A New Species of the Terrestrial Earthworm of the Genus *Metaphire*Sims and Easton, 1972 from Thailand with Redescription of Some Species

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ABSTRACT.— A project on an inventory of earthworm fauna in Nan province, north Thailand and nearby areas has been carried out since 2009, with a focus on the two common genera *Amynthas* Kinberg, 1867 and *Metaphire* Sims and Easton, 1972. In this paper we report on the occurrence of species of the genus *Metaphire*. The cosmopolitan species *Metaphire peguana* (Rosa, 1890) was found in all areas and nearby localities. For the following four widely-distributed species we provide morphological data: *M. anomala* (Michaelsen, 1907), *M. birmanica* (Rosa, 1888), *M. houlleti* (Perrier, 1872), *M. posthuma* (Vaillant, 1868). Morphological divergences from previous records of these common species are noted, but the available data do not support description of new taxa. One new species, *Metaphire grandipenes* n.sp. is so named because of the extreme expansion of ventral segment xviii and the male porophores protruded as large two penes or large two knobs. It has large genital markings and genital marking glands at xx. This new species is compared to *M. abdita* (Gates, 1935).

KEY WORDS: Earthworm, new species, Metaphire, Thailand

INTRODUCTION

The earthworm inventories in Thailand have been recorded over seventy years ago when Gates (1939) listed 24 species of the family Megascolecidae, most of which belong to the genera Amynthas Kinberg, 1867 and Metaphire Sims and Easton, 1972. The two genera are very common in Southeast Asia and are found coexisting almost everywhere (Gates, 1972). The identification of the two genera is quite complicated because they are so similar, differing by the presence of copulatory pouches containing the male pores in Metaphire, but male pores are superficial in Amynthas (Sims and Easton, 1972). The genus Metaphire is the second largest group after *Amynthas*, and was reported to have 112 nominal species in 13 species groups (Sims and Easton, 1972). Subsequent descriptions from various parts of the world increased the diversity of *Metaphire* to almost 200 species (Blakemore, 2006).

The genus *Metaphire* is one of the dominant terrestrial earthworm genera throughout Thailand and nearby countries. Following the summaries by Gates (1939, 1972) 6 *Metaphire* species have been reported from Thailand: *M.anomala*, *M. bipora*, *M. peguana*, *M. planata*, *M. posthuma*, and *M. virgo*. However, in addition Kosavititkul (2005) has reported two species of *Metaphire* from Khao Yai National Park, which included an unknown species, Chantaravisoot (2007) reported four

species of Metaphire from various areas in Thailand that were all commented to be new to science, and Somniyam (2009) recorded species from Nakhon five *Metaphire* Ratchasima province of which many are still unidentified. Recent publications from other SE Asian areas, include Tsai et al. (2004) who described a new gigantic earthworm (Metaphire taiwanensis) from Taiwan. Shen and Yeo (2005) reported three Metaphire species in Singapore. From the above data it is clear that there are still many species waiting to be discovered and described. The Animal **Systematics** Research Chulalongkorn University's members have surveyed terrestrial earthworms throughout Thailand since 2005 and a part of their results has been summarized in Chantaravisoot (2007). In the present paper we describe an additional new species of the genus Metaphire and redescribe five previously known species collected from Nan province, north Thailand. The new species is known only from the type locality, but as more intensive collecting is undertaken in Thailand and other Asian countries, the known range and habitats of these species may be extended. The habitat of new species was in the topsoil layer covered with leaf litter of deciduous forests. The localities were in Nan province, in the north of Thailand, as shown in Figure 1.

MATERIALS AND METHODS

An inventory of earthworm fauna in Nan province in northern Thailand and nearby areas started in 2009. The collecting has been carried out in various ecological settings such as limestone karst, granitic mountains, along the Nan River and tributaries, small canals, ponds, swamps, farms and rice paddy fields. Earthworms were collected by carefully digging up the

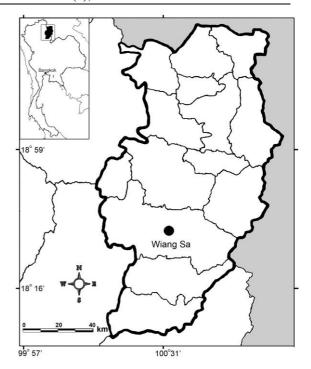


FIGURE 1. Map of type locality of *Metaphire grandipenes* n. sp. Ban Bun Rueng, Lainan Subdistrict, Wiang Sa District, Nan Province.

topsoil near casts and by hand sorting the leaf litter. The worms were killed in 30% (v/v) ethanol, photographed, transferred to 4% (w/v) formalin for fixation for approximately 12 hours, and then transferred to 70% (v/v) ethanol for longer term preservation and subsequent morphological studies.

The descriptions of each species were made during observation under a Stemi DV 4 ZEISS stereoscopic light microscope. Drawings were made of the body segments and the distinct external characters and internal organs, as mentioned above, and are shown in Figures 2–6 for the five described and Figure 7 for a new species, respectively. The number of segments and the body width and length were measured in both full adults and juveniles, and are presented as the range (min-max) and mean ± standard deviation.

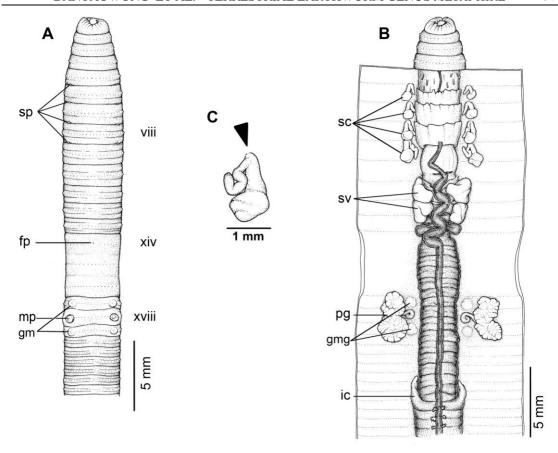


FIGURE 2. External and internal morphology of *Metaphire posthuma*. (A) External ventral view, (B) internal dorsal view and (C) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore.

Holotype and paratype specimens are deposited in the Chulalongkorn University, Museum of Zoology, Bangkok, Thailand (CUMZ). Additional paratypes will be housed in the Biozentrum Grindel und Zoologisches Museum, Hamburg, Germany (UHH), and the Natural History Museum, London (NHM).

Anatomical abbreviations.— fp, female pore; gm, genital markings; gmg, genital marking glands; ic, intestinal caeca; mp, male pores; pg, prostate gland; sc, spermathecae; sp, spermathecal pores; sv, seminal vesicles.

SYSTEMATICS

Metaphire Sims and Easton, 1972

Type species.— *Rhodopis javanica* Kinberg, 1867, by monotypy

Metaphire posthuma (Vaillant, 1868) (Figure 2)

Perichaeta posthuma Vaillant, 1868: 228, pl. 1, figs 1-8. Type locality: Java. Pheretima posthuma—Stephenson, 1923: 309; Gates, 1930: 321, 1931: 405, 1932:

487, 1936: 391, 1937: 363, 1939: 104, 1972: 212.

Metaphire posthuma-Sims and Easton, 1972: 217.

External characters.— Length 64-126 mm, diameter 3-6 mm. Segments 90-126. Prostomium epilobous. First dorsal pore at 12/13. Clitellum annular xiv-xvi, annular, setae present. Setae regularly arranged around each segment, retained at ventral of clitellum; 85-99 on vii, 58-67 on xx, 17-20 between male pores,

Male pores paired in xviii, minute, each in small disc, 0.30 circumference apart. Female pore single, midventral location at xiv.

Spermathecal pores 4 pairs at 5/6-8/9, minute, superficial, 0.33 circumference apart.

Genital markings paired intrasegmental at xvii, xix.

Internal characters.— Septa, 5/6-8/9 thick, 9/10 aborted. Gizzard large within ix-x. Intestine origin at xv. Intestinal caeca paired at xxvii extending forward to xxv, simple with smooth margin. Last hearts at xiii. Lymph glands present at intestinal segments posteriorly from xxvii.

Male sexual system holandric, testis sacs paired and ventral at x, xi. Seminal vesicles paired, rather small at xi, xii. Prostate glands racemose, paired in xviii, two main branches extending from xvii-xx. Prostatic ducts each a U-shaped loop.

Ovaries paired at xiii. Spermathecae four pairs in vi-ix. Ampulla small oval, duct stout. Diverticulum with short stalk, convoluted terminal section. Typhlosole present. Genital marking glands, nearly spheroidal, sessile at xvii and xix.

Diagnosis.— Spermathecal pores minute and superficial, four pairs in 5/6-8/9. Male pores minute in xviii, each in a small disc. Genital markings two rather large pairs in xvii and xix. Intestinal caeca simple in xxvii-xxv. Testis sacs paired in x and xi. Seminal vesicles paired in xi and xii. Prostate glands paired in xvii-xx, each has two main branches.

Distribution.— Pua District, Santi Suk District, Thung Chang District, Bo Kluea District

Habitat.— This species is generally found near anthropogenic habitats, for example paddy fields, ridges of paddy fields, and in wastewater saturated soil from households. These worms are very active, and twist away when touched.

Metaphire houlleti (Perrier, 1872) (Figure 3)

Perichaeta houlleti Perrier, 1872: 99, pl. 2, figs 37-39, 41-44; pl. 3, figs 45-47, 50-55, 60-62. Type locality: Calcutta.

Perichaeta campanulata Rosa, 1890: 115. Type locality: Palon.

Amyntas houlleti—Beddard, 1900: 613 (part). Pheretima houlleti—Stephenson, 1923: 304, Gates, 1958: 16, 1972: 190.

Pheretima houlleti var. tortuosa Gates, 1926a: 157. Type locality: Rangoon. Gates, 1926b: 454.

Pheretima campanulata-Gates, 1930: 307, 1931: 373.

Metaphire houlleti-Sims and Easton, 1972: 238; Shen et al., 2005: 13, fig. 2.

External characters.– Length 82-193 mm, diameter 3.3-5.4 mm. Segments 89-138. Prostomium epilobic. First dorsal pore at 11/12 or 12/13. Clitellum annular at xiv-xvi.

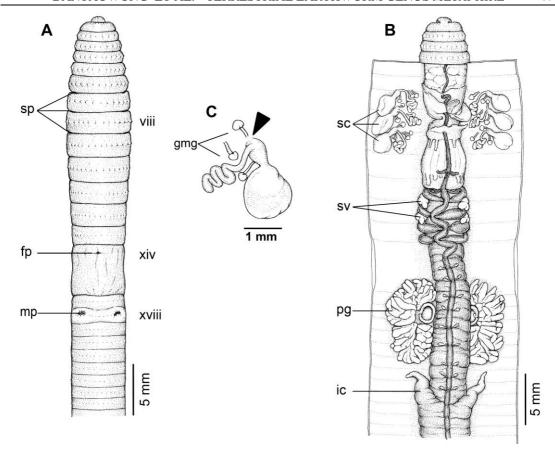


FIGURE 3. External and internal morphology of *Metaphire houlleti*. (A) External ventral view, (B) internal dorsal view and (C) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore.

annular, smooth with no setae. Setae regularly arranged around each segment, usually absent from the clitellar segment; 35-48 at vii, 48-60 at xx, 8-17 between male pores.

Male pores discharged into paired copulatory pouches opening onto the surface through tumescent lips at xviii, 0.30 circumference apart. Female pore single, midventral location at xiv.

Spermathecal pores 3 pairs at 6/7-8/9, slit-like, 0.33 circumference apart. Genital markings within spermathecal pores.

Internal characters.— Septa, 8/9-9/10 aborted, 5/6-7/8 thickened. Gizzard large within ix-x. Intestinal origin at xv. Intestinal caeca paired at xxvii extending forward to xxv, simple with smooth margin. Last hearts in xiii. Lymph glands in post-caecal intestinal segments.

Male sexual system holandric, paired testis sacs in x, xi. Seminal vesicles small, paired at xi, xii. Prostate glands racemose, paired in xviii, large expansion from xvii-xxi. Prostatic ducts fairly slim, U-shaped.

Ovaries paired at xiii. Spermathecae 3 pairs in vii, viii, ix. Ampulla oval, large, duct stout, with a swollen basal portion.

Diverticulum originates below swelling, enlarged and greatly coiled toward distal end. Typhlosole present. Genital marking glands, long stalked, with a round or slightly lobed head, near the spermathecae.

Diagnosis.— Dark dorsal pigmentation. Three pairs ventrolateral slit-like spermathecal pores at 6/7-8/9. Female pore medio-ventral at xiv. Male porophores paired at xviii, each in copulatory pouch with C-shaped opening (slit), tumescent lips. Spermathecal diverticulum enlarged and greatly coiled toward distal end. Genital marking glands stalked. Testis sacs paired in x and xi. Seminal vesicles paired in xi and xii. Prostate glands large, paired in xvii-xxi.

Distribution.— Sapan waterfall, Silapet waterfall, Tham Pha Tub Arboretum, Srinan National Park, Chieng Klang District, Sunti Suk District, Wiang Sa District, Song Khwae District. Tha Wang Pha District, Bo Kluea District.

Habitat.— This worm was found in rotten leaves in anthropogenic areas as well as in jungle.

Metaphire birmanica (Rosa, 1888) (Figure 4)

Perichaeta birmanica Rosa, 1888: 164.
Type locality: Bhamò (alto Irawaddi).
Pheretima birmanica—Stephensen, 1923: 295. Gates, 1926a: 152, 1930: 307, 1931: 372, 1932: 428, 1933: 510, 1972: 207.
Metaphire birmanica—Sims and Easton, 1972: 239.

External characters.– Length 85-210 mm, diameter 5.8-6.7 mm at clitellum. Segments 105-111. Prostomium epilobic. First dorsal pore at 12/13. Clitellum annular on xiv-xvi,

annular, smooth with no setae. Setae regularly arranged around each segment, usually absent from the clitellar segments; 65-70 at vii, 63-69 at xx, 12-17 between male pores.

Male pores discharged into paired copulatory pouches opening onto the surface through tumescent lips at xviii, 0.30 circumference apart. Female pore single, midventral location at xiv.

Spermathecal pores 3 pairs at 5/6-7/8, visible, 0.45 circumference apart.

Genital markings absent.

Internal characters.— Septa, 8/9-9/10 aborted, 5/6-7/8 thickened. Gizzard large within ix-x. Intestinal origin in xv. Intestinal caeca paired in xxvii extending forward to xxv or xxiv, manicate, dorsal most of 3-6 secondary caeca the longest. Last hearts in xiii. Lymph glands in post-caecal intestinal segments. Typhlosole present.

Male system holandric, paired testis sacs in x, xi. Small seminal vesicle paired in xi, xii. Prostate glands racemose, paired in xviii, large extending from xvii-xxi. Prostatic ducts U-shaped.

Ovaries paired at xiii. Spermathecae 3 pairs in vi, vii, viii. Ampulla large sac, duct short. Diverticulum stalk coiled, no terminal chamber.

Diagnosis.— Three pairs ventrolateral spermathecal pores at 5/6-7/8. Female pore medio-ventral at xiv. Male porophores paired at xviii, each in copulatory pouch, tumescent lips. Spermathecae irregular sacshaped, diverticulum coiled, no terminal chamber. Genital markings absent. Testis sacs paired in x and xi. Seminal vesicles paired in xi and xii. Prostate glands large, paired at xvii-xxi.

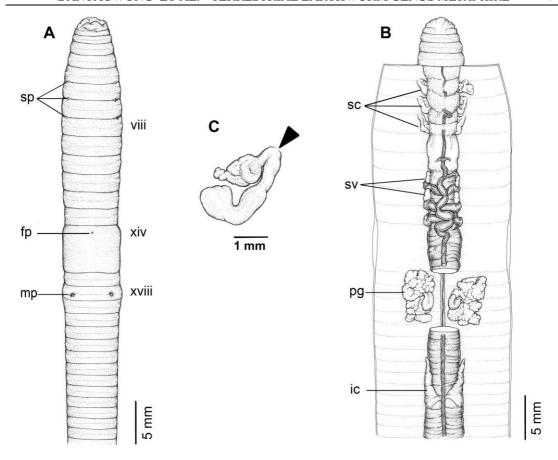


FIGURE 4. External and internal morphology of *Metaphire birmanica*. (A) External ventral view, (B) internal dorsal view and (C) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore.

Distribution.— Chaloem Phra Kiat District, Silapet waterfall.

Habitat.— This worm is generally found on the hills or mountains, and lives in the deep clay substrates.

Metaphire peguana (Rosa, 1890) (Figure 5)

Perichaeta peguana Rosa, 1890: 113. Type locality: Rangoon.

Amynthas peguana—Beddard, 1900: 628. Pheretima peguana—Stephenson, 1923: 308. Gates, 1926a: 152, 1929: 14, 1930: 318, 1931: 404, 1932: 481, 1933: 540, 1936: 444, 1937: 327, 1939: 102, 1972: 207. *Metaphire peguana*, Sims and Easton, 1972: 239.

External characters.– Length 95-220 mm, diameter 5-7 mm. Segments 77-118. Prostomium epilobous with tongue open. First dorsal pore at 12/13. Clitellum annular at xiv-xvi, annular, smooth with no setae. Setae regularly arranged around each segment, usually absent from the clitellar segments; 42-52 at vii, 54-66 at xx, 8-13 between male pores.

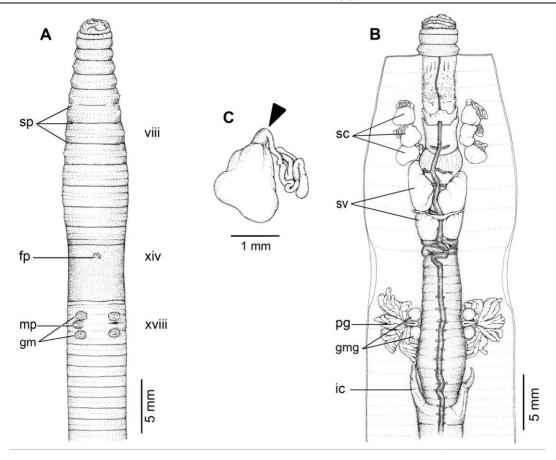


FIGURE 5. External and internal morphology of *Metaphire peguana*. (A) External ventral view, (B) internal dorsal view and (C) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore.

Male pores paired at xviii, ventro-lateral, each in circular porophore, invaginated within transversely slit-like structure, 0.30 circumference apart. Female pore single, midventral at xiv.

Spermathecal pores 3 pairs at 6/7-8/9, minute, superficial, 0.28 circumference apart.

Genital markings two pairs at 17/18 and 18/19.

Internal characters.— Septa, 8/9-10/11 aborted; 5/6-7/8, 11/12-12/13 thickened. Gizzard large within ix-x. Intestinal origin

in xv. Intestinal caeca paired at xxvii extending forward to xxiii, simple with smooth margin. Typhlosole present. Last hearts xiii.

Male system holandric, ventral testis sacs in x, xi. Seminal vesicles paired, large in xi, xii. Prostate glands racemose, paired in xviii, large extending from xvii-xx. Prostatic ducts short.

Ovaries paired at xiii. Spermathecae 3 pairs in vii, viii, ix. Ampulla oval, large, duct narrow. Diverticulum with slender stalk, tightly folded, leading to oval terminal chamber. Genital marking glands, nearly

spheroidal, slightly protuberant at 17/18 and 18/19.

Diagnosis.— Spermathecal pores at 6/7-8/9. Male pores within copulatory pouches at xviii. Two large genital markings paired at 17/18 and 18/19. Intestinal caeca simple. Testis sacs paired in x and xi. Seminal vesicles paired in xi and xii. Prostate glands large, paired in xvii-xx.

Distribution.— Silapet waterfall, Maejarim National Park, Tadman waterfall, Mueang Nan, Chiang Klang District, Santi Suk District, Wiang Sa District, Song Khwae District, Tha Wang Pha District, Bo Kluea District, Chaloem Phra Kiat District, Thung Chang District.

Habitat.— This worm was found widely throughout Nan Province in dipterocarp forest, deciduous forest, and anthropogenic areas, even in wastewater saturated soil from households.

Metaphire anomala (Michaelsen, 1907) (Figure 6)

Pheretima anomala Michaelsen, 1907: 167, fig. 14. Type locality: Botanical Gardens, Sibpur, Calcutta. Stephenson, 1923: 294, Gates, 1926a: 151, 161, 1930: 307, 1931: 372, 1932: 387, 1933: 496, 1936: 396, 1939: 82, 1972: 166.

Metaphire anomala–Sims and Easton, 1972: 239.

External characters.— Length 134-154 mm, diameter 4.6-6.6 mm at clitellum. Segments 116-126. Prostomium epilobic. First dorsal pore at 12/13. Clitellum annular at xiv-xvi, annular, smooth with no setae. Setae regularly arranged around each segment, usually present on ventral side of

xvi; 84-96 at vii, 58-67 on at xx, 12-20 between male pores.

Male pores within paired copulatory pouches opening onto the surface through tumescent lips at xx, 0.33 circumference apart. Female pore single, midventral at xiv.

Spermathecal pores 3 pairs at 5/6-7/8, slit-like; 0.30 circumference apart.

Genital markings present at 17-19 (Fig. 6A), or 17-23 (Fig. 6E), or absent (Fig. 6D).

Internal characters.— Septa, 8/9-9/10 aborted, 5/6-7/8 thickened. Gizzard large within ix-x. Intestinal origin in xv. Intestinal caeca paired in xxvii extending forward to xxiii, simple. Typhlosole present. Last hearts in xiii.

Male system holandric, paired testis sacs in x, xi. Seminal vesicles paired, large in xi, xii. Prostate glands racemose, paired in xx, large extending from xvii-xxii. Prostatic ducts long, hairpin shaped.

Ovaries paired in xiii. Spermathecae 3 pairs in vi, vii, viii. Ampulla slender oval, duct shorter than ampulla. Diverticulum with slender multiply kinked stalk, coiled terminal chamber. Genital marking glands, sessile corresponding to external genital markings.

Variation.– Individuals of a hermaphroditic morph (H morph after Gates (1955)) having a full hermaphroditic complement of reproductive usually organs have spermathecal pores at 5/6-7/8, and male pores at xx, with three pairs genital markings on xvii. xviii. xix. spermathecae as well as prostate glands (seven specimens) (Fig. 6A-C). R morphs are distinguished by absence of male terminalia as well as of all genital marking glands of xvii-xix. Spermathecal pores are occasionally found in intersegments 5/6-7/8, and with them usually spermathecae (nine

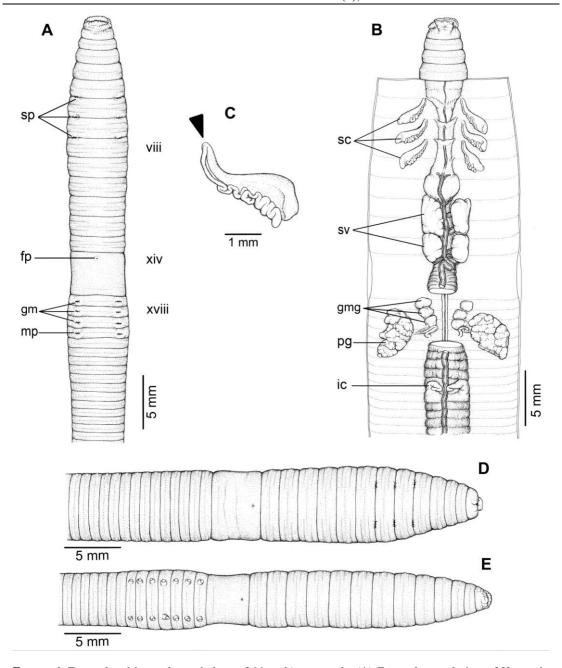


FIGURE 6. External and internal morphology of *Metaphire anomala*. (A) External ventral view of H morph, (B) internal dorsal view of H morph and (C) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore, (D) External ventral view of R morph, (E) External ventral view of A morph.

specimens) (Fig. 6D). Reproduction in this morph must often be uniparental (Gates, 1955). An A morph (Gates, 1955) is distinguished from the H by the presence of three pairs of genital markings and GM glands behind xx and by absence of six spermathecae (Fig. 6E). Male terminalia are present and apparently normal (prostates large) but seminal vesicles usually are small (six specimens). Reproduction in this morph must be not only uniparental but also parthenogenetic (Gates, 1955).

Diagnosis.— Three pairs spermathecal pores at 5/6-7/8, slit-like. Female pore medioventral at xiv. Secondary male pores paired at xx, copulatory pouch present. Spermathecal diverticulum with coiled terminal chamber. Genital markings at xvii-xix. Testis sacs paired in x and xi. Seminal vesicles paired in xi and xii. Prostate glands large, paired in xvii-xxii.

Distribution.— Tham Pha Tub Arboretum, Tadman waterfall, Chaloem Phra Kiat District, Santi Suk District, Wiang Sa District, Song Khwae District.

Habitat. This species can be found in both natural forest and anthropogenic areas.

Metaphire grandipenes Bantaowong and Panha, n. sp. (Figures 1 and 7)

External characters.— Length of head fragment 40 mm, diameter 4.5 mm at clitellum. Number of segments 71. Prostomium epilobic. First dorsal pore at 12/13. Clitellum annular at xiv-xvi, with no setae. Setae regularly arranged around each segment, except in x-xiii, and xvii- xxvii where they are ventrally scattered; 81 at vii, 86 on at xx, 42 between male pores.

Male pores paired, on large protruding alate porophores on segment xviii, 0.5 circumference apart, distance between male pores 5 mm. Female pore single, midventral xiv.

Spermathecal pores 3 pairs at 5/6-7/8, slit-like, 0.4 circumference apart, distance between spermathecal pores 4.5 mm.

Genital marking very large, oval suckerlike, one pair on intersegmental boundary 19/20.

Internal characters.— Septa, 8/9-9/10 aborted, 5/6-7/8 thickened; 10/11-12/13 membranous. Gizzard large within ix-x. Intestinal origin in xv, no lymph glands observed. Typhlosole small from xxvii. Intestinal caeca paired at xxvii extending forward to xxiii, simple. Hearts esophageal in x-xiii.

Male system holandric, paired testis sacs in x, xi. Seminal vesicles paired, large in xi, xii. Pseudovesicles in xiii. Prostate glands racemose, paired xviii, large extending from xvii-xx. Prostatic ducts U-shaped.

Ovaries paired in xiii. Spermathecae 3 pairs in vi, vii, viii. Ampulla ovate, duct shorter than ampulla. Diverticulum with slender straight stalk, terminal chamber with five zig-zags pattern. Genital marking glands in xix-xx very large, sessile corresponding to each external genital marking.

Variation.— The holotype head fragment measures 40 mm body length with 71 segments; the two paratypes range in size from 35 mm body length with 73 segments (incomplete body), and 100 mm body length with 195 segments.

Type locality.— Community forest of Ban Bun Rueng, Lainan Subdistrict, Wiang Sa District, Nan Province, Thailand, 18° 34'

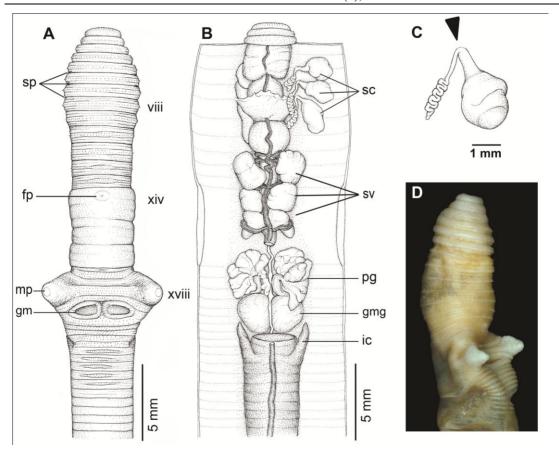


FIGURE 7. External and internal morphology of holotype (CUMZ 3213) of *Metaphire grandipenes* n. sp. (**A**) External view, (**B**) internal dorsal view and (**C**) spermathecae, black arrow indicates the connection of the spermathecae and spermathecal pore, (**D**) male porophores protruded as two penes.

0.8" N, 100° 44' 7.3" E, 275 meters elevation (10^{th} December 2008).

Etymology.— This species was named after the character of large male pores at xviii extremely protruded as two penes.

Type material.— The holotype (CUMZ 3213), and 2 paratypes (CUMZ 3214) are deposited in Chulalongkorn University, Museum of Zoology. Each two paratypes are deposited in UHH and NHM.

Habitat.– Found in the top soil at about 10 cm depth, the soil surface was covered with

leaf litter in a diperocarp forest. Most surrounding areas have been modified to agricultural fields.

Diagnosis.— Three pairs spermathecal pores at 5/6-7/8, slit-like. Female pore mid-ventral on xiv. Male porophores paired, extremely large, alate on xviii. Spermathecae large ovate ampulla, diverticulum with straight stalk and long zig-zag terminal chamber. Genital markings extremely large, two sucker rings at 19/20, each with sessile genital marking glands. Testis sacs paired in x and xi. Seminal vesicles paired in xi, xii. Prostate glands paired in xvii-xx.

Characters	M. posthuma	M. houlleti	M. birmanica	M. peguana	M. anomala	M. grandipenes n. sp.
Body length (mm)	64-126	82-193	85-210	95-220	134-154	>40-100
Clitellum width (mm)	3-6	3.3-5.4	5.8-6.7	5-7	4.6-6.6	4.2-5.1
Number of segments	90-126	89-138	105-111	77-118	116-126	>71-195
Setae between male pores	17-20	8-17	12-17	8-13	12-20	37-42
First dorsal pore	12/13	11/12, 12/13	12/13	12/13	12/13	12/13
Spermathecal pores	5/6-8/9	6/7-8/9	5/6-7/8	6/7-8/9	5/6-7/8	5/6-7/8
Male pores	xviii	xviii	xviii	xviii	XX	xviii
Genital markings	xvii, xix	absent	absent	17/18, 18/19	xvii-xix (H morph) xvii-xxiii (A morph)	19/20
Prostate glands	xvii-xx	xvii-xxi	xvii-xxi	xvii-xx	xvii-xxii	xvii-xx
Genital marking glands	sessile, xvii, xix	stalked, vii, viii, ix	absent	sessile, 17/19,	xvii-xix (H morph),	sessile, 19/20
				18/19	xvii-xxiii (A morph)	
Intestinal caeca	simple, xxvii-	simple,	manicate,	simple,	simple,	simple,

xxvii-xxv

xxvii-xxv

TABLE 1. The characteristics of the six *Metaphire* species from Nan Province, Thailand.

Remarks.- Metaphire grandipenes n. sp. looks very close to Metaphire abdita (Gates, 1935) from Suifu, Szechuan, China in many characteristics for examples the three pairs of spermathecae located in vi, vii, viii corresponding with spermathecal pore locations 5/6-7/8. However at after investigating the type specimens of M. abdita of U.S.N.M 20094, it is clear that the two species are different by the following comparisons. Male pores at ventral end of tubular penes located in eversible deep parietal invaginations with elongately slitlike apertures of M. abdita look similar to the new species, however in the case of M. grandipenes n. sp. the porophores do not have tubular penes and appear not to have copulatory associated an pouch. Furthermore the ventral expansion of xviixx into a triangular alate protrusion does not appear in M. abdita. The prominent large pair of oval sucker-like genital markings at 19/20 is different, because M. abdita has

XXV

small paired presetal markings on xviii and xix, and the GM glands are not so prominent. The absence of septa 8/9-9/10 in this new species is in contrast to their muscular presence in *M. abdita*. The spermathecal ampulla shape of the two species look similar, but the diverticulum stalk is straight in the new species, and convoluted in *M. abdita*. The diverticulum chamber is a small oval in *M. abdita* but it is a zig-zag slender shape in the new species.

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DISCUSSION

Metaphire grandipenes n. sp. was discovered in the remote valley of the Nan River, surrounded by mountains covered with deciduous forest. The type locality is modified by farmers to be agricultural but they keep the forest remnants as community forest for biodiversity conservation. This is showing that there are some good areas remaining for conservation purposes and/or any organic farming which is increasingly

important in the world. *Metaphire grandipenes* n. sp. is probably an isolated endemic occurring in the Nan River valley, for which the habitat characteristics are quite different from those of the other five species mentioned here. It was found coexisting with the common *M. peguana* but the latter was not so abundant as in other areas (Table 1).

The other five Metaphire species have been previously recorded in Burma (Gates, 1972) but here we report them in the forest remnants in Nan on the opposite side of Thailand from the border of Burma. Metaphire birmanica proved to be a new record Thailand. The morphological characteristics of the five species are mostly the same as described elsewhere, but there are also some elements of differentiation. For example M. posthuma of Nan has four pairs of spermathecae in viix which corresponds with four pairs of spermathecal pores at 5/6-8/9. Other characters from the Nan specimens agree with M. posthuma as described before but the additional pair of spermathecae in Nan specimens suggest they should be classified as an unknown species within the M. posthuma group.

Chantaravisoot (2007)reported unidentified taxa keying to the M. houlleti complex, collected from many areas in Thailand. This probably indicates a level of cryptic or semi-cryptic diversity and should be tested at molecular levels. The common M. peguana is less variable, generally agreeing with previously described forms in other areas. The morphological variation within the M. peguana group is relatively small, with 3 unidentified taxa recorded in Chantaravisoot (2007). Metaphire peguana is a cosmopolitan species distributed all over Thailand and nearby countries such as Malaysia, Singapore, Philippines and Laos

(Gates, 1939, 1972; Shen and Yeo, 2005; Kosavititkul, 2005; Chantaravisoot, 2007; Somniyam, 2009; Panha, unpublished data). *Metaphire anomala* shows a high degree of variation as seen in the above description. This may be the initial step of the current time and space of new forms for *M. anomala*, but much of the variation appears to be associated with parthenogenesis.

Molecular analyses combined with morphological data and geological history analysis could show the evolutionary relationships among the species reported here, the history of introductions of common species, and the evolution in general of this area of endemism.

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LITERATURE CITED

Beddard, F.E. 1900. On a new species of earthworm from India belonging to the genus *Amynthas*. Proceedings of the Zoological Society of London, 1900: 998–1002.

Blakemore, R.J. 2006. Chapters In: A Series of Searchable Texts on Earthworm Biodiversity, Ecology and Systematics from Various Regions of the World - Supplemental. Eds. N. Kaneko and M.T. Ito. COE Soil Ecology Research Group, Yokohama National University, Tokiwadai, Yokohama Japan. Available from http://bio-

- eco.eis.ynu.ac.jp/ eng/database/earthworm/ (accessed 21st December 2010)
- Chantaravisoot, N. 2007. Species Diversity of Terrestrial Earthworms Family Megascolecidae in Thailand. Senior Project Report, Department of Biology, Faculty of Science, Chulalongkorn University, 36 pp. (with English Abstract).
- Gates, G.E. 1926a. Notes on earthworms from various places in the province of Burma, with descriptions of two new species. Records of the Indian Museum, 28: 141-170.
- Gates, G.E. 1926b. Notes on Rangoon earthworms. The peregrine species. Annual Magazine Natural History Series 9. 17: 439-473.
- Gates, G.E. 1929. A summary of the earthworm fauna of Burma with descriptions of fourteen new species. Proceedings of the United States National Museum. 75: 1-41.
- Gates, G.E. 1930. The earthworms of Burma I. Records of the Indian Museum, 32: 257–356.
- Gates, G. E. 1931. The earthworms of Burma II. Records of the Indian Museum, 33: 327–442.
- Gates, G.E. 1932. The earthworms of Burma III. Records of the Indian Museum, 34: 357–549.
- Gates, G.E. 1933. The earthworms of Burma IV. Records of the Indian Museum, 35: 413–606.
- Gates, G.E. 1936. The earthworms of Burma V. Records of the Indian Museum, 38: 377-468.
- Gates, G.E. 1937. The genus *Pheretima* in north America. Bulletin of the Museum of Comparative Zoology, 80: 339–373.
- Gates, G.E. 1939. Thai earthworms. Journal Thailand Research Society. 12: 65-114.
- Gates, G.E. 1955. Reproductive organ polymorphism in earthworms of the oriental Megascolecine genus *Pheretima* Kinberg, 1867. Evolution 10: 213-227.
- Gates, G.E. 1958. On some species of the Oriental earthworm genus *Pheretima* Kinberg, 1867, with a key to species reported from the Americas. American Museum Novitates, 1888: 1-33.
- Gates, G.E. 1972. Burmese earthworms, an introduction to the systematics and biology of megadrile oligochaetes with special reference to the Southeast Asia. Transactions of the American Plilosophical Society, 62: 1–326.
- Kosavititkul, P. 2005. Species Diversity of Terrestrial Earthworms in Khao Yai National Park. Ph.D Thesis in Environmental Biology, School of Biology, Institute of Science, Suranaree University of Technology, 193 pp.

- Michaelsen, W. 1907. Neue Oligichaeten von Vorder-Indien, Ceylon, Birma und den Andaman-Inseln. Mitteilungen aus dem Naturhistorischen Museum, 24: 143-188.
- Perrier, E. 1872. Recherches pour server a l'histoire des Lombricins terrestres. Nouvelles Archives du Museum d'histoire Naturelle, 8: 5-198.
- Rosa, D. 1888. Viaggio di Leonardo Fea in Birmanica e regioni vicine, V-Preichetidi. Annali del Museo Civico di Storia Naturale, Giacomo Doria, 6: 155-167.
- Rosa, D. 1890. Viaggio di Leonardo Fea in Birmanica e regioni vicine, XXVI. Preichetidi. Annali del Museo Civico di Storia Naturale, Giacomo Doria, 10: 107-122.
- Shen, H.-P., Tsai, S.-C. and Tsai, C.-F. 2005. Occurrence of the Earthworms *Pontodrilus litoralis* (Grube, 1855), *Metaphire houlleti* (Perrier, 1872), and *Eiseniella tetraedra* (Savigny, 1826) from Taiwan. Taiwania, 50: 11-21.
- Shen, H.-P. and Yeo, D.C.J. (2005) Terrestrial earthworms (Oligochaeta) from Singapore. The Raffles Bulletin of Zoology, 53, 13-35.
- Sims, R.W. and Easton E.G. 1972. A numerical revision of the earthworm genus *Pheretima* (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.
- Somniyam, P. and Suwanwaree, P. 2009. The Diversity and Distribution of Terrestrial Earthworms in Sakaerat Environmental Research Station and Adjacent Arae, Nakorn Ratchasima, Thailand. World Applied Sciences Journal, 6: 221-226.
- Stephenson, J. 1923. Oligochaeta. In: Shipley, A.E. and Scott, H. (eds). The Fauna of British India including Ceylon and Burma. Taylor and Francis Inc., London, 518 pp.
- Tsai, C.-F., Tsai, S.-C. and 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.
- Vaillant, L. 1868. Note sur l'anatomie de deux especes du genre *Perichaeta* et essai de classification des Annelides Lombricines. Annales des Sciences Naturelles, 10: 225-256.