTTATCACTACAGTTATCAATATACGCTGAAGAGGCCTACGATTAGAACGAATTCCTTATTCGTAT
GAGCTGTAGTAATTACAGTAATTCTATTACTTCTATCCCTTCCAGTACTTGCAGGAGCCATTACCATATTACTAACAGATCGA
AACTTAAATACCTCATTCTTCGATCCTGCAGG

megaBLAST: shows similarity no better than 84% for lumbricids from Europe.

BLASTn: similarity < 93% (Id=414/445) with Enoshima *E. japonica* topotype An-417.

> WO2 *A. tralfamadore* Blakemore, 2012 holotype ex NIBR Incheon Jeju Gotjawal display (RJB)

GTGTTGGTATAGGATTGGATCTCCCCCTCCTGCTGGATCAAAGAATGATGTATTGAGGTTTCGATCTGTTAGTAATATGGTA
ATTGCCCTGCCAGTACAGGTAGGGACAATAGTAATAGTACTACAGTGATTACTACTGCTCACACAAATAGTGGGATTCGTT
CTAGACGTAACCCAGATCATCGTATGTTAATTACTGTTGTGATAAAGTTAATAGCTCCAAGAATTGACGAGGCACCTGCAAG
ATGAAGTGAGAAAATTGCAAGGTCTACTGAAGGACCGGCATGTGCTATATTTCTTGCTAAGGGTGGGTAAACTGTTCAACCT
GTGCCAGCCCCCTTTTCTACCGCGGCAGATCTTACCAATAAAATTAATGATGGCGGGAGTAGTCAAAATCTTATGTTATTTA
ATCGAGGGAACGCCATATCTGGGGTTCCAAGTATTAGGGGTAGTAACCAGTTTCCGAATCCACCAATAAAATACTGGCATTAC
TAGAAAGAAGATTATTAATAAATGCATGTGCTGTTACAATTGTATTATATAGCTGGTCCCTACCAAGGAAGGATCCAGGTTGT
CTTAACCTCAATACGAATAAGTAGTCTTATTCTGCACCAATTATTCTGCTCAAATTCCTAGAATAAAGTATAGGG

> WO12 *Eisenia sp. (fetida?)* red from Korean mainland (RJB)

ATAAATGTTGGTAGAGAATAGGGTCGCCACCTCCAGCAGGGTCAAAGAATGAGGTATTTAGGTTTCGATCTGTCAATAGTAT
AGTGATAGCTCCCGCAAGTACTGGAAGAGATAAAAAGTAGTAACACCACGGTAATAACTACAGCTCATACAAATAGGGGGAT
TCGTTCTAGTCGAAGCCCACTTCATCGTATGTTAATAACTGTAGTAATGAAGTTAATTGCCCTAAAATTGAGGAGGCACCT
GCTAAATGGAGGGAAAAAATAGCCAGGTCCACTGAGGGCCCCGCGTGCCTAAGTTACTGGATAGGGGTGGGTAAACTGTC
CACCTGTTCAGCACCTTTTCCACTGCAGCAGAGGATACTAGGAGAATTAGGGAAGGGGGCAGAAGTCAAATCTTATG
TTGTTGAGACGTGGAAAGGCTATGTCTGGAGCTCCTAGTATAAGAGGTAGAAGTCAGTTTCCAAATCCACCAATAAATACA
GGTATTACCAGAAAGAAAATTATTACAAATGCATGGGCTGTAACAATTGTATTATATAGTTGGTCCCTTCCCTAGGAAGGCAC
CTGGTTGCCCTTAGCTCGATTGGAATGAGAAGGCTTATACCAGCACCAACCATACTGCTCAGACCCCGAGAATGAAATAGAG

> WO18 *Eisenia sp. (xanthurus?)* red colour Korea 4th April, 2012 (RJB) - *Eisenia xanthurus* (Templeton, 1836) has priority over *E. andrei* Bouche, 1972 (see Blakemore, 2012f; 2013b: 64).

TCAGAATAAATGTTGGTAGAGAATAGGATCGCCACCTCCAGCAGGGTCAAAGAATGAGGTATTCAGGTTTCGATCTGTCAA
TAGTATAGTGATAGCTCCCGCAAGTACTGGAAGAGATAAAAAGTAGTAACACCACGGTAATAACTACAGCTCATACAAATAG
GGGGATTTCGTTCTAGTCGAAGCCCACTTCACCGTATGTTAATAACTGTAGTAATGAAGTTAATTGCCCTAAAATTGAGGAG
GCACCTGCTAAATGGAGGGAAAAAATAGCCAGGTCCACTGAGGGCCCCGCGTGCCTAAGTTACTGGATAGGGGTGGGTAA
ACTGTCCACCTGTTCCAGCACCTTCTCCACTGCAGCAGAGGATACTAGGAGAATTAGGGAAGGGGGCAGAAGTCAAAT
CTTATGTTGTTGAGACGTGGAAAGGCTATGTCTGGAGCTCCAGTATAAGAGGTAGAAGTCAGTTTCCAAATCCACCAATAA
ATACAGGCATAACCAGAAAGAAAATTATTACAAATGCATGGGCTGTAACAATTGTATTGTATAGTTGGTCCCTTCCCTAGGAA
GGCACCTGGTTGCCCTTAGCTCGATTGGAATGAGAAGGCTTATACCAGCACCAACCATACTGCTCAGACCCCGAGAATGAA
ATAGAGA

> WO19 *E. japonica?* large specimen 4th April, 2012 (NIBRIV0000250888) Korea (RJB)

AACTTTATACTTCATTCTAGGGGTTTGAGCTGGTATAGTCGGTGCTGGTATAAGCCTTCTTATTTCGAATTGAGTTAAGACAGC
CAGGTGCCTTTCTAGGCAGAGATCAGCTATATAATAACAATTGTAACAGCTCATGCATTTGTAATAATCTTTTTCTTAGTAATA
CCTGTGTTTATCGGGGGTTTCGAAACTGACTACTTCTCTAATACTAGGAGCCCCGATATGGCATTCCACGACTCAACA
ATATAAGATTTTGACTTCTACCTCCTCCCTCATCCTTCTGGTGTCTTCTGCCCGTAGAAAAAGGTGCAGGTACCGGATGA
ACAGTATATCCACCTCTATCTAGTAATTTAGCCACGCAGGTCTTCAGTAGATTTAGCCATTTTTCTTACACTTAGCAGG
AGCTTCTCAATTTTAGGAGCTATTAATTTTATTACTACAGTCATCAATATACGATGAAGAGGTCTACGTCTAGAACGAATTC
CTTTATTTGTGTGAGCTGTAGTAATTACAGTAATTTTATTATTACTTCTTTACCTGTACTCGCAGGAGCTATACCATACTTC
TAACAGATCGAAATTTAAATACTTCTTTCTTCGACCCTGCTGGTGGTGGAGATCCTATTCTATATCAACATCTTTTT

> WO20 *Eisenia japonica vaga* (paratype) small specimen 4th April, 2012 (IV0000250889)

AACTCTATACTTCATTCTCGGGGTTTGAGCTGGTATAGTGGGCGCAGGTATAAGACTTCTTATTTCGAATCGAACTAAGCCAA
CCGGGAGCTTTTCTAGGGAGAGACCAACTATACAACACAATTTGTGACAGCCCATGCATTTGTAATAATCTTCTTTCTAGTGA
TACCTGTATTTAATTGGGGGTTTGGAAATTGACTTCTACCCCTTATATTAGGAGCTCTGATATAGCCTTCCCACGGCTAAAT
AACATGAGATTTTGACTTTTACCCCATCCCTTATCCTTCTAGTGTCTTCTGCCGCTGTCGAAAAGGGGGCAGGGACGGGAT
GAACAGTATATCCCCCCTATCAAGGAATCTTGCTCATGCGGGCCCTCAGTAGACTTAGCTATTTTTTCTCTGCACTTAGCA
GGAGCTTCTCAATTTCTAGGGGCTATCAACTTTATTACCACAGTTATTAACATACGATGAAGCGGATTACGTTTAGAGCGAA
TCCCTCTATTTGTATGAGCCGTAGTTATCACAGTAGTACTTACTTCTATCCCTGCCAGTCTTGCAGGAGCCATTACCATA
TTATTAACAGACCGAAACCTTAATACCTCATTCTTTGACCCTGCGGGGGGGCAGGACCCCTATCTATATCAACATCTTTTT

> WO22 *Drawida sp.* 2 fat no penis NIBR 3rd April, 2012 (RJB)

CAAAACAAATGCTGATATAAAATAGGGTCGAGACGGAAAAACACCGGGGGGGAAAAAAACTTTAAATTTTCGATCAGTTAA
AAGCATAGTAATTGCACCAGCTAGCACGGGTAAAGATAAAAAGTAATAGTACTACAGTAATTAATAACTCCTCACACAAATAA
TGGAATTCGTTCAAAGTGTATACCCACTCAACGCATATTAATAACAGTTGTAATAAAAATTAATAGCTCCTAAAATAGAAGAT
GCACCCGCCAAGTGTAAAGAAAAAATCGCCAAATCAACAGATGGGCTGCATGTGCTAAATTAAGTCCAGTGGGGGATAA
ACTGTTTATCCGGTACCAGCACCTTTTTCCACTGCTGCTGATGATACTAACAGAATCAAGGCCGGGGCAAAGTCAAATC
TTAAATTTAAGTTCGAGGGAAAGCCATGTCTGGGGCACCAAGTATTAAGGTAGCAATCAATTACCAAAGCCGCCAATAA
ATACAGGCATTACCAAGAAAAAATTATAATAAAGCATGTGCAGTAACAATAGTATTATAAAGCTGATCCCTACCTAAAA
ATGAACCCGGTTGCTTAACTCAATTCGAATTAATAGTCTCATACCCGCACCAATTAATTCGGCTCACACACCTAAAATAAA
ATAAAGAGTCC

> WO23 *Drawida sp.* 1 thin with penis NIBR 3rd April 2012 (RJB)

ACTCTTTATTTTATTTTATTTAGTGTGTGGGCCGGTATAATTGGCGCCGGCATGAGACTATTAATTCGAATTGAGTTAAGACAGCC
GGGCTCATTTTATTTAGTGTAGATCAGCTTTATAATACTATTGTTACTGCACATGCTTTTATTATAATTTTTTTCTTAGTAATGCC
CGTATTTATTTGGTGGCTTTGGTAATTGATTGCTACCTTTAATGCTTGGTGCCCCAGACATGGCTTTTCTCGACTAAATAATTT
AAGATTTGACTTTTACCTCCGGCCTTGATCCTATTAGTATCTTCAGCAGCAGTGGAAAAGGGGGCCGGTACCGGATGAACA

GTTTATCCTCCGCTAGCTAGTAATTTAGCACATGCAGGTGCATCTGTTGATCTGGCAATTTTTCTTTACTTAGCAGGTGC
ATCTTCTATTTTAGGGGCTATTAATTTATTACAACCTGTTATTAATATGCGTTGAGTGGGCATGCATTTTGAACGAATTCCAT
TATTTGTATGGGGAGTTTAACTTACTGTAGTATTACTACTTCTATCTTTGCCCGTGTAGCTGGTGAATTACTATGCTTTTAA
CTGACCGAAATTTAAATACTTCACTTCTCGACCCGCTGGCGGAGGCGATCTATTTTATATCAACATTTATTT

> WO24 *A. carnosus* from NIBR 3rd April, 2012 (RJB)

TTTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTTCGAATCGAGCTCAGGCAACCG
GGCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTTCCTTATAATTTTCTTTCTAGTTATAACC
AGTATTCATTTGGGGGATTTGGAAATTGGCTACTACCACTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAACAAC
ATGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGAGTAGAGAAAGGAGCAGGTACAGGATGAA
CAGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGCTGG
GGCATCCTCAATTTGGGGGCCATTAACCTTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGAATC
CCGCTATTTGTATGAGCAGTAGTAATTACTGTCGTAATACTACTATTACTATCATTACCAGTCTTGGCCGAGCTATTACAATACT
ACTAACAGACCGAAACCTAAATACATCATTCTTTCGACCCAGCGGGAGGGGGGACCCAATTCTGTACCAACACCTATTTTG

> WO27 *D. koreana*? pale Shindo specimen1 (RJB)

TCTTTATTTTATTTTAGGTGTGTGAGCCGGAATAATTGGTGCCGGTATGAGACTATTAATTCGAATTGAGTTAAGACAACCG
GGTTCATTTTLAGGTAGGGATCAGCTTTATAACTATTGTTACTGCACATGCTTTTATTATAATTTTTTCTTTGGTAATGCCT
GTATTTATTGGCGCTTTGGTAATTGATTGCTACCTTAATACTTGGTGCCCCAGACATGGCTTTCCCTCGACTTAATAATTT
AAGATTTTGACTTTTGGCCCGCCCTTGATTCTGTTAGTATCATCAGCAGCAGTGGAAAAAGGTGCTGGTACCGGATGAACA
GTTATCCCCCACTGGCTAGTAATTTAGCACATGCAGGCCCATCTGTTGATTGGCGATTTTTTCTTTACTTTGGCGGGTGC
ATCTTCTATTTTAGGAGCTATTAATTTTATTACAACCTGTTATTAATATGCGTTGAGTGGGTATACACTTTGAACGAATTCCAT
TATTTGTGTGAGGAGTTTAACTTACTGTAGTACTATTACTTTTATCTTTACCCGCTGTAGCTGGTGAATTACTATGCTTTTAA
CTGATCGAAATTTAAATACTTCACTTCTCGACCCAGCTGGTGGGGGCGACCTATTTTATATCAGCATTTGTTT

> WO28 *D. koreana*? blue Shindo specimen2 (=mixed WO32a+WO17) (RJB)

TCTTTATTTTATTTTAGGTGTATGAGCTGGGATAATTGGTGCCGGAATGAGGCTATTAATTCGAATTGAGTTAAGACAACCA
GGTTCGTTTTTAGGCAGAGATCAGCTTTATAACTATTGTTACTGCACATGCTTTTATTATAATTTTTTCTTTAGTAATACCC
GTATTTATTGGTGGCTTTGGTAATTGATTATTACCTTAATACTTGGGGCTCCAGACATGGCTTTCCCTCGACTTAATAATTT
AAGATTTTGGCTCTTACCGCCGGCTTTAATCTTTTAGTGTGCTCAGCAGCAGTGGAAAAAGGGTGTGGTACCGGGTGAACA
GTTTATCCGCTCTGGCTAGTAATTTAGCACATGCGGGGCCATCAGTCGACCTAGCGATTTTTTCTTTACATTTGGCAGGAGC
ATCTTCTATTTTAGGGGCCACTAATTTTATTACAACCTGTTATTAACATACGATGGGTGGCATAACACTTTGAGCGAATTCCGC
TATTCGTGTGGGGAGTTTAACTTACTGTAGTACTATTACTTCTATCTTTGCCTGTGTTGGCCGGCGCAATTACTATGCTTTTAA
CCGATCGAAACTTAAATACTTCACTTCTTGGATCCGCGCGAGGCGGAGACCTATTTTATACCAGCATTTGT

> WO32 *A. carnosus* from Sammock (RJB)

TTTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTTCGAATCGAGCTCAGGCAACCG
GGCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTTCCTTATAATTTTCTTTCTAGTTATAACC
AGTATTCATTTGGGGGATTTGGAAATTGGCTACTACCACTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAACAAC
ATGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGAGTAGAGAAAGGAGCAGGTACAGGATGAA
CAGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGCTGG
GGCATCCTCAATTTGGGGGCCATTAACCTTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGAATC
CCGCTATTTGTATGAGCAGTAGTAATTACTGTCGTAATACTACTATTACTATCATTACCAGTCTTGGCCGAGCTATTACAATACT
ACTAACAGACCGAAACCTAAATACATCATTCTTTCGACCCAGCGGGAGGGGGGACCCAATTCTGTACCAACACCTATTT

> WO34 *A. carnosus* from Sido (RJB)

TTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTTCGAATCGAGCTCAGGCAACCGG
GCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTTCCTTATAATTTTCTTTCTAGTTATAACCA
GTATTCATTTGGGGGATTTGGAAATTGGCTACTACCACTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAACAACA
TGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGAGTAGAGAAAGGAGCAGGTACAGGATGAAC
AGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGCTGGG
GCATCCTCAATTTGGGGGCCATTAACCTTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGAATCCC
GCTATTTGTATGAGCAGTAGTAATTACTGTCGTAATACTACTATTACTATCATTACCAGTCTTGGCCGAGCTATTACAATACTAC
TAACAGACCGAAACCTAAATACATCATTCTTTCGACCCAGCGGGAGGGGGGACCCAATTCTGTACCAACACCTATTTTGA

> WO35 *A. masatake* from NogeYama, Japan (RJB+TJT)

GGAACCTTATACTTTATCTTGGGAATTTGAGCAGGAATAATTGGTGCCGGAATAAGACTACTTATTTCGTATTGAATTAAGAC
AACC GGATCCTTCTTGGTAGAGACCAACTATATAATACAATTGTAACAGCACATGCATTTTAATAATCTTCTTTCTAGTA

ATGCCAGTATTTATTGGTGGATTTCGAAACTGGCTACTGCCCCAATACTTGGAAACCCAGATATGGCGTTCCCCGATTAA
ATAACATAAGATTTTGGACTACTTCCACCATCCCTAATTTTATTAGTAAGATCGGCCCGGGTAGAAAAAGGAGCGGGCACAG
GTTGGACAGTTTACCACCCCTAGCAAGAAATATAGCACATGCCGGCCCTCAGTAGACCTTGAATTTTCTCACTCCATCTT
GCAGGTGCCTCATCAATTCTTGGAGCTATTAACCTTTATCACAACAGTAATTAACATGCGATGATCTGGGCTACGTCTAGAAC
GAATCCCCTACTTTGTATGAGCAGTAGTGATCACCGTAGTACTATTATTATTATCCCTACCTGTACTGGCTGGGGCAATTACC
ATATTACTAACGGATCGAAACCTTAATACATCATTCTTTGACCCAGCAGGAGGGGGGGACCCAGTTCTATACCAACACCTAT
T

> WO44 *E. japonica* from Jeju June 2012 (mixed w18b, WO45) (RJB)

TCTATACTTCATTCTCGGGGTTTGGAGCTGGTATAGTGGGCGCCGGTATAAGACTTCTTATTTCGAATCGAACTAAGCCAACCG
GGAGCTTTTCTAGGGAGAGACCAATTATACAACACAATTGTGACAGCACATGCATTTGTAATAATCTTCTTTCTAGTAATAC
CTGTATTTATCGGGGGTTTGGAAATTGACTTCTACCCCTTATATTAGGAGCTCCTGATATAGCCTTCCCGCGGCTAAATAAC
ATAAGATTTTACTTTTACCCCATCCCTTATCCTTCTAGTATCTTCTGCCGCTGTGCAAAAAGGCGCGGGGACGGGATGAA
CAGTTTATCCCCCTATCAAGAAATCTTGCTCATGCGGGCCCTCAGTAGACCTAGCTATTTTTTCTCTACACTTAGCAGGA
GCTTCTCAATTCTGGGGGCTATCAACTTTATTACCACAGTTATTAATATACGATGAAGCGGATTACGTTTAGAGCGAATCCC
CCTATTTGTATGAGCCGTAGTTATCACAGTAGTACTTACTCTATCCCTACCAGTCTTGACAGGAGCCATTACCATATTAT
TAACAGACCGAAACCTTAATACCTCATTCTTTGACCCGTGACAGGAGCGGAGACCCTATCTTATATCAACATCT

> WO67 *A. carnosus* brown from Jeju (RJB)

GGTCAACAAATCATAAAGATATTGGAACCTTTATATTTTATCTTGGGAATCTGGGACGGAATAATTGGTGGCGGTATGAGACT
ACTTATTGAAATCGAGCTCAGGCAACCGGGCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGC
ATTCCTTATAATTTTCTTCTAGTTATACCAGTATTCATTGGGGGATTGGAAATTGGCTACTACCCTCATACTGGGAACAC
CGGACATAGCATTTCACGCTAAACAACATGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGC
AGTAGAGAAAGGAGCAGGTACAGGATGAACAGTATATCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGA
CCTGGCAATTTTCTCACTCCACTTAGCTGGGGCATCCTCAATTTGGGGGCCATTAACTTTATTACTACAGTAATTAATATGC
GCTGGTCTGGACTACGTCTAGAGCGAATCCCGCTATTTGTATGAGCAGTAGTAATFACTGTGCTACTACTATTACTATCATT
CCAGTTCTTGGCGAGCTATTACAATACTACTAACAGACCGAAACCTAAATACATCATTCTTCGACCCAGCGGGAGGGGG
GACCAATTCTGTACCAACACCTATTTTGATTTTGGTCCACCTGAAGTTA

> w11 *Eisenia sp. (andrei?)* red colour from Jeju (see Blakemore, 2013a) (RJB)

GATATTGGAACCTCTATTTTCACTTCTCGGGTCTGAGCAGGTATGGTTGGTGGTATAAGCCTTCTCATTTCGAATCGAGCT
AAGGCAACCAGGTGCCTTCTTAGGAAGGGACCAACTATATAATAACAATTGTTACAGCCCATGCATTTGTAATAATTTCTTT
CTGGTAATACCTGTATTTATTGGTGGATTGGAACTGACTTCTACCTTATACTAGGAGCTCCAGACATAGCCTTTCCACG
TCTCAACAACATAAGATTTTGGACTTCTGCCCCCTCCCTAATTTCTCCTAGTATCCTCTGCTGAGTGGAAAAGGGTGTGGAA
CAGGGTGGACAGTTTACCACCCCTATCCAGTAACCTAGCGCACGCGGGCCCTCAGTGGACCTGGCTATTTTTCCCTCCA
TTTAGCAGGTGCCTCCTCAATTTTAGGGGCAATTAACCTCATTACTACAGTTATTAACATACGATGAAGTGGGCTTCGACTA
GAACGAATCCCCCTATTGTATGAGCTGTAGTTATTACCGTGGTGTACTACTTTTATCTCTTCCAGTACTTGGGGGAGCTAT
CACTATACTATTGACAGATCGAAACCTAAATACCTCATTCTTTGACCCGTG

> w28b *A. masatake* from Cheyonji, Jeju (see Blakemore, 2012a) (RJB)

AACCTTATACTTTATCCTGGGAATTTGAGCAGGAATAATTGGTGCAGGAATAAGACTACTTATTTCGATTGAATTAAGACAA
CCGGGATCCTTCTTGGTAGAGACCAACTATATAATAACAATTGTAACAGCACATGCATTCTTAATAATCTTCTTTCTAGTAAT
GCCAGTATTTATTGGTGGATTTCGAAACTGGCTACTGCCCCAATACTTGGAAACCCAGATATGGCGTTCCCCCGATTAAAT
AACATAAGATTTTGGACTACTTCCACCATCCCTAATTTTATTAGTAAGATCGGCCCGGGTAGAAAAAGGAGCGGGCACAGGTT
GGACAGTTTACCACCCCTAGCAAGAAATATAGCACATGCCGGCCCTCAGTAGACCTTGAATTTTCTCACTCCATCTTGC
AGGTGCCTCATCAATTCTTGGAGCTATTAACCTTTATCACAACAGTAATTAACATGCGATGATCTGGGCTACGTCTAGAACGA
ATCCCCTACTTTGTATGAGCAGTAGTGATCACCGTAGTACTATTATTATTATCCCTACCTGTACTGGCTGGGGCAATTACCAT
ATTACTAACGGATCGAAACCTTAATACATCATTCTTTGACCCAGCAGGAGGGGGGGACCCAGTTCTATACCAACACCTATTT

> w37 *A. carnosus* from Geoman (RJB)

AACTTTATATTTTATCTTGGGAATCTGGGACGGAATAATTGGTGCAGGAATAAGACTACTTATTTCGAATCGAGCTCAGGCAA
CCGGGCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTCTTATAATTTTCTTTCTAGTTAT
ACCAGTATTCATTGGGGGATTGGAAATTGGCTACTACCCTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAAC
AACATGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGAGTAGAGAAAGGAGCAGGTACAGGAT
GAACAGTATATCCCCCTAGCAAGAAATATTGCTCATGCGGGGCCATCAGTAGACCTGGCAATTTTCTCACTCCACTTAGC
TGGGGCATCCTCAATTTGGGGGCCACTAACTTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGA
ATCCCGCTATTGTATGAGCAGTAGTAATTAAGTGTACTACTATTACTATCATTACCAGTTCTTGGCGGAGCTATTACAAT
ACTACTAACAGACCGAAACCTAAATACATCATTCTTCGACCCAGCGGGAGGGGGGGACCAATTCTGTACCAACACCTATTT

> w54 *A. carnosus/pingi* from Ullungdo (see Blakemore, 2013b, 2103d)

TTATACTTTATCTTAGGAATCTGAGCCGGAATAATTGGTGCTGGTATAAGACTTCTTATTTCGAATTGAGCTCAGACAACCAG
GATCCTTTCTGGGAAGAGATCAACTATATAACACAATTGTAACAGCACACGCATTCCTGATAATTTTCTTTCTAGTTATAACCC
GTATTCATTGGGGGATTGGAATTTGACTACTACCCTTATACTAGGAACACCGGATATAGCATTTCGCGCCTAAACAATA
TGAGATTTTACTATTACCACCATCACTAATCTTACTAGTCTCTCAGCCGCAGTAGAGAAAGGAGCGGGGACAGGATGGAC
AGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGCCATCAGTAGACCTAGCAATTTTCTACTCCACTTAGCTGGA
GCATCCTCAATTTTAGGGGCCATTAACCTTACTACTACAGTAATTAATATGCGCTGGTCTGGACTACGCTAGAGCGGAATTC
CACTATTTGTGTGAGCTGTAGTAATTACTGTGGTGCTACTATTACTATCGTTACCAGTTCCTGCGGAGCTATTACAATACTA
CTAACAGACCGAAACTAAATACATCATTCTTCGACCCAGCGGGAGGGGGGAGACCCAATTTTATACCAACATCTATTT

> w55 *A. carnosus* from Incheon (200 mm) (RJB)

TTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTTCGAATCGAGCTCAGGCAACCGG
GCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTCCTTATAATTTTCTTTCTAGTTATAACCA
GTATTCATTGGGGGATTGGAATTTGGCTACTACCCTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAACAACA
TGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGCAGTAGAGAAAGGAGCAGGTACAGGATGAAC
AGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGCTGGG
GCATCCTCAATTTGGGGGCCACTAATTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGGAATCC
CGTATTTGTATGAGCAGTAGTAATTACTGTCTACTACTATTACTATCATTACCAGTTCCTGCGGAGCTATTACAATACTA
CTAACAGACCGAAACCTAAATACATCATTCTTCGACCCAGCGGGAGGGGGGACCCAATTTCTGTACCAACACCTATTT

> w56 *A. carnosus roki* subsp. n. H from Incheon (300 mm long) (RJB)

CTATACTTTATCTTGGGAATCTGAGCCGGAATAATTGGTGCCGGTATAAGACTCCTTATTTCGAATCGAGCTCAGACAGCCGG
GATCCTTTCTAGGAAGAGATCAGCTATAACAATAAATTGTGACAGCCACGCATTCCTCATAATCTTCTTTCTAGTTATAACCC
GTATTCATCGGGGGTTTGGAAATTTGACTACTACCCTCATACTAGGAACACCGGATATAGCATTTCACGCCTAAACAACA
TGAGATTTTACTATTACCACCATCACTAATCTTACTAGTCTCTTCTGCGGCAGTAGAGAAAGGGGGCGGGAACAGGGTGGAC
AGTATATCCTCCCCTAGCAAGAAATATCGCTCATGCGGGCCATCAGTAGACCTAGCAATTTTCTACTCCACTTAGCTGGG
GCGTCTCAATTTGGGGGCCATTAACCTTTATTACTACAGTAATTAATATGCGCTGATCTGGACTACGTCTAGAACGGATTCC
ACTATTTGTGTGAGCAGTAGTAATCACTGTGGTACTACTATTACTATCATTACCAGTTCCTGCGGAGCTATTACAATACTAC
TAACAGACCGAAACCTAAATACATCATTCTTTGACCCAGCGGGAGGGGGGACCCAATTTTATACCAACACCTATT

megaBLAST result: Id=583/638 (91%) *A. carnosus* (AB543184) from Japan; and similarly from other Korean/Japanese specimens (Fig. 1).

BLASTn: w56 vs. w54 from Ullungdo Id=607/652 (93%), i.e. different taxa.

H4 vs w56 large “*A. carnosus*” from Incheon (300 mm long)=653/653 (100%), i.e. same taxon and thus meriting a new name as these differ from other Korean *A. carnosus*.

> w57 *A. carnosus* from Incheon (280 mm) (RJB)

TTATATTTTATCTTGGGAATCTGGGCAGGAATAATTGGTGCCGGTATGAGACTACTTATTTCGAATCGAGCTCAGGCAACCGG
GCTCCTTTCTGGGAAGGGACCAACTATATAACACAATTGTAACAGCACACGCATTCCTTATAATTTTCTTTCTAGTTATAACCA
GTATTCATTGGGGGATTGGAATTTGGCTACTACCCTCATACTGGGAACACCGGACATAGCATTTCACGCCTAAACAACA
TGAGATTCTGGCTACTGCCCCATCACTAATCTTACTAGTATCTTCCGCCGCAGTAGAGAAAGGAGCAGGTACAGGATGAAC
AGTATATCCCCCCTAGCAAGAAATATTGCTCATGCGGGCCATCAGTAGACCTGGCAATTTTCTACTCCACTTAGCTGGG
GCATCCTCAATTTGGGGGCCATTAACCTTTATTACTACAGTAATTAATATGCGCTGGTCTGGACTACGTCTAGAGCGGAATCC
GCTATTTGTATGAGCAGTAGTAATTACTGTCTACTACTATTACTATCATTACCAGTTCCTGCGGAGCTATTACAATACTAC
TAACAGACCGAAACCTAAATACATCATTCTTCGACCCAGCGGGAGGGGGGACCCAATTTCTGTACCAACACCTATT

> H1 *Drawida songae yeongdo* subsp. n. holotype (RJB)

TCTTTATTTTATTTAGGTGTATGAGCTGGAATAATTGGAGCAGGGATAAGTCTTCTTATTTCGTATCGAATTAAGACAACCAG
GTGCCTTTTAGGAAGCGATCAACTATATAACACTATTGTAACAGCACATGCTTTTATTATAATTTTTTCTTAGTAATACCT
GTTTTTATTGGTGGGTTTCGGAAATTTGATTACTACCTTAAATATTAGGAGCTCCAGATATAGCTTTCCACGACTTAATAATCT
AAGATTTGACTTCTTCTCTGCTCTTATTTTATTAGTTTCATCCGCAGCAGTAGAAAAAGGTGCTGGTACAGGATGAACAG
TTTATCTCCACTAGCTAGAAACATTTGCTCATGCTGGACCATCAGTAGACCTTGCTATTTTTCATTACATTTAGCTGGGGCT
TCATCAATTTTAGGCGCCATTAATTTTATTACAACAGTAATTAATATACGTTGATCAGGGCTTCAATTAGAACGAAATCCCAT
ATTTGTATGGGCTGTCTTTATTACAGTAATTTTATTACTTTTATCTCTTCTCTGTTTTAGCAGGTGCTATTACTATATTATTAAC
AGACCGAAATCTAAACACATCATTTTTTGACCCTGCAGGTGGAGGGGGACCCAATTTTATACCAACACCTATT

megaBLAST result: *Amyntas* sp. 14 JS-2012 (JQ982486), 83%, i.e., nothing similar on GenBank.

> H2 *Drawida songae yeongdo* (paratype, P), an immature specimen (RJB)

TCTTTATTTTATTTAGGTGTATGAGCTGGAATAATTGGAGCAGGGATAAGTCTTCTTATTTCGTATCGAATTAAGACAACCAG
GTGCCTTTTAGGAAGCGATCAACTATATAACACTATTGTAACAGCACATGCTTTTATTATAATTTTTTCTTAGTAATACCT

GTTTTATTGGTGGGTTTCGGAAATTGATTATTACCTTTAATATTAGGAGCTCCAGATATAGCTTTCCCACGACTTAATAATCT
 AAGATTTTGACTTCTTCCCTCCTGCTCTTATTTTATTAGTTTCATCCGCAGCAGTAGAAAAAGGTGCTGGTACAGGATGAACAG
 TTTATCCTCCACTAGCTAGAAAACATTGCTCATGCTGGACCATCAGTAGACCTTGCTATTTTTTTCATTACATTTAGCTGGGGCT
 TCATCAATTTTAGGCGCCATTAATTTTATTACAACAGTAATCAATATACGTTGATCAGGGCTTCAATTAGAACGAATCCCATT
 ATTTGTATGGGCTGTCTTTATTACAGTAATTTTATTACTTTTATCTCTTCTGTTTTAGCAGGTGCTATTACTATATTATTAAC
 AGACCGAAATCTAAACACATCATTTTTTGACCCTGCGGGTGGAGGGGACCCAATTTTATACCAACACCTATTC

megaBLAST result: Glossoscolecidae sp. (sic!) voucher EW-SJ-498 (GU013845), 83%.

BLASTn: H1 vs. H2=651/655 (99%), i.e. sufficiently similar to class as same taxon.

> **H3 *Amyntas masatacae*** Busan larger (180 mm) specimen (RJB)

TTATACTTTATCCTGGGAATTTGAGCAGGAATAATTGGTGCAGGAATAAGACTACTTATTCGTATTGAATTAAGACAACCGG
 GATCCTTCCCTGGTAGAGACCAACTATATAATAACAATTGTAACAGCACATGCATTCTTAATAATCTTCTTTCTAGTAATGCCA
 GTATTTATTGGTGGATTTCGGAAACTGGCTACTGCCCCAATACTTGGAAACCCAGATATGGCGTTCCCCCGATTAAATAACA
 TAAGATTTGACTACTTCCACCATCCCTAATTTTATTAGTAAGATCGGCCCGGTTAGAAAAAGGAGCGGGCACAGGTTGGAC
 AGTTTACCCACCCCTAGCAAGAAATATAGCACATGCCGGCCCTTCAGTAGACCTTGCAATTTTCTCACTCCATCTTGCAGGT
 GCCTCATCAATCTTGGAGCTATTAACCTTTATCACAAACAGTAATTAACATGCGATGATCTGGGCTACGTCTAGAACGAATCC
 CACTATTTGTATGAGCAGTAGTGATCACCGTAGTACTATTATTATCCCTACCTGTACTGGCTGGGGCAATTACCATATTA
 CTAACGGATCGAAACCTTAATACATCATTCTTTGACCCAGCAGGAGGGGGGACCCAGTTCTATACCAACACCTATTT

megaBLAST result: Closest Id.=99% *Amyntas triastriatus* voucher 06-211 (EF077537) from China - these Chinese misidentifications, as expected.

BLASTn: H3 vs. WO35 *A. masatacae* from Nogyama (IV0000251098), Id.=653/653 (100%) and ditto vs. w28b *A. masatacae* from Cheyonji, Jeju (see Blakemore, 2013a), i.e., all same taxon.

H3 vs. WO2 *A. tralfamadore* holotype from Incheon (ex Jeju), Id.=608/647 (94%), i.e., different taxa (see data for >HY2 barcode below).

H3 vs. *M. ryunome* Blakemore, 2012, Id.=553/652 (85%) i.e. quite different taxa.

> **H4 *Amyntas carnosus roki*** subsp.n. (paratype, P) from Busan (RJB)

ACTCTATACTTTATCTTGGGAATCTGAGCCGGAATAATTGGTGCAGGATAAAGACTCCTTATTCGAATCGAGCTCAGACAGC
 CGGGATCCTTTCTAGGAAGAGATCAGCTATAACAATAAATTGTGACAGCCCACGCATTCTCATAATCTTCTTTCTAGTTATA
 CCCGATTCATCGGGGGTTTGGAAATTGACTACTACCCTCATACTAGGAACACCGGATATAGCATTTCCACGCCTAAACA
 ACATGAGATTTGACTATTACCACCATCACTAATCTTACTGGTCTTCTGCGCAGTAGAGAAGGGGGCGGGAACAGGGTG
 GACAGTATATCCTCCCTAGCAAGAAATATCGCTCATGCGGGGCCATCAGTAGACCTAGCAATTTTCTCACTCCACTTAGCT
 GGGGCGTCTCAATTTTGGGGCCATTAACCTTTTACTACAGTAATTAATATGCGCTGATCTGGACTACGTCTAGAACGGA
 TTCCACTATTTGTGTGAGCAGTAGTAATCACTGTGGTACTACTATTACTATCATTACCAGTTCTTGGCGGAGCTATTACAATA
 CTAATAACAGACCGAAACCTAAATACATCATTCTTTGACCAGCGGGAGGAGGGGACCCAATTTTATACCAACACCTATTT

> **H5 *Eisenia japonica vaga*** subsp. n. (holotype, H) from Busan (RJB)

TCTATACTTCACTTCTCGGAGTTTGGAGCTGGTATAGCGGGCGCAGGTATAAAGACTTCTTATTCGAATCGAATAAGCCAACCG
 GGAGCTTTTCTAGGGAGAGACCAACTATAACAACAATTGTGACAGCACATGCATTTGTAATAATCTTCTTTCTAGTAATAC
 CTGTATTTATTGGGGTTTGGAAATTGACTTCTACCCCTTATATTAGGAGCTCCTGATATAGCCTTCCCACGGCTAAATAAC
 ATGAGATTTGACTTTTACCCCATCCCTTATCCTTCTAGTGTCTTCTGCGCTGTGAAAAGGGGGCAGGAACGGGATGAA
 CAGTATATCCCCCTATCAAGGAATCTTGTCTATGCGGGCCCTCAGTAGACTTAGCTATTTTTTCTCTGCCTTAGCAGGA
 GCTTCTCAATTTAGGGGCTATCAACTTTTATTACCACAGTTATTAACATACGATGAAGCGGATTACGTTTAGAGCGAATCC
 CTCTATTTGTATGAGCCGATGTTATCACAGTAGTACTTACTTCTATCCCTGCCAGTCTTGCAGGAGCCATTACCATATTA
 TTAACAGACCGAAACCTTAATACCTCATTCTTTGACCCTGCGGGGGCGGAGACCCATCCTATATCAACATCTTTTT

megaBLAST result: Identities=93% Lumbricidae sp. (sic) Esik52 (AB607078); 92% *Eisenia japonica* from Japan (GenBank AB542698).

BLASTn: H5 vs. WO20 "*E. japonica*" from 4th April, 2012 survey on Korea=650/655 (99%), i.e. a similar and possibly same taxon but both different from *E. japonica* from Japan, thus probably meriting a new name.

H5 vs. JET 170-11 *E. japonica japonica* topotype Identities=540/652 (83%);

H5 vs. JET 173-11 *E. japonica hiramoto* holotype Id.=363/445 (82%) i.e. dissimilar taxa.

> **H6 *Eisenia fetida*** Busan specimen (RJB)

TCTTTATTTTATTCTTGGGGTTTGGGCAGGAATAGTAGGTGCTGGTATGAGGCTCCTTATTCGAATTGAGCTAAGACAACCA
 GGTGCCTTCCCTAGGAAGTGACCAATTATACAATAAATTGTACAGCCCATGCATTCGTGATAATTTTCTTTCTTGTATACC
 TGTTTTTATTGGTGGATTGGAAATTGGTTGCTACCTTATACTGGGGGCTCCAGATATAGCTTTCCCACGTCTAAATAACA
 TAAGCTTCTGACTTCTGCCCCCTCTCTAATTTACTAGTCTCCTCCGCTGCAGTAGAAAAGGGGGCTGGGACAGGATGAAC
 GGTATATCCCCCTATCCAGTAATTTAGCCCATGCTGGGCCCTCAGTAGACCTAGCCATTTTTTCTTTACTTAGCAGGTG
 CTCTCAATTTCTCGGAGCAATTAATTTTATTACCACAGTAATCAACATACGATGAAGGGGGCTTGCAGTAGAACGAATTC
 CCTATTTGTTTGGAGCAGTAGTTATTACAGTAGTACTTACTTCTATCCTTACCAGTTCTTGTGCGGGCTACTACTACTTTT

AACAGATCGAAACCTCAACACCTCATTCTTTGATCCTGCTGGTGGTGGGGATCCCATTCTATACCAACATCTATTT

megaBLAST result: *Eisenia fetida* strain (sic!) Ef2 (FJ214216), 100%.

BLASTn: H6 vs. w11 *Eisenia* sp. red from Jeju (same as WO12?)=527/618 (85%), different taxa but apparently the closest of other Korean “*E. fetida*” from current studies (see Blakemore, 2012a-c). These two species are part of the *E. fetida* spp-complex (as per Blakemore, 2010) but cannot currently be associated definitively with any of the 15+members (as listed by Blakemore, 2002, 2010, 2012f, 2013b, c).

Cf. other *E. fetida* spex. eg. WO12, WO18, w11... and from Mongolia (Blakemore, 2013c).

> **H7 *Drawida cf. koreana?*** Busan specimen (RJB)

TCTTTATTTTATTTTAGGTGTGTGGGCCGGTATAATTGGCGCCGGCATGAGACTATTAATTTCGAATTGAGTTAAGACAGCCG
GGCTCATTTTATTTAGGTAGAGATCAGCTTTATAATACTATTGTTACTGCACATGCTTTTATTATAATTTTTTCTTAGTAATGCC
GTATTTATTGGTGGCTTTGGTAATTGATTGCTACCTTAAATGCTTGGTGCCCCAGACATGGCTTTTCTCGACTAAATAATTT
AAGATTTTGACTTTTACCTCCGGCCTTGATCCTATTAGTATCTTCAGCAGCAGTGGAAAAGGGGGCCGGTACCGGATGAACA
GTTTATCCTCCGCTAGCTAGTAATTTAGCACATGCAGGTGCATCTGTTGATCTGGCAATTTTTTCTTTACACTTAGCAGGTGC
ATCTTCTATTTAGGGGCTATTAATTTTATTACAACCTGTTATTAATATGCGTTGAGTGGGCATGCATTTTGAACGAATTCCAT
TATTTGTATGGGGAGTTTAATTACTGTAGTATTACTACTTCTATCTTTGCCCGTGTAGCTGGTGAATTACTATGCTTTTAA
CTGACCGAAATTTAAATACTTCACTTCTCGACCCCGCTGGCGGAGGCGATCCTATTTTATATCAACATTTATTT

megaBLAST result: *Drawida* sp. from Japan (AB592437), 92%.

BLASTn: H7 vs WO23 *Drawida* sp. from NIBR 3rd April thin longer=655/655 (100%), i.e. same taxon, but the question now is: Is it *D. koreana* proper? Cf. WO22 NIBR 3rd April, 2012.

> **HY2 *M. ryunome*** Blakemore, 2012; small (116 mm), prostatic Busan specimen (RJB)

TATACTTTATTTAGGAATTTGAGCCGGAATAATCGGAGCTGGGATAAGCCTACTTATCCGCATTGAACTAAGTCAACCGGG
GTCTTTCCTTGGAAGAGACCAGTTATATAATACGATTGTAACAGCACATGCATTCCCTCATAATTTTCTTCTTAGTAATACCAG
TATTTATTGGGGGGTTTCGAAACTGGTTGCTACCACTAATACTAGGAACACCAGATATAGCATTCCCCGACTAAATAACAT
AAGATTTTGACTACTCCCCCTTCCCTAATTCTCCTAGTGAGATCAGCTGCCGTAGAAAAGGAGCAGGTACAGGTTGAACA
GTATACCCACCCCTAGCAAGAAATATAGCACATGCGGGCCCTCAGTAGATCTTGCAATCTTCTACTACATTTAGCAGGTG
CCTCGTCAATTTTAGGAGCTATTAATTTTATTACCACAGTGATCAATATACGATGGTCAGGACTACGACTAGAACGAATTCC
ATTATTTGTTTGAGCAGTAATAATTACTGTAGTACTACTACTATTATCACTCCCTGTACTAGCCGGTGCAATTACTATACTAC
TAACAGACCGAAATCTTAACACATCCTTCTTTGATCCAGCTGGTGGTGGAGACCCAATTCTATACCAACACTTATTC

megaBlast result: *Amyntas incongruus* (EF077551 from China), Id. (99% - misidentified).

BLASTn result: HY2 *Metaphire ryunome* Busan vs. *M. ryunome* H from Hikone (Tokyo An-457); results: Id. 653/653 (100%), i.e. same taxon - its wide distribution now allows concession of possibility to yield to prior records of some as yet unknown species from Asia.

HY2 *M. ryunome* vs. H3 *Amyntas masatacae* Busan specimen (180 mm) no prostates; results: Id. 553/652 (85%), i.e., different taxa as 15% dissimilar COI gene.

Note: These latter data are not shown in Fig. 1 phylotree.