Zootaxa 3774 (4): 324–332
www.mapress.com/zootaxa/

Article

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)

http://dx.doi.org/10.11646/zootaxa.3774.4.2
http://zoobank.org/urn:lsid:zoobank.org:pub:1E9E30D8-392F-454B-B6F6-68E2276B35F9

New species of earthworms belonging to the *Metaphire formosae* species group (Clitellata: Megascolecidae) in Taiwan

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Abstract

The *Metaphire formosae* species group is a member of the *Pheretima* complex of the family Megascolecidae. It is composed of 12 nominal taxa, *Metaphire bununa* Tsai *et al.*, 2000, *Metaphire feijani* Chang & Chen, 2004, *Metaphire formosae* (Michaelsen, 1922), *Metaphire glareosa* Tsai *et al.*, 2000, *Metaphire nanaoensis* Chang & Chen, 2005, *Metaphire paiwanna paiwanna* Tsai *et al.*, 2000, *Metaphire paiwanna hengchunensis* (James *et al.*, 2005), *Metaphire paiwanna liliumfordi* Tsai *et al.*, 2000, *Metaphire tahanmonta* Chang & Chen, 2005, *Metaphire taiwanensis* Tsai *et al.*, 2004, *Metaphire trutina* Tsai *et al.*, 2003, and *Metaphire yuhsi* (Tsai, 1964). In this study, we describe a new species, *Metaphire tengjhihensis* **sp. nov.**, and two new subspecies, *Metaphire nanaoensis truku* **ssp. nov.** and *Metaphire taiwanensis tsaii* **ssp. nov.**, belonging to this species group. DNA barcodes (partial sequences of the mitochondrial cytochrome *c* oxidase subunit 1, COI) from type specimens of *M. feijani*, *M. tengjhihensis* **sp. nov.**, *M. nanaoensis truku* **ssp. nov.**, *M. tahanmonta* and *M. taiwanensis tsaii* **ssp. nov.** have been deposited in GenBank in previous studies and are explicitly linked to the type specimens for the first time, enabling unambiguous identification using both morphology and DNA barcodes. Finally, we comment on the systematics of the *M. formosae* species group and suggest an integrative taxonomic approach that combines morphology and DNA barcodes for future descriptions of new species of *Amynthas* and *Metaphire*.

Key words: *Amynthas*, *Pheretima* complex, systematics, taxonomy

Introduction

The *Metaphire formosae* species group (Chang *et al.* 2008) of the *Pheretima* complex within the family Megascolecidae is an earthworm group endemic to Taiwan. With ten new species described since 2000, this group is currently composed of 12 species or subspecies: *Metaphire bununa* Tsai *et al.*, 2000, *Metaphire feijani* Chang & Chen, 2004, *Metaphire formosae* (Michaelsen, 1922), *Metaphire glareosa* Tsai *et al.*, 2000, *Metaphire nanaoensis* Chang & Chen, 2005, *Metaphire paiwanna paiwanna* Tsai *et al.*, 2000, *Metaphire paiwanna hengchunensis* (James *et al.*, 2005), *Metaphire paiwanna liliumfordi* Tsai *et al.*, 2000, *Metaphire tahanmonta* Chang & Chen, 2005, *Metaphire taiwanensis* Tsai *et al.*, 2004, *Metaphire trutina* Tsai *et al.*, 2003, and *Metaphire yuhsi* (Tsai, 1964) (see also Chang & Chen 2005b, 2008; Chang *et al.* 2008, 2009; Tsai *et al.* 2009). These species are gigantic anecic earthworms with lengths around 30 cm or longer and widths about 1 cm. In addition to their large body size, these species share many other similarities in their morphology: all of them are octothecate species with similar but distinguishable spermathecae in 6–9, and have racemose prostate glands, simple caeca, and male pores characterized by one or two genital pads or their vestiges. They generally have an allopatric distribution throughout the forested mountain areas in Taiwan, from hills to an elevation of about 3,000 m (Chang *et al.* 2009). The systematics of this species group was recently revised, and the monophyly of the group and each species and subspecies within it is supported by molecular data (Chang & Chen 2005b; Chang *et al.* 2008).

This study describes a new species and two new subspecies of *M. taiwanensis* and *M. nanaoensis*, respectively, reported by Chang *et al.* (2008) in their molecular phylogenetic analyses. The current text of external and internal

 Accepted by R. Schmelz: 31 Jan. 2014; published: 7 Mar. 2014

"degradation", which makes it difficult in some cases to decide between presence or absence of that character. James prefers to restrict *Metaphire* to species with well-developed copulatory pouches (James 2005; James *et al.* 2005) and assigns species with "degraded" pouches to *Amynthas*, while Blakemore (2010) prefers to restrict *Amynthas* to species with superficial male pores, arguing that all species with the derivative character state of non-superficial male pores should belong to *Metaphire*. As a result, species in the *formosae* species group were assigned to *Amynthas* and *Metaphire* by James and Blakemore, respectively. We follow the conventional practice of assigning species in the *formosae* species group to *Metaphire* (Chang & Chen 2004, 2005a, b; Tsai et al. 2000, 2003, 2004), but recognize the different forms of copulatory pouches presented by Easton (1979) and James (James 2005; James *et al.* 2005), particularly the intramural and intracoelomic ones. Furthermore, we doubt that the assignment of species with the intramural form and the various "degradations" of both forms to either *Amynthas* or *Metaphire* is satisfying regarding preserving information of character states and evolution. Recent molecular phylogenetic studies strongly suggest that *Metaphire* is not monophyletic (Chang *et al.* 2008; James 2005); it is indeed polyphyletic (unpublished data) regardless of difference in opinions on the definition of *Metaphire* or *Amynthas*. Considering this nature and the viewpoints already discussed by the above authors, any further arguments on the *Amynthas*-or-*Metaphire* issue seem meaningless before someone takes on the task of revising the whole group.

Hypotheses regarding synonyms of earthworms can usually be tested by comparing DNA barcodes. Blakemore *et al.* (2010) further recommended that any new earthworm taxa described should be accompanied by DNA barcodes from types to meet current standards. Although some of the DNA barcodes published by Chang and Chen and their coauthors (Chang & Chen 2005b; Chang *et al.* 2008) are from type specimens of the *M. formosae* species group, this connection has never been established either explicitly or implicitly until the present study (Table 1). DNA barcodes from non-type specimens are also available for all species and subspecies in the *M. formosae* species group (see Chang *et al.* 2008). These genetic data, together with all the published morphological descriptions, enable unambiguous identification.

Drawing a line between intraspecific and interspecific morphological variations is sometimes difficult in *Amynthas* and *Metaphire*. The studies regarding the *M. formosae* species group published after 2000 (as cited in this study) have collectively demonstrated an integrative taxonomic approach through which hypotheses regarding species are tested using morphological, molecular, biogeographical and, to some extent, ecological data. Considering the complexity of morphological variations and the huge numbers of described (and undescribed) species in *Amynthas* and *Metaphire*, and in order to prevent interminable arguments about synonyms, we strongly suggest preserving DNA-friendly samples on a regular basis and using an integrative taxonomic approach that combines morphological and molecular data when it comes to describing a new species of *Amynthas* or *Metaphire* that may raise debates about synonyms.

Acknowledgements

We are grateful to the people who helped us during field surveys, especially Y.-H. Lin, I-H. Chen, S.-P. Wu and C.-C. Huang. We thank H.-P. Shen and Y.-H. Wang for their help in inspecting type specimens, and H.-P. Shen and S. James for their comments on an earlier draft of this manuscript. This study was mainly supported by the National Science Council (NSC92-2621-B-002-019), Taiwan and partly supported by National Science Foundation (EAR-0748442), USA.

References

Blakemore, R.J. (2010) Unravelling some Kinki earthworms (Annelida: Oligochaeta: Megadrili: Megascolecidae) - Part II. *Opuscula Zoologica Budapest*, 41, 191–206.

Blakemore, R.J., Kupriyanova, E.K. & Grygier, M.J. (2010) Neotypification of *Drawida hattamimizu* Hatai, 1930 (Annelida, Oligochaeta, Megadrili, Moniligastridae) as a model linking mtDNA (COI) sequences to the 'Can of Worms' theory of cryptic species. *ZooKeys*, 41, 1–29.
http://dx.doi.org/10.3897/zookeys.41.374

Chang, C.-H. & Chen, J.-H. (2004) A new species of earthworm belonging to the genus *Metaphire* Sims and Easton 1972 (Oligochaeta: Megascolecidae) from southern Taiwan. *Taiwania*, 49, 219–224.

Chang, C.-H. & Chen, J.-H. (2005a) Three new species of octothecate pheretimoid earthworms from Taiwan, with discussion on the biogeography of closely related species. *Journal of Natural History*, 39, 1469–1482. http://dx.doi.org/10.1080/00222930400004586

Chang, C.-H. & Chen, J.-H. (2005b) Taxonomic status and intraspecific phylogeography of two sibling species of *Metaphire* (Oligochaeta: Megascolecidae) in Taiwan. *Pedobiologia*, 49, 591–600. http://dx.doi.org/10.1016/j.pedobi.2005.07.002

Chang, C.-H. & Chen, J.-H. (2008) *Earthworm Fauna of Taiwan:* Metaphire formosae *species group*. National Taiwan University Press, Taipei, Taiwan, 60 pp.

Chang, C.-H., Lin, S.-M. & Chen, J.-H. (2008) Molecular systematics and phylogeography of the gigantic earthworms of the *Metaphire formosae* species group (Oligochaeta: Clitellata: Megascolecidae). *Molecular Phylogenetics and Evolution*, 49, 958–968. http://dx.doi.org/10.1016/j.ympev.2008.08.025

Chang, C.-H., Shen, H.-P. & Chen, J.-H. (2009) *Earthworm Fauna of Taiwan*. National Taiwan University Press, Taipei, Taiwan, 174 pp.

Easton, E.G. (1979) A revision of the "acaecate" earthworms of the *Pheretima* group (Megascolecidae: Oligochaeta): *Archipheretima*, *Metapheretima*, *Planapheretima*, *Pleionogaster*, and *Polypheretima*. *Bulletin of the British Museum of Natural History (Zoology)*, 35, 1–126.

ICZN (1999) International Code of Zoological Nomenclature (4th edition). International Trust for Zoological Nomenclature, c/o Natural History Museum, London, 306 pp.

James, S.W. (2005) Preliminary molecular phylogeny in the *Pheretima* group of genera (Crassiclitellata: Megascolecidae) using Bayesian analysis. *In*: Pop, V.V. & Pop, A.A. (Eds.), *Advances in earthworm taxonomy II (Annelida: Oligochaeta)*. Cluj University Press, Cluj-Napoca, Romania, pp. 129–142.

James, S.W., Shih, H.-T. & Chang, H.-W. (2005) Seven new species of *Amynthas* (Clitellata: Megascolecidae) and new earthworm records from Taiwan. *Journal of Natural History*, 39, 1007–1028. http://dx.doi.org/10.1080/00222930400001434

Michaelsen, W. (1922) Oligochaeten aus dem Rijks Museum van Natuurlijke Historie zu Leiden. *Capita Zoologica*, 1, 1–68. [in German]

Sims, R.W. & Easton, E.G. (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. http://dx.doi.org/10.1111/j.1095-8312.1972W.tb00694.x

Tsai, C.-F. (1964) On some earthworms belonging to the genus *Pheretima* Kinberg collected from Taipei area in north Taiwan. *Quaternary Journal of Taiwan Museum*, 17, 1–35.

Tsai, C.-F., Tsai, S.-C. & Liaw, G..-J. (2000) Two new species of protandric pheretimoid earthworms belonging to the genus *Metaphire* (Megascolecidae: Oligochaeta) from Taiwan. *Journal of Natural History*, 34, 1731–1741. http://dx.doi.org/10.1080/00222930050122156

Tsai, C.-F., Chen, J.-H., Tsai, S.-C. & Shen, H.-P. (2003) A new species of the earthworm belonging to the genus *Metaphire* Sims and Easton (Megascolecidae: Oligochaeta) from the northeastern Taiwan. *Endemic Species Research*, 5, 83–88.

Tsai, C.-F., Tsai, S.-C. & Shen, H.-P. (2004) A new gigantic earthworm of the genus *Metaphire* Sims and Easton (Megascolecidae: Oligochaeta) from Taiwan with reference to evolutional trends in body sizes and segment numbers of the *Pheretima* genus-group. *Journal of Natural History*, 38, 877–887.

Tsai, C.-F., Shen, H.-P., Tsai, S.-C., Lin, K.-J., Hsieh, H.-L. & Yo, S.-P. (2009) A checklist of oligochaetes (Annelida) from Taiwan and its adjacent islands. *Zootaxa*, 2133, 33–48.