

# Taxonomic Review and Phylogeny of Marine Long-legged Flies (Diptera: Dolichopodidae) in Peninsular Thailand

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Thesis Title	Taxonomic Review and Phylogeny of Marine Long-legged Flies
	(Diptera: Dolichopodidae) in Peninsular Thailand
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Major Program	Biology
Academic Year	2016

#### ABSTRACT

A taxonomic review of marine long-legged flies (Dolichopodidae) in peninsular Thailand was investigated. The flies were randomly sampled and collected from rocky shores, sandy beaches and mangroves in eight provinces (namely Pattani, Songkhla, Nakhon Si Thammarat, Surat Thani, Chumphon, Satun, Krabi, and including Phang Nga) along the Thai seacoasts from November 2014 until May 2015. Four fly collection methods such as Malaise trap, yellow pan trap, sweep net and hand collecting method were used. In addition, external morphology features and Next Generation Sequencing techniques, NGS barcodes were employed in order to identify species. Totally, 60 species belonging to 19 genera, 7 subfamilies and one unplaced genus (genus *incertae sedis*) of marine long-legged flies were recognized from Thai seacoasts. All of these, consisting of 22 species from both sides of Peninsular Thailand, 23 species from the Andaman Sea, and 15 species from the Gulf of Thailand.

Seventeen new species have been described from this study: Asyndetus sp.1, Cymatopus mayakunae sp. nov., Diaphorus sp.1, Hercostomus propermeieri sp. nov., Ngirhaphium chutamasae sp. nov., N. meieri sp. nov., Ornamenta siamese sp. nov., gen. nov., Paraclius sp.1, Phoomyia talumpuk sp. nov., Thinophilus boonrotpongi sp. nov., T. langkawensis sp. nov., T. minutus sp. nov., T. parvulus sp. nov., T. parvulus sp. nov., T. parmatoides sp. nov., T. spinatus sp. nov., T. spinatoides sp. nov. and including T. variabilis sp. nov. Furthermore, traditional identification and molecular phylogenetic analysis confirmed the monophyly of marine long-legged flies with reference to Ngirhaphium Evenhuis and Grootaert and Hercostomus Loew. The results clearly showed that the recent Ngirhaphium Evenhuis and Grootaert was divided into five major clades consisting of N. caeruleum, N. chutamasae sp. nov., N. meieri sp

*N. murphyi*, and *N. sivasothii*, and the genus *Hercostomus* Loew was also separated into five distinct clades, namely, *H. lanceolatus*, *H. plumatus*, *H. obtusus*, *H. brevicornis* and *H. brevidigitalis*.

Moreover, the results of haplotype network analysis and distribution pattern of mangrove *Hercostomus lanceolatus* Zhang, Yang and Grootaert from Thailand and Singapore population clearly indicated that haplotype pattern of Surat Thani assemblage was the parental population or the origin of *H. lanceolatus* in this region. Geographical distances and human anthropogenic threats were suggested to play an important role on genetic variation of marine long-legged flies.

Key words; Dolichopodidae, peninsular Thailand, NGS barcoding

ชื่อวิทยานิพนธ์	อนุกรมวิชานและวงศ์วานวิวัฒนาการของแมลงวันขายาวในคาบสมุทร
	ไทย
ผู้เขียน	นายอับคุลเลาะ ซาเมาะ
สาขาวิชา	ชีววิทยา
ปีการศึกษา	2559

### บทคัดย่อ

การทบทวนทางอนุกรมวิธานของแมลงวันขายาวทางทะเล (วงศ์แมลงวันขายาว) ในคาบสมุทรไทย โดยการศึกษาตัวอย่างแมลงวันขายาวทางทะเลที่สุ่มเก็บและรวบรวมจากหาดหิน หาดทรายและป่าชายเลนจากชายฝั่งทะเลของอ่าวไทยบริเวณจังหวัดปัตตานี สุราษฎร์ธานี สงขลา นกรศรีธรรมราช ชุมพร และชายฝั่งทะเลอันดามันบริเวณจังหวัดสดูล กระบี่ และพังงา ระหว่าง เดือนพฤสจิกายน พ.ศ. 2556 ถึงเดือนพฤษภาคม พ.ศ. 2557 อาศัยวิธีการเก็บตัวอย่าง 4 วิธี คือ กับดัก มุ้งแบบ Malaise ถาดดักจับแมลงสีเหลือง สวิงจับแมลง และสุ่มเก็บด้วยมือ โดยอาศัยหลักการจัด จำแนกทางอนุกรมวิธานสองรูปแบบ ได้แก่ สัณฐานวิทยาภายนอกและรหัสแท่งดีเอ็นเอ (DNA barcode) ด้วยเทคนิค Next Generation Sequencing (NGS) สามารถระบุชนิดแมลงวันขายาวทาง ทะเลจากคาบสมุทรไทยได้ทั้งสิ้น 60 ชนิด 19 สกุล 7 วงศ์ย่อย และ 1 กลุ่มสกุลที่ไม่สามารถจัดเข้า กลุ่มใด ๆ (genus *incertae sedis*) แมลงวันขายาวทางทะเลที่พบกระจายได้ทั้งสองฝั่งทะเลของ คาบสมุทรไทยมีจำนวน 22 ชนิด พบเฉพาะทางชายฝั่งอันดามัน 23 ชนิดและพบเฉพาะทางชายฝั่ง

ผลศึกษาพบแมลงวันขายาวทางทะเลชนิดใหม่ของโลก 17 ชนิด ได้แก่ Asyndetus sp.1, Cymatopus mayakunae sp. nov., Diaphorus sp.1, Hercostomus propermeieri sp. nov., Ngirhaphium chutamasae sp. nov., N. meieri sp. nov., Ornamenta siamese sp. nov., gen. nov., Paraclius sp.1, Phoomyia talumpuk sp. nov., Thinophilus boonrotpongi sp. nov., T. langkawensis sp. nov., T. minutus sp. nov., T. parvulus sp. nov., T. parmatoides sp. nov., T. spinatus sp. nov., T. spinatoides sp. nov. และ T. variabilis sp. nov. การจำแนกและวิเคราะห์ความสัมพันธ์ทาง วิวัฒนาการของแมลงวันขายาวทางทะเลบางสกุล ได้แก่ สกุล Ngirhaphium Evenhuis & Grootaert และสกุล Hercostomus Loew พบว่าจำนวนชนิดที่ได้จากการจัดจำแนกแมลงวันขายาวทั้งสอง วิธีการมีความสอดคล้องกันและเป็นสายวิวัฒนาการเดี่ยว อีกทั้งยังบ่งชี้ให้เห็นว่าแมลงวันขายาวทาง ทะเลสกุล Ngirhaphium Evenhuis & Grootaert สามารถจัดจำแนกออกได้เป็น 5 กลุ่มได้แก่ N. caeruleum, N. chutamasae sp. nov., N. meieri sp. nov., N. murphyi และ N. sivasothii สำหรับ สกุล Hercostomus Loew สามารถจัดจำแนกออกได้เป็น 5 กลุ่มเช่นกัน ได้แก่ H. lanceolatus, H. plumatus, H. obtusus, H. brevicornis และ H. brevidigitalis

ผลจากการการวิเคราะห์รูปแบบที่แตกต่างกันของความแปรผันทางพันธุกรรม (haplotype network) และรูปแบบการแพร่กระจายของประชากรแมลงวันขายาวทางทะเลชนิด *Hercostomus lanceolatus Z*hang, Yang และ Grootaert จากประเทศไทยและประเทศสิงคโปร์ ซี้ชัด ได้ว่าประชากรแมลงวันขายาวทางทะเลชนิดนี้จากจังหวัดสุราษฎร์ธานีเป็นประชากรเริ่มต้นของ แมลงวันขายาวกลุ่มนี้ในภูมิภาค ทั้งนี้ระยะห่างของแหล่งอาศัยและกิจกรรมของมนุษย์อาจจะเป็น ปัจจัยหลักที่ส่งผลกระทบต่อความผันแปรและความหลากหลายทางพันธุกรรม

กำสืบค้น; Dolichopodidae, peninsular Thailand และ NGS barcoding

### ACKNOWLEDGEMENTS

Foremost, I would like to express my sincere gratitude to my advisor, Associate Professor Dr. Chutamas Satasook, for the continuous support of my Ph.D. study and research, for her patient guidance, encouragement, advice, motivation, enthusiasm, and immense knowledge over the past four years. Besides my advisor, I would like to show my greatest appreciation to my co-advisor, Dr. Singtoe Boonrotpong, who gives me constructive comments. Without his guidance and persistent help this thesis would not have been possible. My deepest appreciation also goes to my co-advisor, Dr. Patrick Grootaert, Head of Entomology Department in Royal Belgian Institute of Natural Sciences (RBINS), best friend, colleague and excellent teacher and dolichopodid fly trainer. Big thanks for advices and moral supports.

My thankfulness goes to Professor Ding Yang, China Agricultural University, for providing me his World Catalog of Dolichopodidae book and beneficial literatures of Dolichopodidae. My heartfelt appreciation also goes to Professor Rudolf Meier, Head of Molecular Evolution Laboratory, Wendy Wang, Sujatha Narayanan Kutty, David Tan, Darren Yeo, Amrita Srivathsan, Kai Qing of the Evolutionary Biology Laboratory, Department of Biological Sciences, National University of Singapore (NUS), for enthusiastic help in Molecular work.

I owe a very important debt to Simon Jack and Dr. Danjuma Solomon, for English editing and proofreading, valuable comments and moral supports. I want to thank my friends at Biology Department, especially my fellow labmates in Entomology Research Unit and Cephalopod Research Unit for their kindly help in the field and laboratory and continuously encouragements.

I would like to express my gratitude to National Research University (NRU) Scholarship, Prince of Songkla Graduate School Funding, Oversea Research Grant of the Faculty of Science, and Department of Biology for their financial support.

Of course, my most influential teachers are my lovely parents, and I owe an immense debt of gratitude to them for their support (of all kinds) over the years. Thank you for helping me get through my many paroxysms of self-doubt.

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### LIST OF PAPERS AND MANUSCRIPTS

Works in this thesis are organized as following papers and manuscripts. These papers and manuscripts were referred to in the text as Paper 1, Paper 2, Manuscript 1, and Manuscript 2, respectively.

### **PAPERS**:

- Samoh, A., Boonrotpong, S. and Grootaert, P. 2015. *Ngirhaphium* Evenhuis and Grootaert from southern Thailand (Diptera: Dolichopodidae) with a description of a new species. *Zootaxa*, 3946 (1): 125-132.
- Samoh, A., Satasook, C. and Grootaert, P. 2017. Eight new species of marine dolichopodid flies of *Thinophilus* Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand. *European Journal of Taxonomy*, 329: 1-40.

#### **MANUSCRIPTS :**

- Samoh, A., Satasook, C., Boonrotpong, S., and Grootaert, P. An annotated checklist of the marine long-legged flies (Diptera: Dolichopodidae) from Thai Peninsula.
- Samoh, A., Satasook, C., Boonrotpong, S., and Grootaert, P. New data on the marine genera *Cymatopus* Kertèsz and *Thambemyia* Oldroyd (Insecta, Diptera, Dolichopodidae) from rocky shores in southern Thailand with the description of a new species.

## LETTER OF ACCEPTANCE

**Paper I :** Samoh, A., Boonrotpong, S. and Grootaert, P. 2015. *Ngirhaphium* Evenhuis and Grootaert from southern Thailand (Diptera: Dolichopodidae) with a description of a new species. *Zootaxa*, 3946 (1): 125-132.

Published by : Zootaxa, Magnolia Press

7/12/2017 Gmail - FW: Accepted ms on Dolichopodidae (Diptera: Empidoidea) from SAMOH, BOONROTPONG & GROOTAERT



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---Original message----From: Bradley Sinclair <Bradley.Sinclair@inspection.gc.ca> Sent: Wed 11-02-2015 15:19 Subject: Accepted ms on Dolichopodidae (Diptera: Empidoidea) from SAMOH, BOONROTPONG & GROOTAERT Attachment: Samoh et al\_Ngirhaphium Thailand\_final.doc To: zed@mapress.com; CC: Patrick.Grootaert@naturalsciences.be; Patrick Grootaert sprootaert@yahoo.co.uk>; Dear Zhi-Qiang: Please find attached an accepted and formatted manuscript entitled: *Ngirhaphium* Evenhuis & Grootaert from southern Thailand (Diptera: Dolichopodidae) with the description of a new species by ABDULLOH SAMOH, SINGTOE BOONROTPONG & PATRICK GROOTAERT Corresponding author: Patrick Grootaert <<u>Patrick.Grootaert@naturalsciences.be</u>>

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7/12/2017

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Figures: 13 References: 5 New species: Diptera: 1

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cheers, Brad

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**Paper II :** Samoh, A., Satasook, C. and Grootaert, P. 2017. Eight new species of marine dolichopodid flies of *Thinophilus* Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand. *European Journal of Taxonomy*, 329: 1-40.

Published by : European Journal of Taxonomy

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ABDULLAH SAMOH <flywizme@gmail.com>

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FYI

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Subject: Your Submission EJT-16-86R2 - Eight new species of marine Thinophilus Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand To: Patrick Grootaert species of marine Thinophilus Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand EUROPEAN JOURNAL OF TAXONOMY Dear Dr Patrick Grootaert, Following the recommendation of the topical editor, I have the pleasure to accept your paper for publication in the EUROPEAN JOURNAL OF TAXONOMY. The editorial office will contact you in due course. May I ask you to fully collaborate with the production process leading up to publication of your manuscript, by providing figures of high technical quality (resolution of at least 300 dpi for photographs and of 1200 dpi for line drawings, in .jpeg or .tiff format), and by returning your proofs within the requested period. Failure to do so might prevent your manuscript from being published, as both scientific and technical quality of papers published in the EUROPEAN JOURNAL OF TAXONOMY must be of high standard.

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#### **SUMMARY OF CONTENTS**

### 1. GENERAL INTRODUCTION AND LITERATURE REVIEW

#### **1.1) General Introduction**

The Dolichopodidae or long-legged flies can be easily recognized by their fivemajor external morphological features, namely, mostly metallic greenish blue to greenish bronze colour, a range in body size from 1-9 mm in length and relatively slender (Robinson and Vockeroth, 1981), elongated legs, reduced wing venation into a single big cell (dm-cm cell), and also display some distinct male genitalia (180° movable) (d'Assis Fonseca, 1978). In terms of their role in the ecosystem, both adult and larvae of longlegged flies are assumed to be predacious flies (Laurence, 1951; d'Assis Fonseca, 1978). Soft-body larvae of mosquitoes, biting and nonbiting midges, wounded arthropods and amphibian embryos, and early instar of caterpillars (Ulrich, 2005) are generally predated by this carnivorous fly. Furthermore, they are highly sensitive to environmental alteration and prove useful as a bioindicator and for long-term environmental monitoring (Pollet, 2009; Grootaert and Meuffels, 2004). It is also known as a greatly diversified dipteran group in brachyceran series. Globally, approximately 6,500-7,600 species, belong to 240 genera have currently been identified (Grichanov, 1999; Yang et al., 2006; Wang et al., 2007a; Pape et al., 2009). But there is little information or have had little attention paid to them, especially marine species. Nearly a hundred species have been recorded from this region. About 85 species have been assumed to occur from Singapore Island (Grootaert 2016, personal communication) and only fifteen species have been reported from Thailand (Grootaert and Meuffels, 2001).

The superfamily Empidoidea could be classified into two families (Empididae and Dolichopodidae) based on morphology features (Yang and Yang, 2004; Yang et al., 2006). The monophyletic lineage of Empididae is evident (Cumming et al., 1995; Collins and Wiegmann, 2002) and recognised by these external morphology features: head rather small, narrower than thorax in dorsal view and more or less rounded in lateral view; thorax weakly to strongly convex upward; eyes with angular inner incision near antennae; hypandrial lobe very large and broad, and isolated from epandrial lobe (Cumming et al., 1995; Sinclair and Cumming 2006). For Dolichopodidae is classified by the following

features: head large hemispherical, distinctly higher than long, nearly as wide as or wider than thorax; thorax not distinctly convex dorsally; eyes without inner incision near antennae; costal vein with break near humeral cross-vein; subcostal vein apically fused with vein R1, not reaching costal vein; 2nd basal cell and discal cell fused; epandrium with genital foramen; hypandrium basally fused with epandrium (Wang et al., 2007a). Although, the traditional classification based on morphology could be divided Empidoidea into two groups, in fact, they are literally missing in the context of systematic concept (Germann et al., 2011) and highly controversial (Sinclair et al., 2008). They further classified the Empidoidea into five families as follows; Empididae, Hybotidae, Atelestidae (including Nemedininae), Brachystomatidae (comprising Brachystomatinae, Ceratomerinae and Trichopezinae), and Dolichopodidae sensu lato (Sinclair et al., 2008).

According to previous investigators, several ideas of classification into subfamily have been projected by previous dolichopodid fly taxonomists, shown as follows: In 1917, 1918, and 1922, very precious taxonomic information of the family dolichopodidae provided by Becker. They are ascertained from eleven subfamilies including Aphrosylinae, Diaphorinae, Dolichopodinae, Hydrophorinae, Medeterinae, Neurigoninae, Rhaphiinae, Plagioneurinae, Stolidosomatinae, Sciapodinae (as Chrysosomatinae) and Sympycninae (as Campsicneminae). Then, Robinson (1970) gave a revised subfamily classification on Becker's with fourteen subfamilies. He recognised Peloropeodinae and erected a new subfamily Enliniinae, and synonymized Aphrosylinae with Hydrophorinae. Recently, two additional subfamilies have also been erected, i.e. Babindellinae by Bickel (1987) and Achalcinae by Grootaert and Meuffels (1997). Whereas one of the most updated that was classified by Yang et al. (2006), they divided the family Dolichopodidae into seventeen subfamilies, including two new subfamilies, composed: Achalcinae, Antyxinae, Babindellinae, Diaphorinae, Dolichopodinae, Enliniinae, Hydrophorinae, Kowmunginae, Medeterinae, Neurigoninae, Plagioneurinae, Peloropeodinae, Rhaphiinae, Sciapodinae, Stolidosomatinae, Sympycninae, and Xanthochlorinae.

Up to this time, a distinct lack of basic knowledge in various aspects of the Dolichopodidae in Thailand is evident. Few reports have been published from this country. For example, three new species of genus *Nanothinophilus* Grootaert and Meuffels, namely, *N. armatus*, *N. dolichurus*, and *N. pauperculus* (Grootaert and Meuffels, 1998) from the Andaman seacoast were discovered. A year later, one genus

such *Terpsimyia* Becker was rediscovered from Gulf of Thailand (South China Sea). A brief data, including species composition, description, and regional generic key have been provided by Grootaert and Meuffels (2001). Fifteen species in seven genera were found, of these, seven species were claimed as species new to science (Grootaert and Meuffels, 2001a). However, the species number previously surveyed is too low and all marine habitats have not been surveyed in Southern Thailand which is considered to be the richest habitat in this country. In addition, the standard methods of flying insects sampling such as Malaise trap and yellow pan trap were not employed. Moreover, in terms of species recognition, only the traditional method of species identification was used and lots of specimens remain unclear. Due to these facts, many gaps of knowledge are open and need elucidating and further understanding.

Interestingly, after a year of fly surveying from several kinds of marine habitats (including mangroves, rocky shores, sandy beaches, mudflats) throughout peninsular Thailand, the preliminary results revealed that the species were far more numerous and plenty of specimens were pending description as new species and genus. Furthermore, the Thai marine long-legged fly has not been studied in other comprehensive contexts such as molecular taxonomy, biogeography, distribution pattern, ecology, and etc. Regarding this study, the author gladly provided an update of the species composition, species distribution, habitat preferences, taxonomic status, species identification, and including population genetic of the marine long-legged fly in Thailand.

#### 1.2) Literature Review

#### 1.2.1) Dolichopodidae s. str. Characteristics, Habitats, and Food Habits

#### 1.2.1.1) Dolichopodidae s. str. Characteristics

Adult long-legged fly (Order: Diptera; Family: Dolichopodidae) (Figure 1) can be easily recognized by their elongated legs and head, body slender in build, reduced wing venation, aristate antenna (Robinson and Vockeroth, 1981), and ranged in size (1 to 9 mm) (d'Assis Fonseca, 1978, Robinson and Vockeroth, 1981). Although, most species are metallic greenish blue of grounded body. Except some marine and non-marine species, which are expressed in various and different colours such as dark brown to blackish in genus *Argyrochlamys* Lamb, *Cymatopus* Kertész, *Diaphorus* Meigen, and; non-metallic yellowish, for example, *Chaetogonopteron* de' Meijere and *Neurigona* Róndani. In the meantime, larvae of these flies also contain a distinct form of body, it's mostly white, cylindrical and relatively slender build with distinct creeping welts on segments 4th to 11th (Brooks, 2005). Whereas, Dyte (1967) and Robinson and Vockeroth (1981) tell us that the pupae of known species of these flies possess spiniferous transverse bands on the abdominal tergites. One of the most unique external morphological features is wing venation pattern. It is obviously absent of a cross vein between the discal cell and 2nd basal cell (cell M and 1st M<sub>2</sub> united to form a large single cell) (Cregan, 1941).

This family is notable as sexual dimorphism. The great majority of adult males show excessive striking secondary sexual characters (MSSCs) and their identification is therefore easily distinguished. MSSCs include modifications of the antennae, palpi, wing apex, and typically, the legs, which play an important role in courtship behaviour (Cregan, 1941; d'Assis Fonseca, 1978; Lunau, 1992; Zimmer et al., 2003; Grootaert, 2004). The male hypopygium is small and partially concealed by preceding abdominal segments, or large, permanently supported by a pedunculated abdominal segment (Snodgrass, 1904; Ulrich 1974; and Cumming et al., 1995). Whereas the adult female is paid less attention to by taxonomists in terms of species identification. It is due to the less striking morphological characteristic than the male (Cregan, 1941) and leading to difficulty to name.



Figure 1. Cymatopus thaicus Grootaert and Meuffels, male habitus

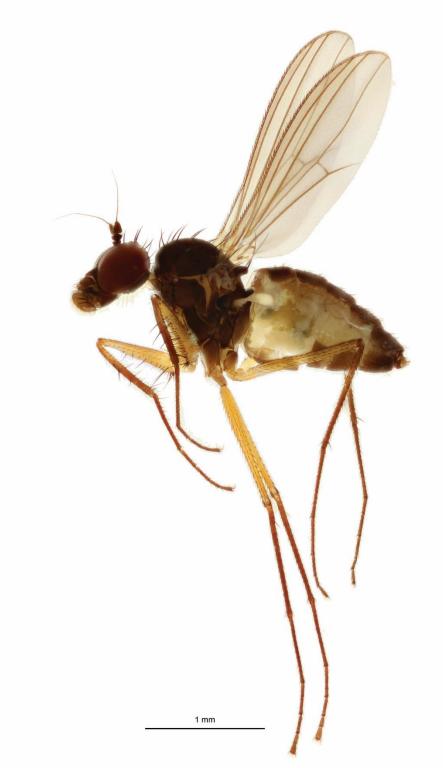


Figure 2. Cymatopus thaicus Grootaert and Meuffels, female habitus

### 1.2.1.2) Habitats

This fly family is widely distributed. They mostly occur in wet and moist areas. Pollet (2000, 2001) tells us that long-legged flies is one dipteran family encountered in all terrestrial habitats from forests to semi-aquatic and they are abundantly diverse in marshy areas such as humid forests, mangroves (Grootaert, 2006a; Zhang et al., 2008), and all kinds of riparian ecosystems (Pollet, 2000; Grootaert and Meuffels, 2004). Similarly, Miall (1934) has long provided very important information about this fly's habitat, he tells us that "the naturalist, in search for aquatic insects, cannot fail to find them almost daily and hourly". This sentence is an evidence to confirm that the long-legged fly is cosmopolitan, abundant, largely diverse, and generally found in the neighborhood of water. He further mentions about this fly that they always fly to rest on the grass and bushes near to water, standing on stones in the bed of streams, and some hover surrounded in the spray of waterfalls (Miall, 1934).

Some genera of long-legged flies, mostly dwelled and predated their preys at the seacoast or seashores. For instance, Ngirhaphium Evenhuis and Grootaert (Evenhuis and Grootaert, 2002; Grootaert and Puniamoorthy, 2014; Samoh et al., 2015), is obviously found in mudflats of back and mid mangroves, and also commonly distributed at the sea front. The genus Asyndetus Loew belongs to subfamily Diaphorinae, is presumed commensal flies of crab burrows, and is commonly found along beaches of seacoast in southern hemisphere (Grootaert and Meuffels, 2002). According to the list of Meuffels and Grootaert (1993), he listed 11 species from Papua New Guinea. In Thailand, four species were recorded (Grootaert and Meuffels, 2002), including, Asyndetus ciliatus, A. aciliatus, A. thaicus, and A. latifrons. Furthermore, Argyrochlamys Lamb, the generic name of this fly related to the place that they were mostly found. They are generally observed near the entrance zone of the ghost crab burrows and seem to be restricted to beaches with high sun-exposure. In addition, they can be easily recognized by their nonmetallic body, pale yellowish brown to dark grey colour, and also contain 5 pairs of dorsocentrals. Males can be easily identified by the distinctive "comma shape" or "bifurcate projection" (Brooks, 2005) near the joint of the hind tibia and basitarsus. This genus is mainly record from Afrotropic, for example, Ghana, Mauritius, Somalia, Sudan (Dyte and Smith, 1980) and is also reported from Oriental realm, Chagos Island Srilanka (Dyte, 1975). Nanothinophilus Grootaert and Meuffels, the holotype of minute

Nanothinophilus hoplites was collected from mudflats in mangroves at Ao Nang, Krabi, Andaman Sea (Indian Ocean), during low tide in 1997 by Grootaert and Meuffels (Grootaert and Meuffels, 2001). The known species in Thailand included N. armatus Grootaert and Meuffels, N. pauperculus Grootaert and Meuffels, N. dolichurus Grootaert and Meuffels, and N. hoplites Grootaert and Meuffels. Thambemyia Oldroyd, the type species of this genus is first designated by Oldroyd in 1956 as a monotypic genus. Thambeyia Oldroyd, 1956, Acymatopus Takagi, Conchopus Takagi are more closely related to each other than Cymatopus Kertész. They are widely distributed throughout Southeast Asia: (Brunei, Malaysia, and Thailand), Hong Kong, Taiwan, China to Japan. In Thailand, they were recently recorded from Pakbara beaches and rocky shores, Satun province in Andaman seacoast. Whereas marine Thinophilus Wahlberg, only three species of *Thinophilus* have been discovered from Thailand until now: *T. nitens* Grootaert and Meuffels, T. parmatus Grootaert and Meuffels and T. setiventris Grootaert and Meuffels. The genus Thinophilus Wahlberg is one of the most diverse groups of dolichopodid flies inhabiting coastal environments (Grootaert et al. 2015). They are adapted to and survive excellently in marine habitats such as front, mid and back mangroves, tide pools, mudflats, sandy beaches and rocky shores. Lastly, Phoomyia Naglis and Grootaert or non-metallic dolichopodine fly, is usually found at beaches near sea front with high sun exposure and are easily recognised by these following combination characters: head and thorax grey, non-metallic body, moreover, one highly distinctive characteristic is its abdomen is mostly yellow or brown with grey pruinosity. In Thailand, only one species (Phoomyia thailandensis) was recoded from Gulf of Thailand (Naglis et al., 2013).

Lots of species of *Tachytrechus* Haliday live very near to sandy brooks and also mostly found resting on the boarder of dams (Schiner, 1862), a species known as *Tachytrechus tessellatus* is a large size Dolichopodinae which is highly active in open, sunny, and wet habitats. They are generally observed near shallow pools, tide pools. It can be recognized by its peculiar clypeus, which mostly extends beyond the lower eye margin and is rounded below. Moreover, this species has a wide range of distribution and is recorded from Thailand, India, to the African continent (Yang et al., 2006).

The terrestrial species of long-legged fly may be observed on tree trunks (Cregan, 1941) such as *Sciapus*, *Medetera*, *Neurigona* (Brooks, 2005), some were found on the

leaves of shrubs, damn soils (Cregan, 1941), some were reported from moss algal mats, sap wounds, leaf litters (Dyte, 1959), while others occur in drier habitats such as gardens, agriculture areas, for instant, some species of the *Medetera* and *Dolichopus* (Brooks, 2005).

Due to their specific habitat requirements, dolichopodids show high potential as bioindicators (Grootaert and Meuffels, 2004; Pollet, 2001; 2009). High diversity of dolichopodid flies is the determiner of undisturbed area or good quality of habitat. Conversely, low diversity is an indicator for a poor quality of habitat (Grootaert and Meuffels, 2004).

#### **1.2.1.3)** Food Habits

Being predacious or carnivorous insects, this pattern is generally a habit of both adults and larvae of Dolichopodidae (Smith and Empson, 1955; Ulrich, 2005). Long-legged flies preferably predate on other small and soft-bodied invertebrates. For example, Doane (1907), provides us a little information about food habits of particular *Scellus virago* found on thinly-encrusted salt areas bordering San Francisco bay, they enjoy consuming blood of smaller and weaker *Rhicnoessa parvula* Lw. (Agromyzidae) using their powerful forelegs. Lutz (1918) similarly states that the adult long-legged flies are all predacious insects, foraging mostly minute and soft-bodied insects, especially flies. Whereas, Aldrich (1922) proposes this fly chiefly preys on microscopic, weaker dipterous families such as small Chironomids and other fly larvae, as well as fleshy oligochaete worms near the edge of the water. Williams (1938) mentions that minute collembola and drossophilid flies seem to be the most preferred prey of *Campsicnemus fumipennis* Parent. While Ulrich (2005) also tells us that long-legged flies mostly predate on various groups of smaller invertebrates such as Chironomidae, Culicidae, Homopterans, Collembolans, mites, and Thysanopterans.

Due to the above facts, it could be said that long-legged flies play such a great role in the ecosystem (as carnivorous insects), to the extent as to warrant special status as a natural enemy for pest management and also a keystone species.

#### 1.2.2) Classification and Phylogeny of Dolichopodidae s.str.

### 1.2.2.1) Classification Based On External Morphology

Absorbingly, lots of fundamental work on taxonomy and systematics of Dolichopodidae s.str. include Loew (1864), Lundbeck (1912), Becker (1917, 1918, 1922), Parent (1938), Robinson (1964, 1975), d'Assis Fonseca (1978), Robinson and Vockeroth (1981), Bickel (1994), Evenhuis and Grootaert (2002), Ulrich (2003, 2004, 2005), Yang and Yang (2004), Brooks (2005), Yang et al. (2006), Sinclair and Cumming (2006), Lim et al. (2010), Germann et al. (2011). Although Brooks (2005) tells us that the number of described species of Dolichopodidae is increasing drastically and the end is not yet in sight. Furthermore, it has long been recognized that there are many problems with the higher-level classification of the family and comprehensive review of world subfamilies, genera, and including species are paramount (Robinson and Vockeroth 1981). Many dolichopodid subfamilies have not been the subject of many studies in a phylogenetic context on a world scale, have uncertain limits and are mostly questionably monophyletic (Diaphorinae, Hydrophorinae, Peloropeodinae, Rhaphiinae, Sympycninae). Moreover, virtually nothing is known about the phylogenetic relationships of the genera within these subfamilies. Moreover, Thailand is still lacking the information pertaining to the family Dolichopodidae in various contexts such as biodiversity, ecology, phylogeny, behaviour, etc. Thus, primary work such as taxonomic review and advancement in information, such as phylogenetic relationships and DNA barcoding, of these flies are needed.

In general, the genera of Dolichopodidae have been proposed into subfamilies by various dolichopodid fly taxonomists over the past 150 years (Lioy, 1863; Schiner, 1864; Aldrich, 1905; Kertész, 1909; Lundbeck, 1912; Becker, 1917, 1918, 1922; Curran, 1934; Robinson 1970a, 1970b; Ulrich 1981; Negrobov, 1986). In addition, some of dolichopodid fly classification ideas are shown as follows:

*Classification of Lioy* (1863). — The earliest classification was that of Lioy (1863), who introduced the concept of the family Dolichopodidae (Table 1.), includes;

Table 1. Lioy's classification idea

Lioy	Lioy (1863)		
	FAMIGLIA SCENOPINITI		
	(= Scepinidae)		
	FAMIGLIA LONCHOPTERITI		
	(= Lonchopteridae)		
	FAMIGLIA PLATYPEZITI		
	(= Platypezidae)		
	FAMIGLIA CEPHALOPSITI		
	(= Pipunculidae)		
	*FAMIGLIA HYDROPHORITI		
	*FAMIGLIA MEDETERITI		

Note that, [\*] indicates the arrangement by Lioy (1863), in which he divided the Dolichopodidae into two groups: Famiglia Hydrophoriti and Famiglia Medeteriti (Lioy,1863).

*Classification of Schiner* (1864).— The classification of Lioy (1863) has been criticized and notably ignored by subsequent taxonomists,— Schiner (1864), who divides the Dolichopodidae into four subfamilies and also published his own catalogue called European Diptera a few months later.

Table 2. Schiner's classification idea

Schiner (1864) DOLICHOPODIDAE RHAPHIINAE DOLICHOPODINAE HYDROPHORINAE DIAPHORINAE *Classification of Aldrich* (1905). — The Nearctic region long-legged flies or the American genera have already been classified by Aldrich (1905) into twelve subfamilies as follows:

Table 3. Aldrich's classification idea

### AGONOSOMINAE

- 1. Psilopodinus
- 2. Agonosoma
- 3. Mesorhaga
- 4. Leptorhethum

### DIAPHORINAE

- 1. Diaphorus
- 2. Asyndetus
- 3. Chrysotus
- 4. Eutarsus
- 5. Teuchophorus
- 6. Campsicnimus

## RHAPHIINAE

- 1. Argyra
- 2. Leucostola
- 3. Porphyrops
- 4. Rhaphium
- 5. Nematoproctus
- 6. Syntormon

### APHROSYLINAE

1. Aphrosylus

## PLAGIONEURINAE

1. Plagioneurus

### SYMPYCNINAE

- 1. Parasyntormon
- 2. Sympycnus
- 3. Nothosympycnus
- 4. Anepsiomyia

## XANTHOCHLORINAE

- 1. Achalcus
- 2. Chrysotimus
- 3. Xanthochlorus
- 4. Xanthima

### THINOPHILINAE

- 1. Thinophilus
- 2. Diostracus
- 3. Hypocarassus
- 4. Phylarchus

## NEURIGONINAE

1. Neurigona

### MEDETERINAE

- 1. Medeterus
- 2. Peloropeodes
- 3. Thrypticus
- 4. Coeloglutus

## HYDROPHORINAE

- 1. Hydrophorus
- 2. Scellus
- 3. Liancalus

## DOLICHOPODINAE

- 1. Dolichopus
- 2. Gymnopterus
- 3. Hercostomus
- 4. Paraclius
- 5. Tachytrechus
- 6. Polymedon
- 7. Sarcinus
- 8. Pelastroneurus
- 9. Leptocorypha
- 10. Orthochile

*Classification of Kertész* (1909).— Kertész (1909) classification, subsequently followed Schiner's (1864) classification in his catalog of the Palaearctic species, but also recognized Aldrich's New World subfamily Plagioneurinae.

*Classification of Lundbeck* (1912). — In his treatment of the Palaearctic fauna is literally followed by the classification of Kertész (1909) and a person who has criticized the classification of Aldrich (1905), he stated that the subdivisions of family of Dolichopodidae in subfamilies is at present not satisfactory, but considered Aldrich's (1905) system to be a more natural arrangement of the genera.

*Classification of Becker* (1922a).— A good classification by Becker was invented in 1922a (in general, Becker's idea has a sequencing series; 1917-1918, and 1922a) and considered as one of the most natural taxonomic arrangements of this particular group of flies according to his idea and widely adopted by dolichopodid fly taxonomists.

Table 4. Becker's classification idea

#### DOLICHOPODINAE

- 1. Dolichopus Latr.
- 2. Hygroceleuthus Lw.
- 3. Hercostomus Lw.
- 4. Paraclius Lw.
- 5. Pelastoneurus Lw.
- 6. Sarcionus Aldr.
- 7. Stenopygium Becker
- 8. Tachytrechus Walk.
- 9. Polymedon O. S.
- 10. Macellocerus Mik.
- 11. Psilichium Becker
- 12. Sybistroma Meig.
- 13. Leptocorypha Aldr.
- 14. Gonioneurum Becker

#### **HYDROPHORINAE**

- 1. Hydrophorus Fall.
- 2. *Scellus* Lw.
- 3. Liancalus Lw.
- 4. Thinophilus Walk.
- 5. Diostracus Lw.
- 6. Hypocharassus Mik.
- 7. Syntomoneurum Becker.
- 8. Phylarchus Aldr.
- 9. Peodes Lw.

#### APHROSYLINAE

1. Paraphrosylus Becker

#### PLAGIONEURINAE

1. Plagioneurus Lw.

Classification of Becker (1922a) (continued)

### MEDETERINAE

- 1. Medeterus Fisch.
- 2. Thrypticus Gerst.

### RHAPHIINAE

- 1. Rhaphium Meig.
- 2. Porphyrops Meig.
- 3. Xiphandrium Lw.
- 4. Syntomon Lw.
- 5. Eutarsus Lw.
- 6. Achalcus Lw.
- 7. Peloropeodes Wheel.
- 8. Systenus Lw.

### NEURIGONINAE

1. Neurigona Rond.

### DIAPHORINAE

- 1. Diaphorus Meig.
- 2. Lyroneurus Lw.
- 3. Chrysotus Meig.
- 4. Coeloglutus Aldr.
- 5. Asyndetus Lw.
- 6. Argyra Meig.
- 7. Leucostola Lw.
- 8. Achradocera Becker
- 9. Symbolia Becker
- 10. Xanthina Aldr.

### STOLIDOSOMINAE

1. Stolidosoma Becker

### CAMPSICNEMINAE

- 1. Campsicnemus Halid.
- 2. Sympycnus Lw.
- 3. Subsympycnus Becker
- 4. Hypteochaeta Becker
- 5. Calysochaetus Big.
- 6. Chrysotimus Lw.
- 7. Xanthochlorus Lw.
- 8. Anepsiomyia Bezzi.
- 9. Teuchophorus Lw.

### CHRYSOSOMATINAE

- 1. Condylostylus Big.
- 2. Megistostylus Big.
- 3. Mesorhaga Schin.
- 4. Leptorhetum Aldr.
- 5. Sciapus Zell.

### **GENUS INCERTAE SEDIS**

1. Anchineura Thoms.

**Remarks**: The subfamily arrangement by Becker (1922a) was mainly based on several external morphological features such as first joint of antenna (in Dolichopodinae); typical male hypopygium lied completely at 6<sup>th</sup> abdominal segment (Hydrophorinae); bare thorax of Aphrosilinae, but nothing was stated about the mouthparts. Interestingly, his classification seemed to be a precious foundation for the current taxonomy of Dolichopodid flies study.

Up to this time, lots of ways of dolichopodid fly classifications have been several times revised as shown in Table 5 below:

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	Robinson (1970)	Yang et al. (2006); Sinclair et al. (2008)	Pollet and Brooks (2008)
			Dolichopodidae s.lat.
1			Microphorinae <sup>A</sup>
2			Parathalassiinae <sup>A</sup>
	Dolichopodidae	Dolichopodidae	Dolichopodidae s.str.
3		Achalcinae <sup>B</sup>	Achalcinae
		[Antyxinae]	
4		Babindellinae <sup>C</sup>	Babindellinae
5	Diaphorinae	Diaphorinae	Diaphorinae
6	Dolichopodinae	Dolichopodinae	Dolichopodinae
7	Enliniinae	Enliniinae	Enliniinae
8	Hydrophorinae <sup>D</sup>	Hydrophorinae	Hydrophorinae
		[Kowmunginae]	
9	Medeterinae	Medeterinae <sup>E</sup>	Medeterinae
10	Neurigoninae	Neurigoninae	Neurigoninae
11	Peloropeodinae	Peloropeodinae	Peloropeodinae
12	Plagioneurinae	Plagioneurinae	Plagioneurinae
13	Rhaphiinae	Rhaphiinae	Rhaphiinae
14	Sciapodinae	Sciapodinae	Sciapodinae
15	Stolidosomatinae	Stolidosomatinae	Stolidosomatinae
16	Sympycninae	Sympycninae	Sympycninae
	[Systeninae]		
17	Xanthochlorinae	Xanthochlorinae	Xanthochlorinae

Table 5. Overview and comparative of the most relevant classifications of Dolichopodidae from the 1970s by Robinson until present

**Note that** <sup>A</sup>Sinclair and Cumming (2006). <sup>B</sup>Grootaert and Meuffels (1997). <sup>C</sup>Bickel (1987). <sup>D</sup>Aphrosylinae are treated as synonyms of Hydrophorinae in all three classifications. <sup>E</sup>Bickel (1986): *Systemus* Loew transferred to Medeterinae, rendering Systeminae (see Robinson, 1970a) obsolete. Moreover, superscript codes in the Table refer to papers holding post Robinson (1970a) taxonomic changes. Subfamilies in brackets are considered controversial (modified from Germann et al., 2011).

#### 1.2.2.2) Classification Based On Molecular Data

At present, molecular markers from mitochondrial DNA such COI (Cytochrome Oxidase subunit I) is largely accepted by modern taxonomists and used in phylogenetic reconstruction within dipteran families (Meier and Wiegmann 2002; Bernasconi et al., 2007; Petersen et al. 2007; Kutty et al. 2007, 2008, 2010; Su Feng Yi et al. 2008; Ståhls et al., 2009; Lim et al., 2010; Germann et al., 2010; Germann et al., 2011; Pollet et al., 2010; Pollet et al., 2011; Renaud et al., 2012; Laurito et al., 2013; Pramual et al., 2016), reference to long-legged flies are shown as follows:

*Classification of Bernasconi et al.* (2007).— the classification treatment of Bernasconi et al. is mainly based on COI and combined with 12S rDNA gene or dataset (1199 characters) and according to the results of the same authors studied, the 101 European species of long-legged flies could be divided into seven subfamilies (Table 6), and only three subfamilies were considered as monophyletic relationships (Figure 3) (Dolichopodinae, Sympycninae, and Hydrophorinae) (all investigation based on various phylogenetic analysis; including Bayesian [BAY], Neighbour-Joining [NJ], weighted-unweighted Maximum Parsimony [MP] analysis) as shown as follows:

Table 6. Overview idea of Bernasconi et al.'s classification

DOLICHOPODINAE	RHAPHIINAE
1. Dolichopus	1. Rhaphium
2. Hercostomus	
3. Sabystroma	SCIAPODINAE
4. Poecilobrthrus	1. Sciapus
5. Gymnopternus	
	SYMPYCNINAE
DIAPHORINAE	1. Campsicnimus
1. Chrysotus	2. Syntormon
2. Argyra	3. Teuchophorus
HYDROPHORINAE	
1. Hydrophorus	
2. Lianchalus	
MEDETERINAE	

1. Medetera

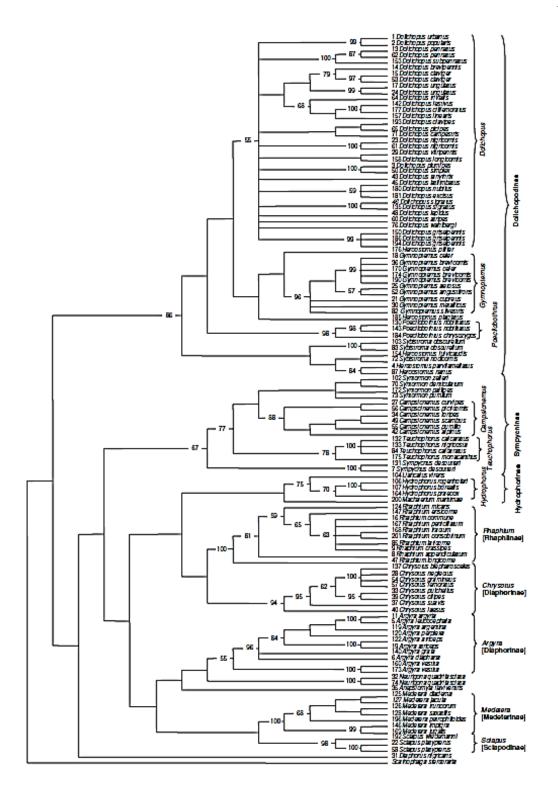


Figure 3. Phylogenetic hypothesis for dolichopodid fly relationships based on Neighbour-Joining (NJ) analysis (modified from Bernasconi et al., 2007)

*Classification of Lim et al.* (2010). — This classification was addressed by Lim and colleagues in 2010 using six genes from both nuclear (18S, 28S) and mitochondrial genes (12S, 16S, Cytb, and COI). In addition, this good analysis was experimented based on 76 Oriental species from twelve dolichopodid subfamilies and uses eight species of Empididae and Hybotidae as outgroups. Finally, they could classify dolichopodids or long-legged flies into twelve subfamilies, confirming the monophyly of five from the twelve subfamilies and also restoring the tribe Aphrosilini to subfamily Aphrosilinae (based on Maximum Likelihoods Analysis) (Table 7, Figure 4), shown as follows:

Table 7. Overview idea of Lim et al.'s classification

APHROSILINAE*	NEURIGONINAE*			
1. Cymatopus	1. Neurigona			
2. Thambemyia	PARATHALASIINAE*			
3. Thinolestris	1. Eothalassius			
DIAPHORINAE	2. Microphorella			
1. Asyndetus	PELOROPEODINAE**			
2. Chrysotus	1. Nepalomyia			
3. Diaphorus	<b>PELOROPEODINAE</b> **			
<b>DOLICHOPODINAE*</b>	1. Griphophanes			
1. Argyrhochlamys	<b>PELOROPEPDINAE**</b>			
2. Dolichopus	1. Acropsilus			
3. Hercostomus	2. Scotiomyia			
4. Licthwardtia	RHAPHIINAE			
5. Paraclius	1. Ngirhaphium			
6. Tachytrechus	SCIAPODINAE*			
HYDROPHORINAE*	1. Amblypsilopus			
1. Nanothinophilus	2. Chrysosoma			
2. Thinophilus	3. Plagiozopelma			
KOWMUNGINAE	SYMPYCNINAE*			
1. Phacaspis	1. Chaetogonopteron			
MEDETERINAE	2. Hercostomoides			
1. Medetera	3. Teuchophorus			
	4. Syntormon			

Note that asterisk (\*) is indicated monophyletic and (\*\*) paraphyletic

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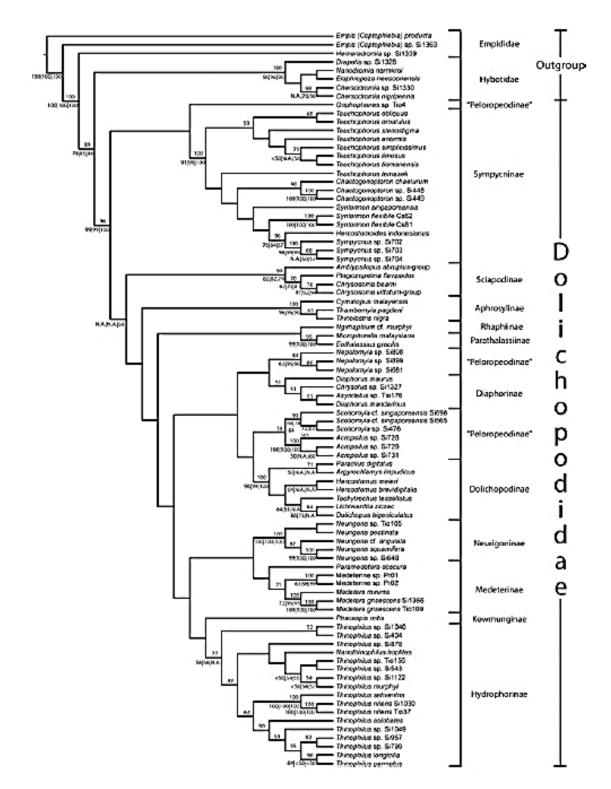


Figure 4. Phylogenetic hypothesis for dolichopodid fly relationships based on Maximum Likelihood (ML) analysis (modified from Lim et al., 2010)

*Classification of Germann et al.* (2011). — This is the latest molecular phylogenetic classification of long-legged flies. Their classification (based on various mitochondrial and nuclear markers, for instance, COI, 12S, 16S, and 18S) is intended to investigate the current, external morphology-based dolichopodid fly classifications such as Robinson, 1970; Yang et al., 2006; and also Pollet and Brooks, 2008. Furthermore, in order to increase the level of understanding of the phylogeny at a higher taxonomic level of the world scale. Finally, they found strong clues from two methods of the phylogenetic relationships, namely, Bayesian (BAY) and Maximum Likelihoods (ML) that the Microphorinae is a sister group of Dolichopodidae *sensu stricto*, and they also revealed that Achalcinae, Dolichopodinae, Parathalassiinae, Sciapodinae are monophyletic relationship, and grouped Stolidosomatinae within Sympicninae. Whereas, Diaphorinae, Medeterinae, Neurigoninae, Rhaphiinae, and Sympycninae are paraphyletic relationship, and presented Hydrophorinae and Peloropeodinae polyphyletic relationships (Table 8, Figure 5) as follows:

Table 8. Overview idea of Germann et al.'s classification

SUPERFAMILY EMPIDO	IDEA			
• FAMILY EMPIDIDAE				
• FAMILY HYBOTIDAE	Ξ			
• FAMILY DOLICHOPO	<b>DDIDAE</b> sensu lato			
Subfamily Microphorinae*				
FAMILY DOLICHOPODIDAE sensu stricto				
MONOPHYLY	PARAPHYLY	POLYPHYLY		
1. Achalcinae	1. Diaphorinae	Hydrophorinae		
2. Dolichopodinae	2. Enliniinae	<ul> <li>Lineage A</li> </ul>		
3. Parathalassiinae*	3. Neurigoninae	<ul> <li>Lineage B</li> </ul>		
4. Sciapodinae	4. Medeterinae	<ul> <li>Lineage C</li> </ul>		
5. Stolidosomatinae <sup>A</sup>	5. Rhaphiinae	Peloropeodinae		
INCERTAE SEDIS	6. Sympycninae	<ul> <li>Lineage A</li> </ul>		
1. Kowmunginae		<ul> <li>Lineage B</li> </ul>		
2. Xanthochlorinae				

\* note that it is traditionally considered as sister taxa to Dolichopodidae sensu lato (Sinclair and Cumming, 2006; Pollet and Brooks, 2008); <sup>A</sup> note that it is monophyletic relationships but placed within Sympycninae and it should be sunk as a separate lineage into Sympycninae (Germann et al., 2011).

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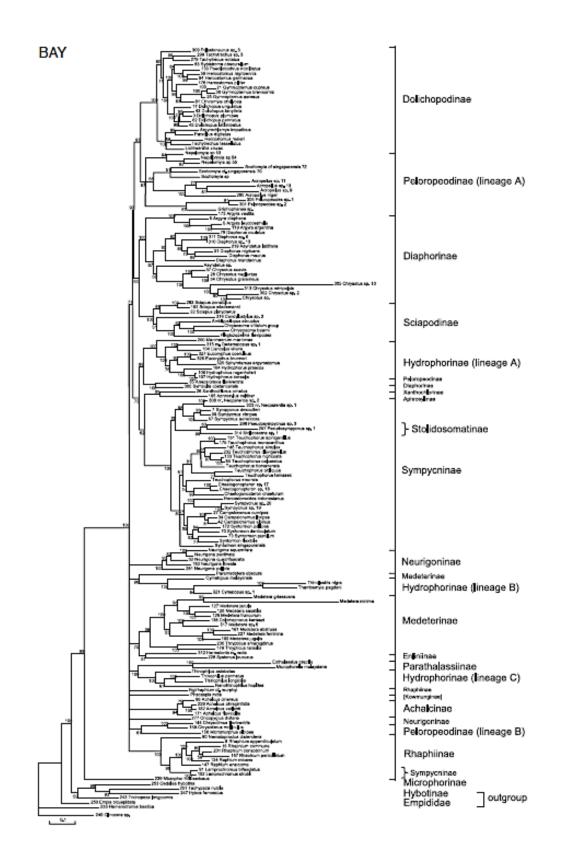


Figure 5. Phylogenetic hypothesis for dolichopodid fly relationships based on Bayesian (BAY) analysis (modified from Germann et al., 2011).

## 1.2.1.3) Diversity of Dolichopodidae sensu stricto

## **1.2.1.3.1) World Scale**

Although family Dolichopodidae *sensu stricto* is meticulously study from all zoogeographic regions (Robinson 1970a, 1970b; Dyte 1975; Dyte and Smith 1980; Bickel and Dyte 1989; Negrobov 1991; Pollet et al. 2004), the species number of long-legged flies is rather ranged and assorted (from 6,780 until 7,755 known species). For instance, Yang et al. (2006) listed 6780 species, 226 genera in their World Catalog of Dolichopodidae; over 7,100 described species in 220 genera (Pape et al., 2009); recently, with 7,755 valid species (including 110 fossil species), 277 valid genera (including 31 fossils) from all zoogeographical regions (Grichanov, 2014), but in fact, this species number is low, and not yet in sight and very far from any finale since many places in tropical regions are utterly poorly surveyed or lacking attention (Grootaert, 2009).

# 1.2.1.3.2) Asia and Southeast Asia Scale

Throughout Asia Continent, only China and Singapore are the leading countries that extensively study family Dolichopodidae, particularly marine species have been most exhaustively studied, resulting in several good publications. Regarding marine and nonmarine dolichopodid flies, many species have been described from this region, shown as follows:

**Singapore**, over forty-four species of marine Dolichopodids have been recognized from the whole of Singapore island (Evenhuis and Grootaert, 2002) and tend to be coherently increased; in addition, both Grootaert and his colleagues expected that the number of marine shore Dolichopodids in Singapore republic much higher than this record.

Regarding Grootaert (2006b, 2006c), he has reported five species of genus *Paramedetera* for the first time from Singapore and also described and illustrated four new species. They were *P. micropyga*, *P. obscura*, *P. chelata*, and *P. digitate* (Grootaert, 2006b). In the meantime, he also reported thirteen species of genus *Teuchophorus* (Dolichopodidae: Sympycninae) from this region. Nine were considered as new species. One species belongs to the *T. conspicuous-notabilis* group and found: *T. ornatulus* Meuffels and Grootaert. Three species classified belong to the *T. gratiosus* group, namely, *T. temasek*, *T. neesoonensis*, and *T. antennatus*. Four new species belong to the

*T. pauper* group: *T. singaporensis*, *T. meieri*, *T. spinulosus*, and *T. acuminatus* (Grootaert, 2006c).

A year later, Zhang et al. (2007) reported nine species of mangrove *Paraclius* Loew from Singapore. Seven of them were considered as new species, namely, *P. asiobates, P. crassatus, P. digitatus, P. obtus, P. polychaetus, P. serratus,* and *P. singaporensis.* 

While, Zhang et al. (2008) extended seven new species of mangrove *Hercostomus* Loew collected from several mangroves in Singapore. Seven species were included *H. brevidigitalis*, *H. brevicornis*, *H. lanceolatus*, *H. limosus*, *H. meieri*, *H. plumatus*, and *H. singaporensis*.

In 2013, Grootaert reported seven new species of *Nepalomyia* Hollis from lowland Singapore: *N. priapus*, *N. negrobovi*, *N. singaporensis*, *N. spinata*, *N. temasek*, and *N. yangi*, while *N. harpago*, was described from South Peninsular Malaysia.

Since the most recent paper of Grootaert and Puniamoorthy (2014) related to a taxonomic revision and re-description of Dolichopodiadae in the genus *Ngirhaphium* Evenhuis and Grootaert was published. In the meantime, they recognised and also provided a new identification key and description of the two new species (including *N. sivasothii* and *N. caeruleum*) from Singapore mangroves.

**China** also provided a great number of marine and non-marine Dolichopodidae, such as six species of genus *Hercerstomus* Loew (Negrobov, 1991) from North China, when six new species of genus *Hercostomus* Loew have been added from this region by Yang (1996), six new species of Dolichopodidae including *H. flavimaculatus*, *H. qingchenganus*, *H. tianmushanus*, *Ludovicius sichuanens*, *Mesorhaga guangxiensis*, and *Amblypsilopus guangxiensis* reported from South China by Yang (1998). The 225 species of *Hercostomus* have been added to the fauna of China since 1995 mainly based on the work of Yang and Yang (1995), Wei (1997), Yang and Grootaert (1999), Yang and Saigusa (1999, 2000, 2001a, 2001b, 2001c, 2001d, 2002), Zhang and Yang (2003a, 2005), Zhang et al. (2004, 2005, 2007b). Recently, 253 species of *Hercostomus* Loew are known to occur in China.

*Asyndetus* Loew is a genus that has been extensively studied in China such as Becker (1922b); Parent (1926); Negrobov (1973) proposed five species; Zhang and Yang (2003b) described two new species: *A. beijingensis, A. guangxiensis.* When Wang et al. (2007b) provided further data of this genus for the country.

2006, was a big highlight year for Chinese Dolichopodid fly taxonomic study. Yang et al. (2006) published a tremendously good "*World Catalog of Dolichopodidae*", with more than 7,000 species listed in their book and also illustrated with great drawing and basic information.

In 2007c, Wang et al. reported, revised and named seven new species of *Acropsilus* Mik, 1878 including *A. guangdongensis, A. guangxiensis, A. jinxiuensis, A. luoxiangensis, A. yunnanensis, A. zengchengensis* and *A. zhuae* from China.

The family Dolichopodidae taxonomy in China has been published in high rate, Zhang and Yang (2008) recognised three new species of *Dolichopus* Latreille from China; including *Dolichopus jiliensis*, *D. longipilosus*, and *D. yangi*.

Wang et al. (2010) described and illustrated three new species of the genus *Neurigona* Róndani (*N. hainana* from Hainan province, *N. sichuana* from Sichuan province and *N. yaoi* from Neimenggu province) and *N. zhangae* is transferred as *Viridigona zhangae*.

Wang et al. (2012) erected three new species of long-legged flies (*C. dalongensis*, *C. huairouensis*, and *C. hubeiensis*) and one new record (*C. apicicurvatus*) of the genus *Chrysotimus* Loew from Palearctic China.

Wang et al. (2015) reported a new data of the two genera, including genus *Acropsilus* and genus *Chrysotimus* from Taiwan, with provided species identification key and also described a species new to science, namely, *C. taiwanensis* Wang and Yang.

Tang et al. (2016a) discovered and described ten new species of *Medetera* Fischer von Waldheim from Inner Mongolia, China. They were *M. albens, M. bisetifera, M. flava, M. ganshuiensis, M. lihuae, M. transformata, M. triseta, M. shiae, M. shuimogouensis*, and *M. xiquegouensis*. Moreover, a key to the species of *Medetera* from Palearctic China is also provided.

Tang et al. (2016b) recorded and described six new species of the genus *Rhaphium* Meigen; *R. apophysatum, R. bilobum, R. bisectum, R. daqinggouense, R. dorsiseta,* and *R. neimengense* from China.

Unfortunately, very little information is available about Dolichopodidae (Grootaert, 2006a, 2006b; Zhang et al., 2007; Grootaert, 2013) have been recorded from other Southeast Asia (SEA) countries such as Brunei, Indonesia, Malaysia, and Philippines. In addition, it is woefully needed for better understanding of large scale of species distribution and composition.

# 1.2.1.3.3) Thai Dolichopodid fly Study and Big Gap of Knowledge

Fly surveys, especially in Southeast Asia (SEA), have been undertaken (Delfinado and Hardey, 1973, 1975, 1977; Oosterbroek, 1998; Grootaert and Meuffels 1997a, 1997b, 1997c, 1998a, 1998b, 1998c, 1999a, 1999b, 2001, 2002; Bickel, 1999; Shamshev and Grootaert, 2004a, 2004b, 2005a, 2005b). Papp and Ševčík, 2005; Papp, 2005a, 2005b; Papp et al., 2006;

Thailand is one of the most targeted areas for overseas researchers to study flies in any contexts. Although Papp et al. (2006) claimed Thai dipteran species survey is not facing endangerment, then this needs some attention!. In addition, lots of first records of species and numerous works have been published in the last three decades than formerly, in fact, very few valuable publications of the marine long-legged fly have been published in the last decade. Taxonomy warrants most concern. Several dolichopodid fly species have been promoted to be new species. Furthermore, in order to provide a regional key and to reach a correct interpretation of phylogenetic relationship among the marine longlegged flies of Thailand, a historical review of literature of these flies recorded from Thailand until now will be needed, concurrently the localities of specimens collected from Thailand by miscellaneous people.

Grootaert and Meuffels (1997a), two taxonomists who are the pioneers on marine long-legged flies in Thailand. They described the three new *Paramedetera* species from Thailand; there were *Paramedetera turschi*, *P. ankarum* and *P. horrorifera*. Furthermore, Grootaert and Meuffels (1997b) recognized a new genus, *Griphomyia* Grootaert and Meuffels from Thailand with *G. gravicaudata* as type species.

Grootaert and Meuffels (1998a) discovered a new genus from Thailand that is Haplopharynx, H. mutilus as a type specimen and H. phangngensis as a new species. Moreover, Grootaert and Meuffels (1998b) described *Nanothinophilus* a new genus from Thailand with *N. armatus* as a new species. Grootaert and Meuffels (1998c) published new locality data on the species *Griphophanes gravicaudata* in Thailand.

Bickel (1999a) described new species of the genus *Mastigomyia* Becker from Thailand; including *Mastigomyia amami* and *M. trangensis*. Grootaert and Meuffels (1999b) rediscovered *Terpsimyia semicincta* Becker from this country. In the same year, Grootaert and Meuffels (1999c) also described a new species of *Chaetogonopteron chaeturum*, which is considered to be very common throughout the country.

Presently, most of the known marine long-legged fly species in Thailand are largely recorded from Andaman seacoasts and Gulf of Thailand, Southern Thailand (Grootaert and Meuffels, 2001). They were investigated in various marine habitats throughout the Thai Peninsula, namely, Satun, Trang, Krabi, Phang Nga, Ranong, and also covering many provinces of the eastern coast of Thailand such as Rayong, Trad, Chantaburi, Chonburi, including the narrowest part of Thailand at Pachuap Khirikhan.

The following are the subfamilies, genera and species of marine long-legged flies occurring in Thailand (Figure 7) (Grootaert and Meuffels, 2001). Included are all types of marine habitat. All marine long-legged flies are characterized by elongated legs, relatively slender bodies, aristate antennae, and reduced wing venations (Robinson and Vockeroth, 1981). On the other hand, marine long-legged flies possess the opposite characteristics. So far as the literature is concerned, there are at least 15 species representing seven genera in two subfamilies and one *incertae sedis* (Figure 4) recorded from Thailand (Grootaert and Meuffels, 2001).

## Genus Phacaspis Grootaert and Meuffels, 1988

Grootaert and Meuffels (1989a) erected the genus *Phacaspis* in 1988. Morphological evidence is mainly based on only 2 species discovered in Thailand. These are *Phacaspis petiolata* Grootaert and Meuffels and *Phacaspis mitis* (new species). They are minute flies, about 1 mm in length, and mostly found on the mudflats in mangroves (Grootaert and Meuffels, 2001).

## Diagnostic characters of the genus Phacaspis

Male and female are described. A small species resembling *Phacaspis petiolata*. Thorax with 3 pairs of about equally long dorsocentrals. Fore femur ventrally with a row of very long, thin, straight setae. Mid and hind femora without longer hairs or bristles or totally bare. Fore tibia nearly as long as femur, without bristles; mid tibia not thickened. Hypopygium very long and reaching beyond base of third abdominal segment. Basal antennal segment with brown colour; Third antennal segment triangular, with an acute apex, longer than deep.

**Distribution**: Coast of Andaman Sea (Thailand) – Holotype – male, from mudflats in mangrove at low tide (near bridge, estuary) in Ao Nang, Krabi province; paratypes – 2 males and 2 females same provenance as holotype; Palian Trang; Pak Bara mangrove, Satun, collected 3 males and 1 female.

#### Genus Nanothinophilus Grootaert and Meuffels, 1988

The new genus *Nanothinophilus* Grootaert and Meuffels was established by Grootaert and Meuffels in 1988, currently known only from the Andaman sea coast in Southern Thailand. It is demonstrated by 4 species: *N. armatus* Grootaert and Meuffels, *N. pauperculus* Grootaert and Meuffels; *N. dolichurus* Grootaert and Meuffels, and *N. hoplites*, new species (see Grootaert and Meuffells, 2001).

#### Diagnostic characters of the genus Nanothinophilus Grootaert and Meuffels, 1988

Fore tibia can be easily recognized with a row of very long and strong, bent bristles. Fore femur ventrally with bristles that are longer than diameter of femur. Hind femur ventrally with 2 irregular series of bristles, 2 of which are longer than depth of femur. In addition, one more key character is Hypopygium that reaching beyond base of 4th abdominal segment.

**Distribution**: Coast of Andaman Sea (Thailand) – Holotype- male, from mudflats in mangrove at low tide in Ao Nang, Krabi province. Paratypes – 1 male and 2 females, same provenance as holotype.

#### Genus Thinophilus Wahlberg, 1844

It was firstly described by Wahlberg, belongs to subfamily Hydrophorinae. This genus sounds like others Dolichopodids genera that are widely distributed and exhibited a large number of male secondary sexual characters (MSSCs) (Grootaert, no year). The species live near water or aquatic flies. Not only observed in freshwater habitats but also commonly found in marine habitats with high sun exposed areas such as mud flats in mangroves, sandy beaches, and along creeks (Grootaert and Meuffels, 2001; Zhu et al.,

2006). Until now, there are 115 described species around the world, with 34 species in the oriental region (Dyte, 1975; Yang and Li, 1998). In addition, eighteen species occur in Palearctic region (Negrobov, 1991). In Thailand, only three species were described, they were *Thinophilus parmatus*, *T. nitens*, and *T. setiventris*. The Thai Peninsula is considered the richest area of the country for marine habitats, thus Grootaert and Mueffels (2001) assumed that *Thinophilus* Wahlberg should be found in high numbers of species and await description from Thailand.

## Diagnostic characters of genus Thinophilus Wahlberg, 1844

They are considered as medium in size of Dolichopodids from 2-7 mm. long, the face of both sexes are wide, metallic green with dull black spots on mesoscutum; pulpi brownish yellow and usually large; fore coxa of legs with yellow, hind tibia brown at basally; antennae yellow, dorsally browned; legs poorly bristled, fore femur of male ventrally with irregular rows of scattered, short bristles, including a row of 3-4 longer (Zhu, et al., 2006) posteroventral on apical third; wing cloud on tip, on apical third of  $R_{4+5}$  and on wing boss; third sternum in male with a cluster of hairs. These are clearly generic characters of marine *Thinophilus* Wahlberg.

# Distribution: Coast of Andaman Sea (Thailand)

Holotype- male of *T. nitens* and *T. setiventris*, from Wat Tapo Taram, river near hot springs, Ranong province, Thailand. Despite *T. parmatus* being found at rivers and estuaries of Takua Pa, Phang Nga province, the paratypes male and female were discovered in a mangrove at Palian district, Trang province.

## Genus Thinolestris Grootaert and Meuffels, 1989

The genus *Thinolestris* Grootaert and Meuffels, depicted by Grootaert and Meuffels, belongs to subfamily Hydrophorinae (as Aphrosylinae) (Grootaert and Meuffels, 1989b), (*Thinolestris* gen. nov., closely allies to *Cymatopus* Kertész). Two species were described from this region, *Thinolestris luteola*, a species from Coral Sea (Papua New Guinea) and *Thinolestris obscura*, a second species from North Sulawesi (Indonesia), although one species was recorded from Thailand, *Thinolestris thaica* (Grootaert and Meuffels, 2001) and it was the most western and northern record of the genus. This genus generally found at shaded areas of intertidal zones of beaches with patches of sand and pebbles.

#### Diagnostic characters of Thinolestris Grootaert and Meuffels, 1989

*Thinolestris* Grootaert and Meuffels, is recognized as small size of marine Dolichopodidae, less than 2.5 mm long. It is colourless metallic green. Face very broad in both sexes, head with convex occiput, palpi moderately large, and no postocellar bristles, antenna short with apical arista (Grootaert and Meuffels, 2001). However, the differences in the species of *T. thaica* and *T. luteola* (Coral Sea-Papua New Guinea) ought to be sought in the male genitalia. The somatic characters are inexplicable to distinguish the species. *Thinolestris luteola* has the tip of the hypandrium indented and differently shaped and bristled appendages on the abdominal sterna (Grootaert and Meuffels, 1989b; 2001).

**Distribution**: Grootaert and Evenhuis (2006) tell us that adults *Thinolestris* Grootaert and Meuffels are active on beaches with small pebbles mixed with sandy patches. It is not found on rocks, pure sandy beaches nor mangroves.

Holotype male and paratypes were reported from Khao Lak and Nang Thong, Phang Nga province, Thailand.

## Genus Thambemyia Oldroyd, 1956

The *Thambemyia* Oldroyd belongs to the subfamily Hydrophorinae of Dolichopodidae. It was proposed by Oldroyd for one species (*T. pagdeni* Oldroyd, 1956) occurring in Malaysia (Penang). This genus was revised by Masunaga et al. (2005). To date, there are 18 more species of the genus known, which are distributed in Japan (16 species), Taiwan (one species) and Hawaii (one species). *T. borealis* (Takagi, 1965), known from Hokkaido and Kuril Island, was probably introduced to North America by human-assisted dispersal (Pollet et al. 2004). The species of the genus are marine, and found foraging in the splash zone (Sunose and Sato, 1994; Grootaert and Meuffels, 2001). On Thailand's sea coast one species was recorded (*Thambemyia pagdeni*) from Phang Nga and Trad Provinces. Furthermore, this scenario did not make them surprised (Grootaert and Meuffels, 2001), due to this region's location close to Pinang island.

# Diagnostic characters of the genus Thambemyia Oldroyd, 1956

These flies are ranged in size from 3.2 - 6.0 mm and considered as a medium to large marine Dolichopodidae. Dark and stout, it is generally recognized from its body. Verex a little wide, not concave. A pair of strong orbitals present on frons above antennae; postocular bristles strong. First flagellomere elongate with short pubescence and small rigid bristles; arista apical, short, bare. Palpus and proboscis sometimes

elongated. Mesonotum with irregular black spots. Scutellum with two pairs of strong bristles. It is always strongly modified in males. Wing usually hyaline, indistinctly tinged greyish, sometimes brown apically in males.

**Distribution**: Rocky shore of the Andaman Sea (Thailand, Malaysia), Indian Ocean (Sumatra), Gulf of Thailand. According to Grootaert and Meuffels (2001) who stated that some specimens were collected from Phang Nga (Southern, peninsular Thailand) and Trad province (Eastern, Thailand).

#### Genus Cymatopus Kertész, 1901

Cymatopus Kertész is a marine genus of flies that are found on rocky shores. The genus is very heterogeneous with several species groups. Eighteen species are momentarily assigned to it and are mostly confined to the Australasian and Oriental regions. Four species from the eastern Pacific and Caribbean regions should probably be removed from Cymatopus (Grootaert and Meuffels, 1993). Cymatopus capensis Parent, 1939 from South Africa was placed in synonymy to Aphrosylus griseatus Curran, 1926 by Dyte and Smith (1980), becoming later the type species of the new genus Cemocarus Meuffels and Grootaert, 1984. Meuffels and Grootaert (1984) and Grootaert and Meuffels (1993) revised the Australasian species. Additionally, Evenhuis and Grootaert (2002) provided some new distributional records for the Oriental region (Singapore). Furthermore, Evenhuis (2005) described an additional four species (i.e., C. baravikai, n. sp. (Fiji), C. flavipes, n. sp. (New Caledonia), C. neocaledonicus, n. sp. (New Caledonia), and C. othniopteryx, n. sp (East Timor)) from Fiji and their neighbouring areas, bringing the total number of species worldwide to fourteen. Interestingly, in Thailand, three species of the genus Cymatopus were observed by Grootaert and Meuffels in 2001. They are Cymatopus longipilus, C. malayensis and C. thaicus.

# Diagnostic characters of Genus Cymatopus Kertész, 1901

*Cymatopus* Kertész represented in Thailand by three species groups: a species group with simple fore-legs, but with long haired hind legs (*C. longipilus*), and two species groups with heavily ornamented fore-legs: *C. malayensis* belongs to a group of larger species where the male has dense whiskers and notched wings with fields of microtrichia; *Cymatopus thaicus* belongs to a group of smaller species with the usual postocular hairs and simple wings (Grootaert and Meuffels, 2001).

**Distribution**: Rocky shores of Andaman Sea (Thailand, Malaysia). These marine Dolichopodids are widely distributed in Thailand from the southern part (Khao Lak, and Nang Thong, Phang Nga Province) until the western part (Koh Chang, Hat Sai Khao, Trad Province and Koh Samet, Ao Tawan, Rayong Province).

The distribution zone of the Thai marine long-legged fly is manifestable in terms of plotting map (Figure 6). Where a dozen new species are discovered every year (Bickel, 2009). Over the last century, the Dolichopodidae has been variously divided into subfamilies both regionally and at a world scale (Aldrich, 1905; Becker, 1917, 1918; Robinson, 1970; Ulrich, 1981; Negrobov, 1986; Yang et al., 2006; Sinclair et al., 2008; Pollet and Brooks, 2008; Lim et al., 2010, Germann et al., 2011).

Although, there have been a few experiments and investigations on marine longlegged flies in peninsular Thailand (Grootaert and Meuffels, 2001) and this brought to the high latest recorded number for Thai marine long-legged flies to fifteen species, in particular the Gulf of Thailand Sea such as Chumphon, Surat Thani, Nakhon Si Thammarat, Songkhla, Pattani and including Narathiwas Provinces were not found and recorded of a single species. In addition, the increasing of the number of species from the latest studies may convince Thai dolichopodid fly taxonomists to pay more attention to this interesting group especially the marine species, but nonetheless there are no researchers still concerned. This could be explained by several reasons hidden behind this problem. For instance, marine long-legged flies are difficult in identification and observation, because of their minute size, taxonomic key is literally out of date. Moreover, long-legged fly taxonomic study is notably deficient and needs to be studied further in every context such as biodiversity and monitoring, biogeography, ecology, evolution, and also a good taxonomy or systematic study.

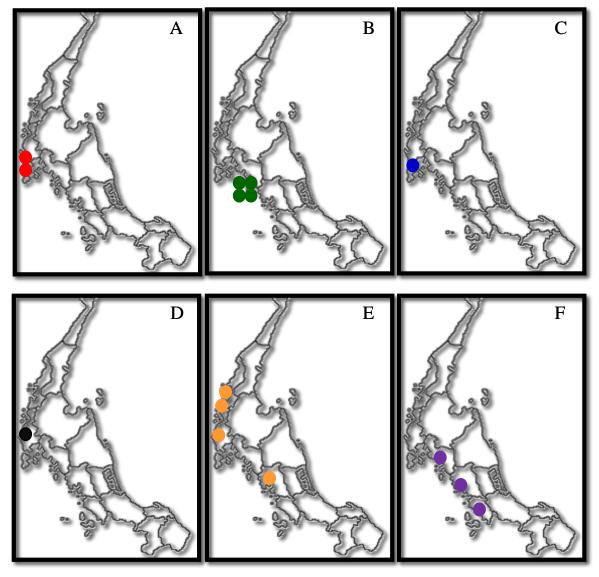
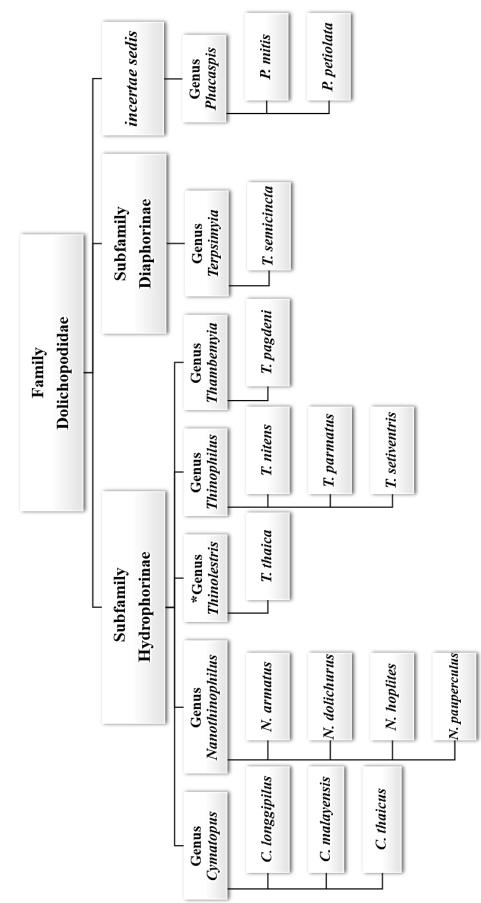
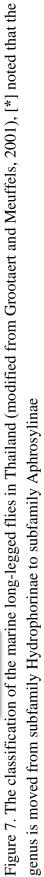


Figure 6. Localities of the marine long-legged flies in Thailand, consisting of the genus *Cymatopus* Kertész in red circles; composed *C. thaicus, C. malayensis* (A). For the genus *Nanothinophilus* Grootaert and Meuffels shows in green circles (B); contained *N. armatus, N. pauperculus, N. dolichurus*, and *N. hoplites*. The genus *Thambemyia* Oldroyd exhibits in blue circle with one member is *T. pagedni* (C). While the black circle shows an endemic species, *Thinolestris thaica* (D). Then, the genus *Phacaspis* Grootaert and Meuffels (E), composed *P. petiolata* and *P. mitis* (orange circles). Further purple circles represent the genus *Thinophilus* Wahlberg, including *T. nitens, T. parmatus, T. setiventris* (F).





Regarding the most updated information of marine long-legged fly fauna in Thailand (Grootaert and Meuffels, 2001) principally conducted upon the basic survey and mainly provided an annotated checklist in lower number of species (fifteen species) and very out date of species identification key, when compared to a small neighbouring country such as Singapore (with 44 known species, recorded in Evenhuis and Grootaert, 2002), due to a lack of basic knowledge on this particular fauna in various aspects, for this reason, this study will be taken in order to increase the understanding of basic knowledge and provide further information such as taxonomic status, species and genetic diversity, distribution patterns, habitat preferences, and also species identification using traditional and modern ways of taxonomic study of marine long-legged flies in this region.

# 2. QUESTIONS AND OBJECTIVES

#### **2.1) QUESTIONS**

2.1.1) What is the species composition of marine long-legged flies (Diptera: Dolichopodidae) in peninsular Thailand?

2.1.2) Are there congruent species number using external morphology based and molecular based identification of marine long-legged flies (Diptera: Dolichopodidae) with reference to genus *Ngirhaphium* Evenhuis and Grootaert and genus *Hercostomus* Loew in peninsular Thailand?

2.1.3) Are there common patterns of divergence in marine long-legged flies (Diptera: Dolichopodidae) with reference to genus *Ngirhaphium* Evenhuis and Grootaert and genus *Hercostomus* Loew in peninsular Thailand?

# **2.2) OBJECTIVES**

This thesis desires to review, identify and understand the distribution pattern, genetic diversity, and explicit phylogeny of marine long-legged flies of peninsular Thailand using traditional and modern approaches of current taxonomic study (integrative taxonomic point of view). Specifically, this study aimed to:

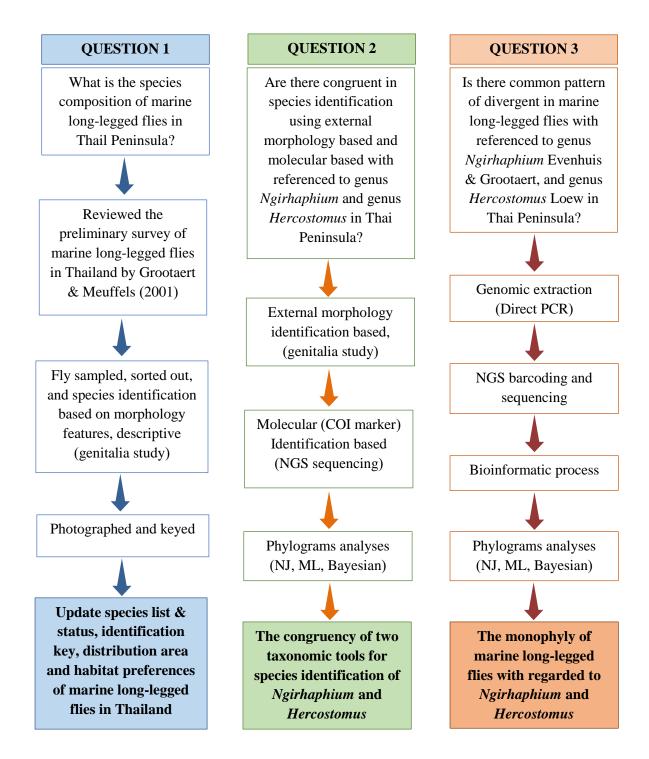
2.2.1) Update, describe, nomenclature and provide a regional key of marine long-legged flies in Thailand.

2.2.2) Integrate the morphological and molecular evidences in delimiting species.

2.2.3) Understand the genetic diversity and distribution pattern of marine longlegged flies in peninsular Thailand using Next Generation Sequencing (NGS) technique.

# 3. RESULT AND DISCUSSION

The overall results and discussion are provided in this part. Full details including review of literatures, methodologies, detailed results and discussion can be found in the attached Manuscript 1, 2 and published Papers 1, 2. Furthermore, research questions, brief methodology and expected outcomes are also summarized in the diagram below.



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#### **3.1 Research Question 1**

*Question 1: What is the species composition of marine long-legged flies (Diptera: Dolichopodidae) in peninsular Thailand?* 

## 3.1.1) Species Composition and Taxonomic Status

The marine long-legged flies were randomly collected for the period of seven months (November 2014 until May 2015) from several mangroves, beaches, tide pools, and rocky shores of the Andaman Sea (Indian Ocean) and the Gulf of Thailand (South China Sea), southern Thailand. Malaise traps, yellow pan traps, visual searching and sweep netting techniques were adopted to collect the fresh specimens from the fields. In order to identify species in this habitat, external morphology characters, especially male genital features were mostly investigated. The result of the present study revealed that with 3,870 of marine long-legged fly specimens which were collected from Thai Peninsula, interestingly, 60 morphospecies, 23 genera, seven subfamilies and including one unplaced subfamily were recognised (Table 9, full detailed in manuscript 1, 2 and published paper 1, 2). Of the 60 morphospecies, seventeen new species were identified and one new genus Ornamenta siamese sp. nov. gen nov. (incertae sedis) (Figure 8), will sooner be given a new scientific name, illustrated, described and keyed. Twenty seven new record species were recorded for the country. This could be concluded that this survey was drastically increased (account for 79.49% or four time expanded) in terms of species number, new species, new genus, and new record when compared to the previous report by Grootaert and Meuffels (2001). Besides, further result also divulged that genus Thinophilus Wahlberg was considered as the richest in species number in this region (see full detailed in manuscript 1 and published paper 2). This result literally supports the suggestion of the previously surveyed marine long-legged flies (Diptera: Dolichopodidae) in Thailand since nearly two decades ago by Grootaert and Meuffels (2001), they mentioned that many species of marine long-legged flies in Thailand were awaiting discovery, description and illustration. Beyond this point, they also found that mangrove Thinophilus Wahlberg were the most diverse genus among them and mostly in concordance with Singapore species (in annotated checklist of dolichopodid flies from Singapore), recorded by Evenhuis and Grootaert (2002), with five new species revealed. This further suggested that this genus was needed to revise species limits and geographical distribution.



Figure 8. Male habitus, *Ornamenta siamese* sp. nov., gen nov., collected from a mangrove at Ban Bakan Toh Thid, Langu district, Satun province (Andaman Sea, Indian Ocean)

Table 9. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach.

Taxa	Status	Distribution Area		Habitat Preference
		Andaman	Gulf of	
		Sea	Thailand	
Diaphorinae				
Asyndetus Loew, 1869				
1) Asyndetus aciliatus	-	+	-	SB
2) Asyndetus thaicus	-	+	-	SB
3) Asyndetus sp.1	New species	+	-	SB
Chrysotus Meigen, 1824		1	[	
4) Chrysotus dot	New record	+	+	М
Diaphorus Meigen, 1824	1	ſ	r	
5) <i>Diaphorus</i> sp.1	New species	+	+	М
Dolichopodinae				
Argyrochlamys Lamb, 1922				
6) Argyrochlamys impudicus	New record	+	+	RB,SB
Hercostomus Loew, 1857				
7) Hercostomus brevicornis	New record	+	+	М
8) Hercostomus brevidigitalis	New record	+	-	М
9) Hercostomus lanceolatus	New record	+	+	М
10) Hercostomus obtusus	New record	+	-	М
11) Hercostomus plumatus	New record	+	+	М
12) Hercostomus propermeieri	New species	+	-	М
Lichtwardtia Enderlin, 1921		I	L	
13) Lichtwardtia ziczac	New record	-	+	М
Paraclius Loew, 1864				
14) Paraclius adligatus	New record	+	+	М
15) Paraclius asiobates	New record	-	+	М
16) Paraclius digitatus	New record	+	+	М
17) Paraclius obtus	New record	-	+	М
18) Paraclius serratus	New record	+	+	М
19) Paraclius singaporensis	New record	-	+	М
20) Paraclius sp. nov.	New species	-	+	М
Tachytrechus Haliday, 1851	I I I I I	I	<u> </u>	<u> </u>
21) Tachytrechus tessellatus	New record	+	+	SB

Table 9. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach (cont.).

Taxa	Status	Distribution Area		Habitat Preference
		Andaman	Gulf of	
		Sea	Thailand	
Phoomyia Naglis and Grootaert, 200	3			
22) Phoomyia singaporensis	New record	+	+	RB,SB
23) Phoomyia talumpuk sp. nov.	New species	-	+	RB,SB
Hydrophorinae				
Cymatopus Kertész, 1901				
24) Cymatopus malayensis	-	+	+	RS
25) Cymatopus thaicus	-	+	-	RS,SB
26) <i>Cymatopus mayakunae</i> sp. nov.	New species	+	-	RS
Nanothinophilus Grootaert and Meu	ffels, 1998			
27) Nanothinophilus hoplites	-	+	-	M.SB
28) Nanothinophilus pauperculus	-	+	-	M,SB
Thambemyia Oldroyd, 1956				
29) Thambemyia pagdeni	-	+	+	RB,SB
Thinophilus Wahlberg, 1844				
30) Thinophilus apicatus	New record	+	+	М
31) Thinophilus boonrotpongi	New species	+	+	М
32) Thinophilus chaetulosus	New record	-	+	М
33) Thinophilus langkawensis	New species	+	-	M,SB
34) Thinophilus melanomerus	New record	+	-	М
35) Thinophilus minutus	New species	+	+	М
36) Thinophilus parmatoides	New species	-	+	М
37) Thinophilus parmatus	-	+	-	М
38) Thinophilus parvulus	New species	-	+	М
39) Thinophilus sp. nov	New species	+	-	М
40) Thinophilus simplex	New record	+	+	М
41) Thinophilus spinatoides	New species	+	-	М
42) Thinophilus spinatus	New species	+	-	М
43) Thinophilus superbus	New record	+	-	М
44) Thinophilus variabilis	New species	+	+	М
45) Thinophilus yeoi	New record	-	+	М

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Table 9. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach (cont.).

Taxa	Status	Distribution Area		Habitat Preference
		Andaman	Gulf of	
		Sea	Thailand	
Parathalassiinae				
Microphorella Becker, 1909				
46) Microphorella malaysiana	-	+	+	RB,SB
Rhaphiinae				
Ngirhaphium Evenhuis and Grootaer	t, 2002			
47) Ngirhahium caeruleum	New record	_	+	М
48) Ngirhahium chutamasae	New species	+	-	М
49) Ngirhahium meieri sp. nov.	New species	+	_	М
50) Ngirhahium murphyi	New record	+	-	М
51) Ngirhahium sivasothii	New record	+	-	М
Sciapodinae				
Amblypsilopus Bigot, 1859				
52) Amblypsilopus abruptum	_	-	+	М
Chrysosoma Guerin-Meneville, 1831	_			
53) Chrysosoma leucopogon	-	-	+	М
Sympycninae				
Chaetogonopteron de' Meijere, 1914				
54) Chaetogonopteron chaeturum	-	+	+	М
55) Chaetogonopteron vexillum	_	+	+	М
Sympycnus Loew 1857				
56) Sympycnus sp.	New record	-	+	М
Teuchophorus Loew, 1857				
57) Teuchophorus krabiensis	-	+	-	М
Incertae Sedis (Unplaced Group)				
Ornamenta gen. nov.				
58) Ornamenta siamese sp. nov.	New species	+	-	М
Phacaspis Meuffels and Grootaert, 1	990			L
59) Phacaspis mitis	_	+	+	М
Terpsimyia Becker, 1922				L
60) Terpsimyia semicincta	-	-	+	М
7 subfamilies, 1 unplaced group				

#### **3.1.2) Species Distribution**

Over 3,800 specimens of marine long-legged flies in Thai Peninsula were investigated and identified based on male genital and non-genital morphological features scrutiny. It was found that the west coast of Thai Peninsula (Andaman Sea, Indian Ocean) contained higher number of species (23 morphospecies) of marine long-legged flies than the Gulf of Thailand Sea (South China Sea, Pacific Ocean) (15 morphospecies). In other words, marine long-legged flies in Thai Peninsula were greater in number of species in the Andaman seacoasts than Gulf of Thailand seacoasts, especially in mangrove habitat (Table 9, Manuscript 1). This study confirms previously surveyed report of marine longlegged flies in Thailand by Grootaert and Meuffels (2001). These authors identified thirteen morphospecies from Andaman Sea side, whereas only six species were observed from Gulf of Thailand. This could be hypothesized that the larger size and complexity of mangrove in Andaman Sea might support a greater number or variety of marine longlegged fly species than the Gulf of Thailand. According to the report of Department of Marine and Coastal Resources (DMCR) in 2009 (Marine knowledge management subcommittee, Aquatic resources research institute, Chulalongkorn University, 2017), it was clearly shown that Andaman seacoasts (1,104,892.87 RAI) contained larger mangrove size than Gulf of Thailand Sea (182,934.01 RAI). Moreover, the result also notably supports the theory of island biogeography of organisms proposed by MacAthur and Wilson (1967), that the larger island may support more number of organismal species on the island.

The richest genera of marine long-legged flies in Thailand elucidated were *Thinophilus* Wahlberg (16 species) and *Paraclius* Loew (7 species), but the distribution pattern and number of specimen are confusing and uninteresting. Because several of the species from both genera were low in number of individual or specimens. For example, *Thinophilus parvulus* sp. nov., only a pair of them that could be captured from Pattani mangrove only, and one more problematic species *Thinophilus spinatus* sp. nov. which was mainly swept from Ban Bakan Toh Thid, Langu district, Satun province also provided little number of individual, when *Paraclius adligatus*, was solely sampled from Ban Khao Than mangrove, Tha Chang district, Surat Thani province. Unluckily, only five female specimens of this species could be trapped by Malaise trap and hand collection. However, one of the most striking results in the context of distribution pattern of the

present study showed that Hercostomus lanceolatus belonging to Hercostomus Loew, 1857 was the largest range of distribution. It was widely distributed in several mangroves from the two sides of Thai Peninsula (Table 9, manuscript 1). For example, in the Gulf of Thailand Sea, Hercostomus Loew occurred abundantly in several mangroves in Pattani province such as replanted mangrove very near sea front at the Prince of Songkla University, Pattani campus; a clumped mangrove at Ban Dato, Yaring district, Pattani province; one back mangrove in Ban Na Thab, Chana district, Songkhla province; and also largely dwelled in disturbed mangroves at Ban Khao Than, Tha Chang district, Surat Thani province; and abundantly observed from Ban Phanangtak, Muang district, Chumphon province. For the Andaman Sea, this species was obviously observed from Tammalang mangrove, Muang district, Satun province; and also sampled from Ban Bang Nai Si and Ban Bang Dong mangroves, Takuapa district, Phang Nga province (Figure 9, circled with red colour). Whereas the genus Ngirhaphium Evenhuis and Grootaert remarkably occurred in different way and provides an interesting data in term of distribution pattern, with majority of species in this genus mostly restricted to particular mangroves. For instance, N. chutamasae sp.nov. is only occurred in Tammalang mangrove, while *N. caeruleum* occurred in mudflats of Surat Thani and Chumphon bays, and a new species N. meieri sp.nov. (pending for description), was restricted to a back mangrove with unique environmental condition at Ban Bang Dong, Takuapa district, Phang Nga province (Figure 10). Due to these facts, it could be concluded that those mentioned species are notably interesting in the context of species identification, distribution, and understanding of the genetic diversity and variation. Further results also elucidated that most species of marine long-legged flies occurring in Thailand are largely congruent to the Singaporean species than the other countries in Southeast Asia (SEA), with more than 37 species conforming to type (http://evolution.science.nus.edu.sg/ MIP.html), especially the species that were specifically collected from the Gulf of Thailand Sea. This could be simplifying this natural event by the locality of geography. If Singapore country and the border of the sea water were taken into account, it could be said that both the Gulf of Thailand and Singapore lies in the same side of the South China Sea (Figure 11), across Malay peninsula and without any natural barrier to limit the dispersal ability of this flies from place to place. This is the reason why most species are similar between the two countries.

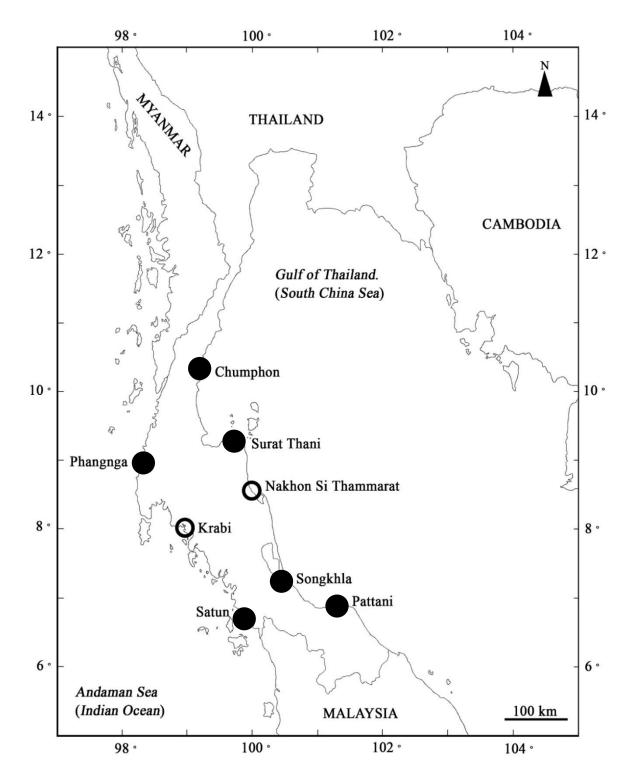


Figure 9. Distribution map of *Hercostomus lanceolatus* Zhang, Yang and Grootaert in Thailand ( note that is the localities of distribution)

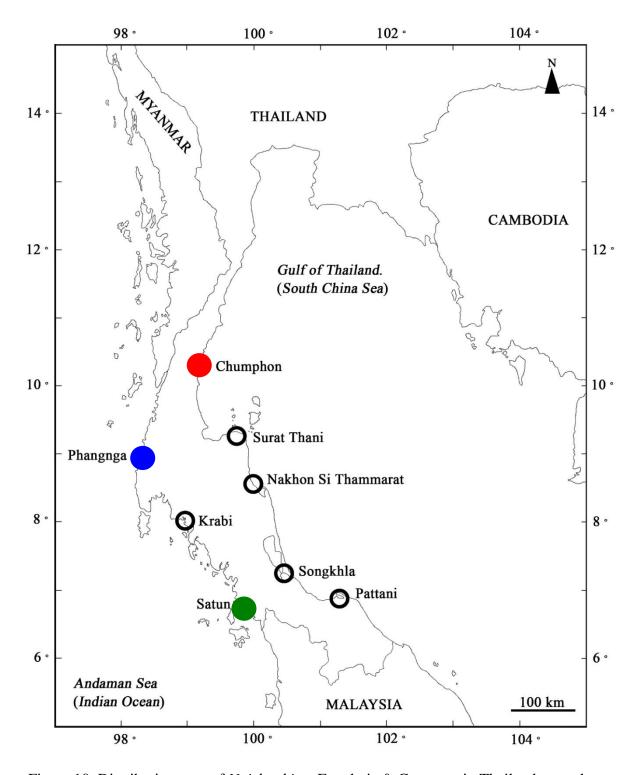


Figure 10. Distribution map of *Ngirhaphium* Evenhuis & Grootaert in Thailand, note that *N. caeruleum*, *N. chutamasae*, *N. meieri*, *N. murphyi*, *N. sivasothii* 

However, due to lack of many available information or no survey from other neighbouring countries such as Malaysia, Brunei, Indonesia, Myanmar, Cambodia, Philippines, and Vietnam (Figure 9) which were also recognised as countries that are endowed with marine habitats, it may provide poor and not precise data of this group of flies in the context of species distribution. Nonetheless, this survey is an evidence to support the marine zoogeographical distribution in this region.



Figure 11. Map of Thailand and Singapore, red pins indicate the countries.

#### **3.1.3) Habitat Preference**

In various available publications (Miall, 1934, Cregan, 1941; Dytes, 1959; Pollet, 2000, 2001; Grootaert and Meuffels, 2004; Brooks, 2005; Ulrich, 2005; Grootaert, 2006a) on natural history of long-legged flies in the world, it was reported that both larvae and adult of long-legged flies abundantly occurred in moist and humid habitats such as in saltmarshes, seashores, lakes, streams, canals, mangroves, rocky shores, humid rocky and sandy beaches, tide pools, waterfalls, freshwater seepages, damn soil, humid forests, swamps (Dytes, 1959; Pollet, 2001; Brooks, 2005; Ulrich, 2005, Grootaert, 2006a), and also occurred in drier habitats such as agricultural fields, grasslands, and urban gardens (Books, 2005). As can be seen (Table 9, Figure 12, and Manuscript 1). The result of habitat preferences of the current study indicated that mangrove habitat composed of the highest number of species and was assumed to be major marine habitats that could support a large number of species. In addition, this finding has led to conclusion that the most preferred habitat by marine long-legged flies in Thailand is mangrove. This could be explained by the very moist environment in containing more complexity of microhabitat than other marine habitats such as rocky and sandy beaches, rocky shores, or even tide pools. If take all those marine habitats (mangroves, rocky shores, sand and rocky beaches, tide pools) into account, and found that mangrove largely goes along with basic information on natural history of long-legged flies that many of the species largely prefer moist environments, and of course mangrove show merely fitted to the definition of high humid and moist atmospheres comparing to other marine habitats such as rocky shores, sandy-rocky beaches, tide pools. Moreover, it also contains various kinds of microhabitats and assumed that it allowed and supported species of marine long-legged flies into this habitat. Similarly, previous study by Grootaert and Meuffels (2001) has suggested that each marine species has their own favored habitats. For example, Terpsimyia semicincta, virtually lives in mangrove mudflats with high exposure to sunlight in the Gulf of Thailand sea side which is generally influenced by sea level of the day Grootaert and Meuffels (2001).

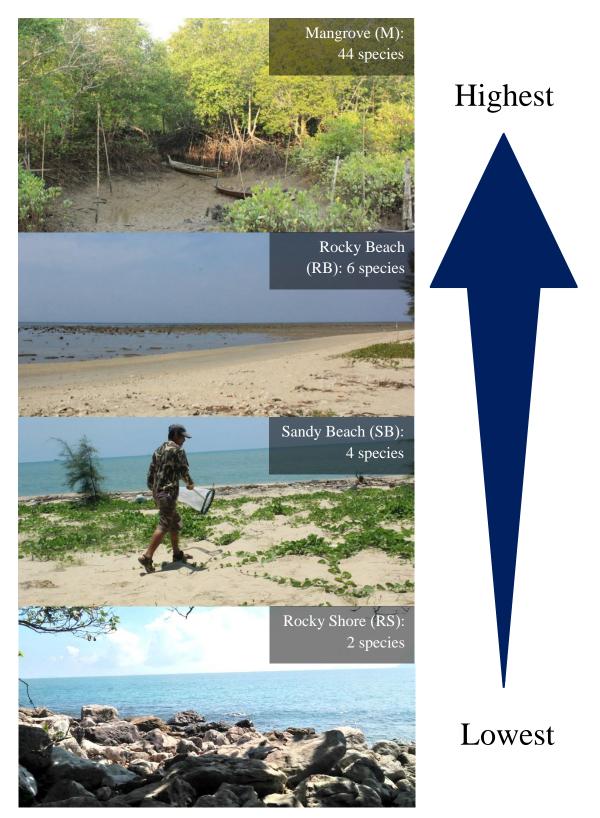


Figure 12. Marine habitat preferences of marine long-legged flies in Thai Peninsula, southern Thailand.

#### 3.2 Research Question 2 And 3

Question 2: Are there congruent species number using external morphology based and molecular based identification of marine long-legged flies (Diptera: Dolichopodidae) with referenced to genus Ngirhaphium Evenhuis and Grootaert, and genus Hercostomus Loew in Thai Peninsula?

*Question 3: Are there common patterns of divergent in marine long-legged flies (Diptera: Dolichopodidae) with referenced to genus Ngirhaphium Evenhuis and Grootaert, and genus Hercostomus Loew in Thai Peninsula?* 

## 3.2.1) Species Identification and Monophyly of marine long-legged flies

The monophyly of long-legged fly family is evident (Wiegmann et al., 1993; Collins and Wiegmann, 2002; Brooks, 2005; Zhang and Yang, 2005; Sinclair and Cumming, 2006; Wang et al., 2007a; Lim et. al., 2010; Germann et al., 2011). According to Pollet et al. (2010) tested the hypothesis of the monophyly of the European Dolichopodinae systematics based on 1,702 mitochondrial DNA characters and genital and non-genital morphological features, and in the end of their study confirmed that there was notably monophyletic relationships in the subfamily Dolichopodinae. In Singapore, based on Lim et al. (2010) study, they used six genes of mitochondrial and nuclear genes to assess the phylogenetic relationships of long-legged flies, and found that there were monophyletic relationships, and further suggested that this fly undergoes rapid speciation.

Current study is the first study from Thailand with reference to two genera of mangrove species group, *Ngirhaphium* Evenhuis and Grootaert and *Hercostomus* Loew. This study employed two taxononic tools such as external morphology features and COI gene. One of the most striking findings from this section was that both taxonomic tools provided a congruent identification in the contexts of species number and the monophyly of mangrove *Ngirhaphium* Evenhuis and Grootaert and *Hercostomus* Loew as follows:

#### 3.2.1.1) Genus Ngirhaphium Evenhuis and Grootaert

## **3.2.1.1.1)** External morphological features based identification

In overall, the author sorted out nearly 3,900 individuals of fresh marine longlegged fly specimens from peninsular Thailand and then identified up to 60 species, belonging to 23 genera, 7 subfamilies and one unplaced subfamily through the use of an external morphology characters, while 74 MOTUs or molecular taxonomic unit (from 1,200 representative flies, 924 succeeded sequences) obtained from DNA-based analysis. Moreover, the results indicated that five species of genus *Ngirhaphium* Evenhuis and Grootaert from peninsular Thailand were recognized by external morphological features as follows:

# SYSTEMATIC ACCOUNT

# Family DOLICHOPODIDAE Subfamily RHAPHIINAE

# Genus Ngirhaphium Evenhuis and Grootaert, 2002

*Ngirhaphium* Evenhuis & Grootaert, 2002: 310. Type species by original designation: *Ngirhaphium murphyi* Evenhuis & Grootaert, 2002.

**Remarks and Diagnosis.** Medium to large sized species (4.5–8 mm) with a metallic green or blue ground colour. Antenna very long in males, a little shorter in females. Arista apical, basal article long. Rostrum in male small with well-developed labellae. Large rostrum in female Vertex excavated (cf. Sciapodinae).

**Mid and hind coxae** without exterior bristle. Femora with inconspicuous bristling. All tibiae with strong bristles. Fore leg in male with tarsomere 4 bearing an asymmetrical, apical dorsal forked protuberance (absent in females); terminal segment with a pair of normal claws and a thickened claw-like structure beneath the posterior claw. Females with the claws as usual, but the terminal segment bears a long dorsal protuberance. Mid and hind legs with tarsomeres 1–4 with an apical comb of spinules ventrally.

Wing with tip of  $M_{1+2}$  sharply bent upwards just before reaching the wing border and ending near tip of  $R_{4+5}$ .

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# Ngirhaphium chutamasae Samoh and Grootaert

(Figures 13) (New species)

**Diagnosis.** A large species differing from the other *Ngirhaphium* species mainly in the structure of the male genitalia. Cercus in lateral view slightly shorter than dorsal surstylus. Cercus brown, tip pointed bearing a single yellow bristle. Dorsal surstylus brown, bordered with short, stout yellow bristles. Outer branch of apical fork on the fore tarsomere 4 slightly longer than inner branch.  $M_{1+2}$  with a short stub on apical bend.

## Material examined.

HOLOTYPE ♂, labelled: "THAILAND: Satun prov., Tammalang (6°32'21.05"N, 100°04'9.42"E), 3.x.2014 (reg. 34030, leg. P. Grootaert)<sup>,</sup> (PSU); PARATYPE:1 ♂, Tammalang (6°32'21.05"N, 100°04'9.42"E), 6.viii.2014 (leg. A. Samoh) (RBINS). **Etymology.** The species is dedicated to Associate Professor Dr. Chutamas Satasook, director of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkhla University, Hat Yai as a token for her dynamic support of our research.

Description. Male. Length body: 7 mm; wing: 5.6 mm.

**Head**. Frons shining metallic green (not dusted). Face greenish brown in ground-colour; apex of face and clypeus yellowish brown in ground-colour, wide, nearly as wide as front of frons, parallel-sided, grey dusted with very short clypeus (less than 0.1 length of face). Eyes pass beyond border of face; eyes densely set with white hairs. Ocellar callus globular protruding from frons with 2 very long ocellars, directed backward, divergent. Vertical bristles long, half as long as ocellars, rather anteriad on frons at level of ocellar callus, close to eye border, long, black, directed forward and cruciate. Pair of long black postverticals directed backward and crossing. Postoculars above strong, black in single row, below white and mixed with very long white hairs below mouth; postcranium greenish in ground-colour but grey dusted. Palpus long, strap-shaped, yellowish with few short black hairs, no bristles. Labella brown with black hairs.



Figure 13. *Ngirhaphium chutamasae* sp. nov., male habitus; inset: apical tarsomeres on fore leg, showing the large fork-like extensions on tarsomere 4 and the additional claw-like structure on tarsomere 5 (photo: J. Brecko). Scale = 1 mm (Samoh et al., 2015)

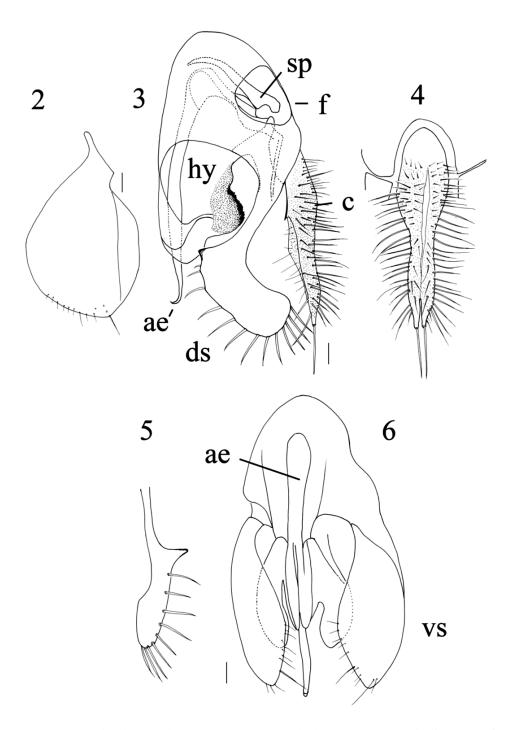


Figure 14. 2–6. *Ngirhaphium chutamasae* sp. nov., holotype male genitalia: 2. Left ventral surstylus; 3. Lateral view of genital capsule with left ventral surstylus removed; 4. Cerci dorsally; 5.Dorsal surstylus in ventral view; 6.Ventral view of genital capsule. Abbreviations: ae: aedeagus; c: cercus; ds: dorsal surstylus; f: foramen; hy: hypandrium lobe; sp: sperm pump; vs: ventral surstylus. Scale = 0.1 mm.

**Antenna** very long, completely black. First segment long, 3X as long as second segment; second segment short, apically with crown of short black bristles. Third segment very long strap-shaped, laterally flattened, about 6X as long as width at base. Arista apical, apical article longer than basal article, gradually tapered towards tip. Length of scape: 0.52; pedicel: 0.13; postpedicel: 0.95; basal aristal article: 0.34; apical aristal article: 0.4 (in mm).

**Thorax** and scutellum dark metallic green in ground-colour (bluish when seen from in front), covered with fine grey dusting. All hairs and bristles black. Pleura more densely grey dusted than mesonotum. Acrostichals biseriate, about 7 pairs, rows widening slightly behind. Presutural dorsocentrals multiseriate; 6 postsutural dorsocentrals: 4 short and 2 long prescutellars; 1 pair of strong scutellars. One long humeral with shorter bristle in front; 1 strong posthumeral, 2 strong notopleurals, 1 postsutural, 1 supraalar, 1 very strong postalar. Propleural bristles black, 6 short upper and 2 longer lower propleural bristles (lower one twice as long as upper).

Legs (Figure. 12) yellow, all bristles black. All coxae greenish black in ground-colour, covered with fine greyish dusting. All trochanters brown. Tip of hind tibia annulated brown at tip. All tarsi yellowish, becoming darker towards tip. Apical tarsomeres completely black. Foreleg. Coxa with short black bristles. Fore femur slightly swollen on basal half; row of minute posteroventrals in apical half. Short preapical posterior bristle and 1 stronger anterior preapical bristle directed forward. Fore tibia with 4 strong ad, 4 strong pd and crown of 4 apicals. All tarsal segments densely set with black hairs and short black bristles. Tarsomere 4 with dorsal asymmetrical fork, extended over tarsomere 5; fork about 1.5X length of tarsomere; outer branch of fork slightly longer than inner branch, tips pointed (Figure. 12, inset). Terminal segment with pair of long normal claws and thicker claw-like structure beneath posterior claw. Two well-developed pulvilli and empodium present. Length of femur, tibia and tarsal segments (in mm): 2: 1.96: 0.98: 0.56: 0.28: 0.28: 0.28. Mid leg. Coxa with short bristles anteriorly; no exterior bristle. Mid femur as wide as fore femur; ventrally with inconspicuous bristles; 1 strong anterior preapical and 2 tiny posterior preapicals. Tibia with 6 ad, 6 pd (might be considered as dorsal), 8 longer av and crown of long apicals. Tarsomeres 1–4 ventrally at tip with pair of short spine-like bristles as well as comb of shorter black spinules. Apical tarsomere dorsally thickly set with long black squamiform bristles. Length of femur, tibia and tarsal segments (in mm): 2: 3.08: 1.68: 0.84: 0.77: 0.35: 0.42. Hind leg. Coxa bare. Hind femur thicker than mid femur, as wide as fore femur; ventrally almost bare; 1 strong anterior preapical, 1 fine posterior preapical. Tibia stronger bristled than mid tibia with 7 long av, 7 ad, 7 pd and crown of long apicals. Tarsomeres 1–4 ventrally at tip with pair of short spine-like bristles as well as comb of shorter black spinules. Length of femur, tibia and tarsal segments (in mm): 2: 3.78: 1.68: 0.98: 0.77: 0.42: 0.35.

**Wing** mostly tinged brownish, but anteriorly between costa and  $R_{4+5}$  with yellowish brownish tinge. Tp brown seamed. Veins dark brown, yellowish at base.  $M_{1+2}$  sharply bent upwards and ending in costa closely near tip of  $R_{4+5}$ . Tp straight, about as long as apical part of  $M_{3+4}$ . Anal vein reaching wing border. Halter with white knob. Squama white with long white cilia.

**Abdomen** shiny dark metallic green; tips and sides of tergites with greyish dusting. Sternites greyish dusted. Tergites densely set with quite long black bristles; hind-marginal bristles slightly longer than other bristles. Only tergite 5 with very long marginal bristles. Sternites with very short hairs except for longer marginals on sternite 4. Genital capsule black. Cercus brown, slightly shorter than dorsal surstylus (Figure 14).Ventrally at base with black sclerotisation. Both cerci fused for almost entire length, only tips free (Figure 14). Tip of cercus pointed, with single yellow apical bristle, dorsally set with long black bristles. Dorsal surstyli brown, much enlarged, forming clasper transverse on cercus, bordered with short, stout yellow bristles, shorter than surstylus is wide (Figure 14). Ventrally with dark spur-like apex (Figure 14). Ventral and dorsal surstyli not fused.Ventral surstylus yellowish, large, rounded (Figure 14); tip with short hair-like bristles; inner carina running parallel to dorsal border. Hypandrium dorsally with large rounded black protuberance set with spinules (Figure 14).

Female. Indistinguishable from females of N. sivasothii

**Remarks.** The new species is morphologically almost identical to *N. sivasothii*, except for the larger forked extension on fore tarsomere 4 in male and the very different male genitalia. The outer branch of the apical fork on fore tarsomere 4 is slightly longer than the inner branch; the outer branch is slightly shorter than the inner branch in *N. sivasothii*. The wing is brownish tinged and only the Tp is dark seamed. In *N. sivasothii* the wing is darker and the longitudinal veins as well as Tp are generally black seamed. Vein  $M_{1+2}$  includes a short stub on the apical bend in the new species. Such a stub was never

observed in the large populations of *N. sivasothii* in Singapore. Presently females of the new species are indistinguishable from females of *N. sivasothii*. Both species have been collected together. For this reason, no females were included in the material examined section.

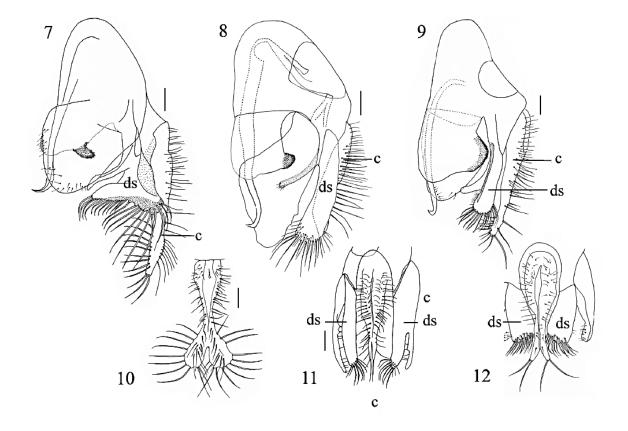


Figure 15. 7–12. Lateral view genital capsule: 7. *Ngirhaphium sivasothii*; 8. *N. caeruleum*; 9. *N. murphyi*; dorsal view cerci: 10. *N. sivasothii*; 11. *N. caeruleum*; 12. *N. murphyi*. Abbreviations: c: cercus, ds: dorsal surstylus (modified after Grootaert and Puniamoorthy, 2014). Scale = 0.1 mm.

## *Ngirhaphium caeruleum* Grootaert & Puniamoorthy, 2014 (New record)

**Materials Examined.** THAILAND:  $2 \stackrel{\circ}{\circ}, 3 \stackrel{\circ}{\circ}$ , Chumphon province, Muang, Ban Paknam (N 10°30'28.7', E 99°14'29.8'), 11.viii.2015;  $1 \stackrel{\circ}{\circ}, 3 \stackrel{\circ}{\circ}$ , Surat Thani province, Chaiya, Phumriang (9°23'34.0"N 99°15'24.0"E)

**Diagnosis.** A larger species with mesonotum and tergites shining metallic blue. Apical aristal segment thin (filiform) and longer than basal aristal segment. Male with dorsaland ventral surstylus at right side fused; separated at left side.

Description. Male: Body length: 7 mm; wing length: 5.6 mm

**Head:** Frons shining dark metallic blue (not dusted), sunken between the eyes, wide in front becoming wider behind; ocellar callus small, raised above frons. Face wide, as wide as front of frons, parallel-sided, silvery dusted with a very narrow clypeus (hardly a tenth of length of face). Eyes pass beyond the border of the face; eyes densely set with silvery hairs.

**Thorax:** Thorax and scutellum with a shining dark metallic bluish ground-colour, covered with a fi ne grey dusting. A black stripe outside each acr-row. All hairs and bristles black. Pleura more densely grey dusted than mesonotum. Acr biseriate, about 7 pairs, the rows widening a little behind. Presutural dc multiseriate; 6 post-sutural dc: 4 short and 2 long prescutellars. All propleural bristles black, 4 short upper and 2 longer black lower propleural bristles (lower one twice as long as upper).

**Legs:** Yellow, all bristles black. All coxae greenish black in ground-colour, covered with a fine greyish dusting. All trochanters pale brown. Tip of mid and hind tibiae darkened at tip. All tarsi brownish yellow, becoming darker towards tip. Terminal segments completely black.

**Wing:** hyaline, faintly tinged brownish, but anteriorly between costa and  $R_{4+5}$  with a yellowish tinge. Membrane along veins  $R_{4+5}$ ,  $M_{1+2}$ ,  $M_{3+4}$  and Tp sometimes brown to black seamed. Veins dark brown.  $M_{1+2}$  sharply bent upwards and ending in costa closely near tip of  $R_{4+5}$ . Tp straight, a little longer than apical part of  $M_{3+4}$ . Anal vein well developed. Halter with white knob. Squama white with long white cilia.

**Abdomen:** Shining dark metallic bluish; tips and sides of tergites with a greyish dusting. Sternites greyish dusted. Tergites densely set with quite long, black bristles; hind marginal bristles a little longer than the other bristles. Only tergite 5<sup>th</sup> with very long marginal bristles. Sternites with very short hairs except for the longer marginals on sternite 4<sup>th</sup>. Hypopygium yellowish brown sessile with tip of cerci hidden in sternite 4<sup>th</sup>. Cercus shorter than dorsal surstylus. Apex of cercus slender with a long bristle. Tip of dorsal surstylus set with a bundle of yellow bristles.

**Remarks.** This large species that author discovered from Cumphon and Surat Thani Province is a bit green compare to type species from Singapore.

## *Ngirhaphium murphyi* Evenhuis & Grootaert, 2002 (New record) (Figure 16)

*Ngirhaphium murphyi* Evenhuis & Grootaert, 2002: 310. Type locality: SINGAPORE: Kranji mangrove. *N. murphyi*: Grootaert & Puniamoorthy, 2014: 147

**Diagnosis.** A large species (5.7–7.3 mm), generally with clear wings. Mesonotum and tergites metallic green. Apical aristal article quite thick, nearly half as long as basal aristal article. Male with cerci longer than surstyli andnthus the tips are visible outside the surstyli

**Materials examined.** THAILAND: 1 ♂, 4 ♀, Satun province, Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 11.viii.2014 (leg. A. Samoh) (PSU)

**Remarks.** A large species (5.7–7.3 mm), generally with clear wings. Mesonotum and tergites metallic green. Apical aristal article quite thick, nearly half as long as basal aristal article. Male with cerci longer than surstyli and thus the tips are visible outside the surstyli

Distribution. Satun, Krabi, and Phang Nga Province (Andaman Sea, Indian Ocean)



Figure 16. Male habitus, Ngirhaphium murphyi Evenhuis and Grootaert, 2002

## Ngirhaphium sivasothii Grootaert & Puniamoorthy, 2014

(New record) (Figure 17)

*Ngirhaphium sivasothii* Grootaert & Puniamoorthy, 2014: 150. Type locality: SINGAPORE: Semakau Island.

**Diagnosis.** A medium-sized species (4.5–5.5 mm), generally with dark infuscate wing and with longitudinal veins and Tp (posterior cross vein) brownish seamed. Mesonotum and tergites metallic green. Apical aristal article shorter, but nearly as long as basal article. Male with dorsal surstylus half as long as cerci, with a rectangular bend, set with very long bristles (Figure 15-7). Cercus much longer than dorsal surstylus, tip wide, rounded, set with many long yellow bristles (Figure 15-10). Outer branch of apical fork on the fore tarsomere 4 slightly shorter than inner branch.

Material examined. THAILAND: Satun province: 1♂, 2♀, Tammalang (6°32'21.05" N, 100°04'9.42" E); 4 ♂ 7♀, 6.viii.2014 (leg. A. Samoh); 1♂, 2♀, 3.x.2014 (reg. 34030, leg. P. Grootaert & A. Samoh); 7 ♂, 20 ♀, Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 12.viii.2014 (leg. A. Samoh); 2♂, 1♀, Tanjong Po (6°36'57.43" N, 99°57'25.66" E), 3.x.2014 (leg. A. Samoh) (PSU).

**Remarks.** Some specimens had quite clear wings without the brown of black seams along the longitudinal veins and the Tp (posterior cross vein).

Distribution. Satun, Krabi, and Phang Nga Province (Andaman Sea, Indian Ocean)



Figure 17. Male habitus, Ngirhaphium sivasothii Grootaert and Puniamoorthy, 2014

### Ngirhaphium meieri sp. nov.

## (Figure 18)

**Diagnosis.** A large species differing from the other *Ngirhaphium* species mainly in the structure of the male genitalia. Cercus in lateral view slightly shorter than dorsal surstylus (Figure 19). Cercus brown, tip pointed bearing a single yellow bristle. Dorsal surstylus brown, bordered with short, stout yellow bristles. Outer branch of apical fork on the fore tarsomere 4 slightly longer than inner branch.  $M_{1+2}$  with a short stub on apical bend (Figure 18).

**Material examined.** HOLOTYPE ♂, labelled: THAILAND: Phang Nga Province, Takuapa, Bang Yai, (8°54'27.5"N, 98°23'59.6"E), sweep netting, 9 February 2015.

**Etymology.** The species is dedicated to Prof. Dr. Rudolf Meier, head of Evolutionary Biology Laboratory, Department of Biological Sciences, at the National University of Singapore (NUS) who provides the authur a great opportunity to visit and study fly's taxonomy by using molecular techniques.

**Distribution.** Phang Nga Province (Andaman Sea, Indian Ocean)

## Key to species of male Ngirhaphium Evenhuis and Grootaert, 2002

1)	Mesonotum and tergites metallic blue. Antenna with apical aristal article filiform and
	generally longer than basal article (Singapore). Genitalia as in Figures 15-11
	N. caeruleum Grootaert & Puniamoorthy
-	Mesonotum and tergites mainly metallic green. Antenna with apical aristal article
	shorter or about half as long as apical article2
2)	Cerci in lateral view nearly as long as dorsal surstyli (Figures 14)
	N. chutamasae sp. nov.
-	Cerci in lateral view longer than dorsal surstyli (Figures 15-7, 15-9)3
3)	Dorsal surstylus elongate digitiform with truncate apex (Figures 15-9, 15-12) slightly
	shorter than cercus. Cerci with narrow apex, set with 2 apical setae (Figure 15-
	12)N. murphyi Evenhuis & Grootaert
-	Dorsal surstylus with very wide apex (Figure 15-7) much shorter than cercus. Cerci
	with expanded apex set with many yellow setae (Figure 15-10)



Figure 18. *Ngirhaphium meieri* sp. nov., male habitus: apical tarsomeres on fore leg, showing the large fork-like extensions on tarsomere 4 and the additional claw-like structure on tarsomere 5. Scale = 1 mm.

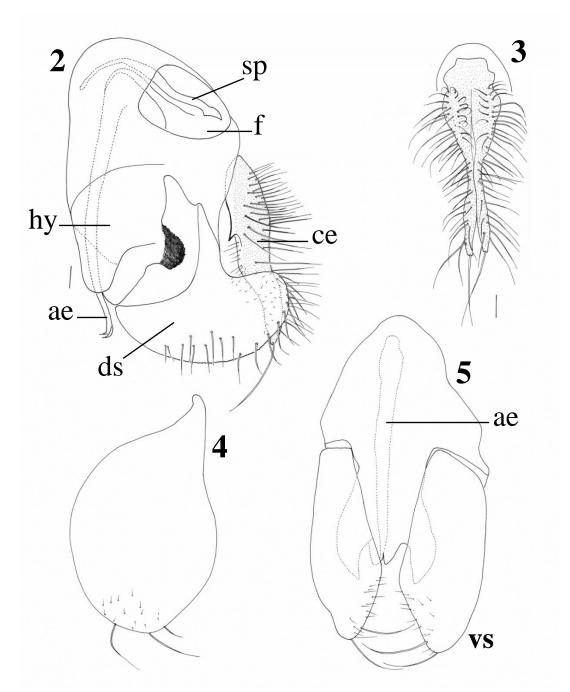


Figure 19. 2–5. *Ngirhaphium meieri* sp. nov., holotype male genitalia: 2. Lateral view of genital capsule with left ventral surstylus removed; 3. Cerci dorsally; 4. Left ventral surstylus; 5. Ventral view of genital capsule. Abbreviations: ae: aedeagus; c: cercus; ds: dorsal surstylus; f: foramen; hy: hypandrium; sp: sperm pump; vs: ventral surstylus. Scale = 0.1 mm.

#### 3.2.1.2) Molecular (COI) Based Identification

## • Neighbour-Joining (NJ) and Maximum Likelihoods (ML)

Regarding to molecular phylogenetic (COI gene) analyses—based on Neighbour Joining (NJ) (Figure 20) and Maximum Likelihoods (ML) methods (Figure 21), overall and with variable support, the close relationships between *Ngirhaphium chutamasae*, *N. caeruleum* (12% genetic distance, with 100 bootstrap support), and *N. sivasothii* sp. nov. (11% genetic distance, with 100 bootstrap support), *N. murphyi*, and *N. chutamasae* sp. nov. (Figure 20).

To illustrate, in Neighbour-Joining (NJ) phylogenetic tree (COI gene) showed three distinct clades (Figure 20) or recognised as monophyletic relationships: 1<sup>st</sup> clade composed with one single species, namely, *N. sivasothii*. 2<sup>nd</sup> clade consisted two species, for instance, *N. chutamasae*, and *N. murphyi*. 3<sup>rd</sup> clade contained only one species such *N. caeruleum*. Depending on this analysis, *N. chutamasae* sp. nov. is literally clustered closer to *N. murphyi*. Moreover, both species can be separated into two species on the basis of the 94% bootstrap support, with 7% genetic distances (Figure 20) which might lead to the conclusion that *N. chutamasae* sp. nov. must be considered as a new species for this genus.

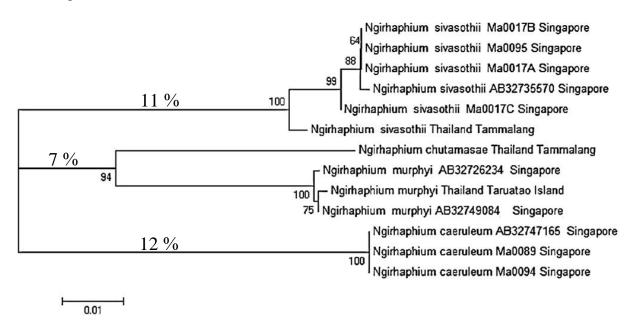
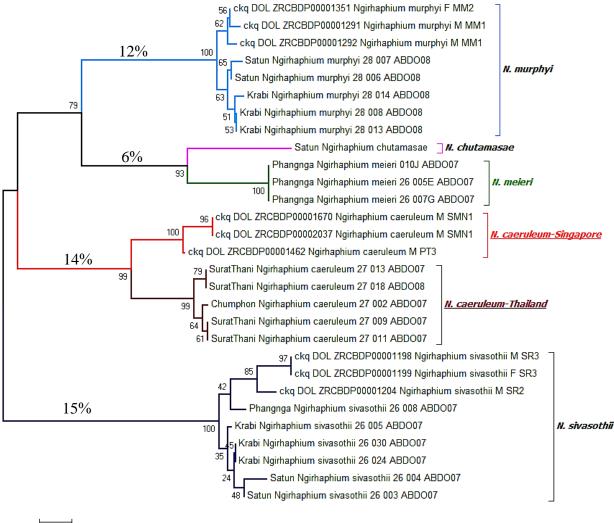


Figure 20. Neighbour-Joining (NJ) tree of the COI barcodes of *Ngirhaphium* Evenhuis and Grootaert with bootstrap values indicated at the nodes. Scale of genetic distance is 1%.

In Maximum Likelihoods (ML) phylogenetic tree was showed in Figure 21. This tree terminology clearly revealed the monophyletic group of *Ngirhaphium chutamasae* sp. nov. and *N. merei* sp. nov (6% genetic distances, 93% bootstrap values). But inspite of that both *N. chutamasae* sp. nov. and *N. meieri* sp nov. was nested with *N. murphyi* (12% genetic distances, 79% bootstrap values), related to *N. caeruleum* in 14% genetic distances with 100% bootstrap support, separated from *N. sivasothii* by 15% genetic distances with 100% bootstrap support (Figure 19).



0.0100

Figure 21. Phylogenetic tree of *Ngirhaphium* Evenhuis and Grootaert used Maximum Likelihood method analysis in MEGA 7.

From the above results (external morphology based identification vs COI based identification), it can be said that both taxonomic tools provided the same answer in term of species number. In general, Ngirhaphium Evenhuis and Grootaert is firstly collected from Singapore mangroves by Evenhuis and Grootaert (2002), but it is the first record for Thailand. Evenhuis and Grootaert (2002) considered N.murphyi differrent relative featuring amount of external morphological characters and noticed as synapomorphous to a species, Rhaphium longicornae (Fallén) which belongs to genus Rhaphium Meigen. However, it can be easily separated from its closely related genus Rhaphium Meigen (Evenhuis and Grootaert, 2002) by these following features and information. In genus Rhaphium, the basal aristal segment is shorter than apical segment (Figure 22), and veins M<sub>1+2</sub> and R<sub>4+5</sub> often slightly converging (Grichanov et al., 2011) (Figure 23A, 23B). There is mainly Holarctic genus (Grichanov et al., 2011; Negrobov et al., 2011, 2012) but also widely distributed in Central Asia such as South Kazakhstan, Kyrgyzstan, Tajikistan, Uzbekistan, and expected to occurred in Iran (Kazerani et al., 2013; Negrobov et al., 2013a, 2013b), and were also found in Sri Lanka (Naglis and Grootaert, 2011), and China (Tang et al., 2016b), except Australasian region (Yang et al., 2006), range from 1.5 to 5.7 mm (Kazerani et al, 2013).

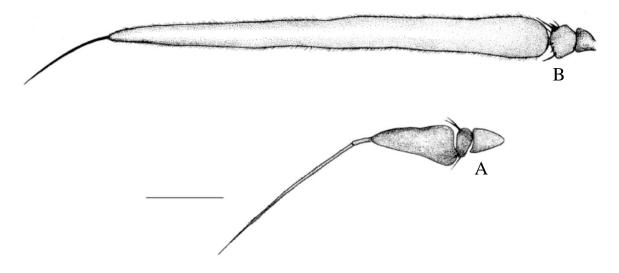


Figure 22. *Rhaphium apophysatum* (A), Male antenna; *Rhaphium bilobum* (B), Male antenna. Scale bar = 0.2 mm (Tang et al., 2016b)

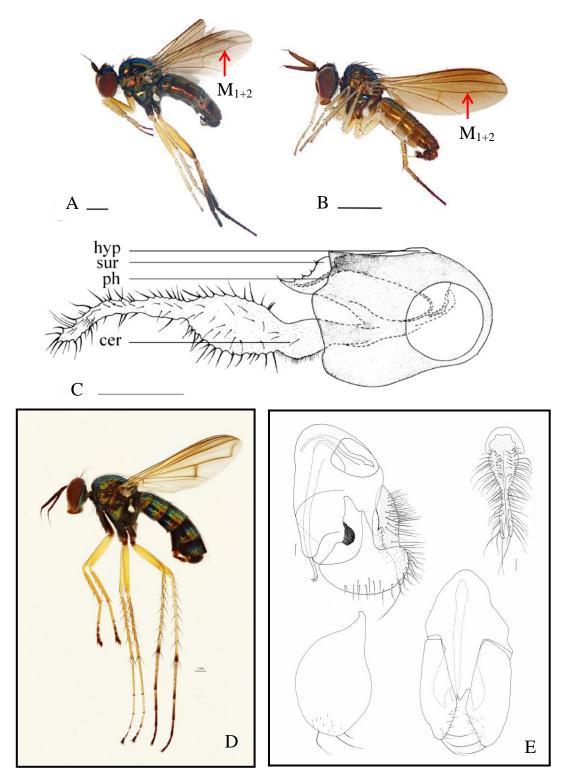


Figure 23. *Rhaphium dorsiseta* (A), *R. neimengense* (B), Male habitus and Wing veins, Scale bar = 0.1 mm; *R. apophysatum* (C), Male genitalia, lateral view, Scale bar = 0.2 mm (Tang et al., 2016b), Male habitus, *N. meieri* sp. nov. (D), Male genitalia (E).

Whereas, in genus Ngirhaphium Evenhuis and Grootaert, is medium sizes (5-7 mm) mangrove long-legged flies with metallic green grounded colour (see also Figure 13, 16, 17, 18)(Evenhuis and Grootaert, 2002), the basal part is longer than apical segment (Evenhuis and Grootaert, 2002; Grootaert and Puniamoorthy, 2014; Samoh et al., 2015) (Figure 23-E), wing vein, the tip of  $M_{1+2}$  is sharply bent upword and ending near vein  $R_{4+5}$  (Figure 23-D) (Evenhuis and Grootaert, 2002). Regarding to the result of this study, five species of mangrove Ngirhaphium Evenhuis and Grootaert have been recognised from Thai Peninsula using traditional way of identification. There were Ngirhaphium caeruleum, N. chutamasae sp. nov., N. meieri sp. nov., N. murphyi, and N. sivasothii. Surprisingly, the results from the modern way of species identification by using a remarkably good mitochodrial DNA marker such as COI also elucidated that five species of mangrove *Ngirhaphium* as well as identified by using external morphology based. This can be concluded that both external morphology features based and COI based identification are completely congruent (Figures 20, 21, 24). Further results also revealed that Ngirhaphium Evenhuis and Grootaert in Thailand was monophyletic relationships based on molecular analysis (COI gene, Neighbour-Joining) (Figure 20). Similarly, a notable revision of mangrove Ngirhaphium Evenhuis and Grootaert by Grootaert and Puniamoorthy (2014), they also recognised three species (including N. caeruleum, *N.murphyi*, *N. sivasothii*) belong to this genus from several mangroves in Singapore used traditional and modern ways of species identification, and the final results revealed that the number of species of genus Ngirhaphium Evenhuis and Grootaert from Singapore mangroves were completely congruent (N. caeruleum is different from its closely related N. murphyi and N. sivasothii described as two new species by 11%, 12% genetic distances respectively) and monophyletic relationships or shared in common for genetic data. In addition, they further mentioned that all three recognised species from Singapore mangroves were easily distinguished based upon the male genital features.

Further COI analysis using Maximm Likelihoods (ML) method showed that two new species, *Ngirhaphium chutamasae* sp. nov. and *N. meieri* sp. nov. were closely related in term of genetic distance. Interestingly, with reference to external morphology based identification, both species are the most identical except a shape of male dorsal surstylus and circus (Figure 21).

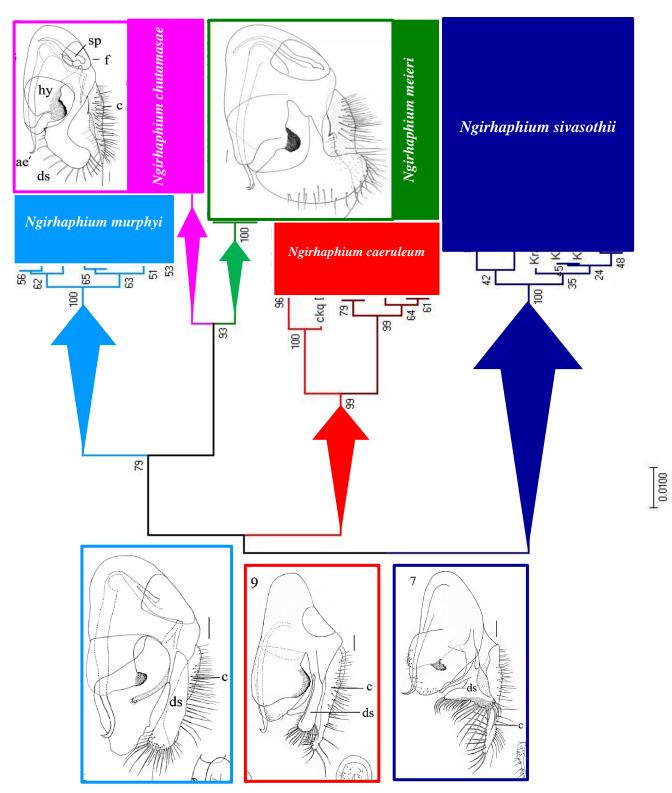


Figure 24. *Ngirhaphium* Evenhuis and Grootaert phylogenetic tree based on Maximum Likelihood (ML) method analysis.

In *N. chutamasae* sp. nov., dorsal surstylus is boot-like in shaped (Figure 25- B) and as equal as cercus is long, whereas *N. meieri* sp. nov., contains mushroom-like dorsal surstylus and a bit shorter than dorsal surstylus (Figure 25-A). It is suggested that using external morphology to assess the species of mangrove *Ngirhaphium* Evenhuis and Grootaert in Thailand is usefull and lead to high success. Similarly, in the contexts of distribution pattern and habitat preference theses are completely differrent and might be sufficiently good for primary consideration and recognition. In *N. chutamasae* sp. nov., solely occurred in small creeks near front sea at Tammalang mangrove, Satun province (Figure 25-A, Figure 11, 26, 27), while *N. meieri* sp. nov., strictly found distributed in back mangrove at Bang Nai Si, Takuapa, Phangnga province (Figure 25-B, Figure 18, 26, 28).

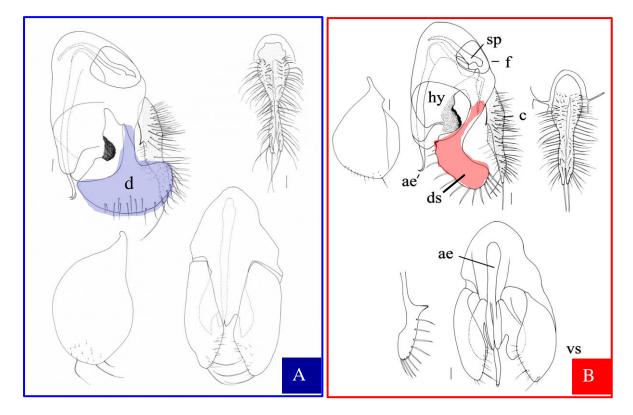


Figure 25. Comparative morphology of dorsal surstylus (ds) shape between two new species of *Ngirhaphium* Evenhuis and Grootaert from Thailand, *N. meieri* sp. nov. (A), and *N. chutamasae* sp. nov. (B)

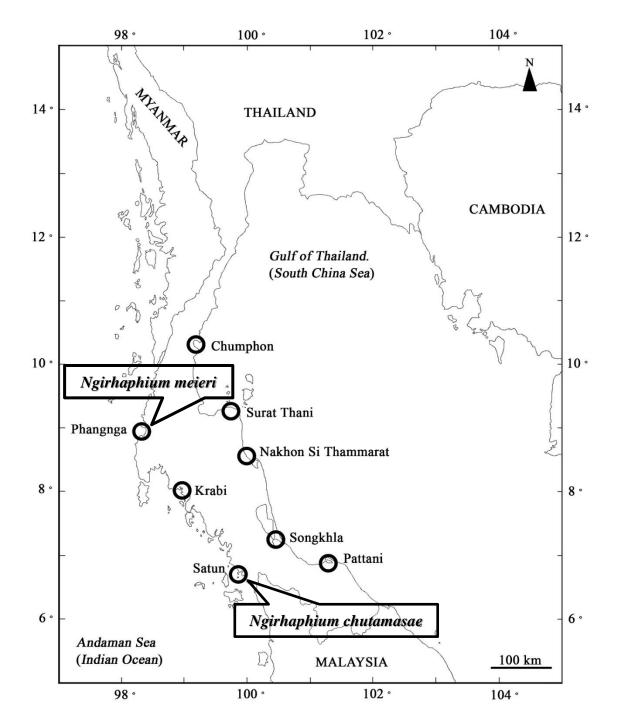


Figure 26. Distribution areas of two new species of *Ngirhaphium* Evenhuis and Grootaert, note that blue indicate the distributed area (Bang Nai Si, Phangnga province) of *N. meieri* sp. nov. and red indicate the distributed area (Tammalang, Satun) of *N. chutamasae* sp. nov.



Figure 27. Habitat preference of *Ngirhaphium chutamasae* sp. nov. At Tammalang mangrove research station, Muang district, Satun province



Figure 28. Back mangrove in Bang Nai Si, Takuapa district, Phangnga Province

Although, this genus is a new member and providing a little information for dolichopodid fly family, but in term of taxonomic arragment of this monotypic genus, it seems to requires more attention. Genus *Ngirhaphium* has been classified into the subfamily Rhaphiinae by Evenhuis and Grootaert (2002), they explained that it is due to their external morphological characters which have remarkable resemblance to a member of long-legged flies genus *Rhaphium* Meigen, 1803, *Rhaphium longicorne* Fallen. However, an exciting molecular phylogenetic data that was provided by Lim et al. (2010) based on six genes could not put this genus among other dolichopodid subfamilies and has remain as unplaced subfamily.

# 3.2.2) Genus Hercostomus Loew 1857

## 3.2.2.1) External morphological features based identification

#### Systematic Account

Subfamily Dolichopodinae Genus *Hercostomus* Loew, 1857 (New record)

### Type genus: Dolichopus Latreille, 1796

Hercostomus Loew, 1857: 9. Type species: Sybistroma longiventris Loew

**Remarks.** Genus *Hercostomus* sensu lato, as defined as, a polyphyletic assemblage of species, related to *Dolichopus*, *Parahercostomus*, and *Poeccilobotrus* (Brooks, 2005), with typical wing vein  $R_{4+5}$  and M gently bent anteriorly beyond crossvein dm-cu. In addition, this genus retained all the major external morphological characters of the subfamily Dolichopodinae with pteropleuron without hairs i.e. no hairs in front of the posterior thoracic spiracle. Fifth pair of dc not or rarely slightly convergent. Male clypleus not bulging, lower margin usually straight and not reaching lower eyes margin, scape and pedicel well developed (Figure 29); arista simple and short with developed pubescent.

Apparently, this group of flies is still largely debatable in term of taxonomic arrangement. Especially one recognized species from Southeast Asian countries, namely, *Steleopyga* (Grootaert and Meuffels, 2001b) as described is a separate genus, by indication of some characters (the possession of a cluster of spines on sternite 8 and one preapical anteroventral row of 4 setae on the hind femur, and the complexes of male genitalia – forming elements of entangled asymmetrical lobes, and Brooks (2005) suggested that to be a homologous with the condition of *Hercostomus longiventris* lineages that is a part of the "*Hercostomus complex*".

In addition, Yang et al. (2006) reported that these flies contained nearly 270 species from the Oriental region and are mostly collected from non-marine habitats. However, some species (seven new species) are also recorded from marine habitat such as mangroves in Singapore (Zhang et. al. 2008). Unfortunately, this genus was never ever recorded from Thailand. This study is the first of it kind (recording with seven known species) from peninsular Thailand.

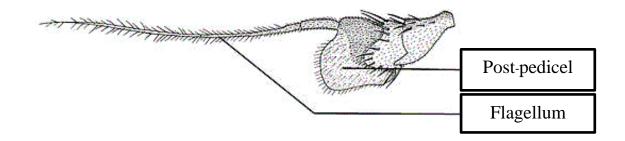


Figure 29. Male antenna of *Hercostomus lanceolatus* (modified from Zhang et al., 2008)

# *Hercostomus brevicornis*, Zhang, Yang and Grootaert (New record) (Figure 30, 31)

**Material Examined**.  $4^{\circ}_{\circ}1^{\circ}_{\circ}$ ; Thailand, Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh;  $5^{\circ}_{\circ}$ ; Laemson Kamphaeng, Satun, (6°56'27.9"N 99°42'12.4"E), sweep netting, 4 May 2015;  $7^{\circ}_{\circ}4^{\circ}_{\circ}$ ; Tammalang, Muang, Satun, (6°32'21.7"N 100°04'09.3"E), sweep netting, 2 May 2015;  $6^{\circ}_{\circ}1^{\circ}_{\circ}$ ; Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh;  $4^{\circ}_{\circ}2^{\circ}_{\circ}$ ; Bang Yai, Bang Nai Si, Takuapa district, Phang Nga, (8°54'27.5"N

98°23'59.6"E), 9 February 2015, coll. A. Samoh; 6∂4♀; Khlong Phon, Khlong Thom, Krabi, (7°48'11.2"N 99°10'11.9"E), sweep netting, 13 May 2015, coll. A. Samoh.

**Remarks**. Body length 2.9–3.1 mm, wing length 2.7–2.8 mm. All coxae yellow, but mid coxa at most pale brownish with a narrow black anterior stripe. Cercus strongly curved, nearly geniculate. Aedeagus with 2 small, inner denticles (Figure 30B).

Distribution. Satun, Phangnga, Krabi, and Surat Thani

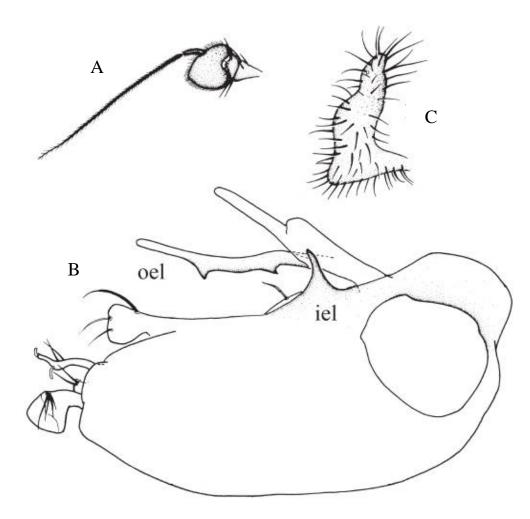


Figure 30. *Hercostomus brevicornis*, Male: A, antenna; B, male genitalia; C, cercus; ile, inner epandreal lobe; oel, outer epandreal lobe (modified from Zhang et al., 2008)



Figure 31. Male habitus, Hercostomus brevicornis Zhang, Yang and Grootaert

## *Hercostomus brevidigitalis*, Zhang, Yang and Grootaert (New record) (Figure 32, 33)

**Material Examined**. Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh;  $2\sqrt[3]{1}$ ; Ban Ramard, Khlong Thom, Krabi, (7°42'17.4"N 99°03'48.4"E), sweep netting, 26 April 2015;  $3\sqrt[3]{1}$ ; Ban Bang Yai, Takuapa, Phang Nga, (8°54'27.5"N 98°23'59.6"E), sweep netting, 11 February 2015, coll. A. Samoh. **Remarks**. This is a small to medium species (body length 3.2–3.6 mm, wing length 3.2–3.5 mm). First flagellomere elongate, 2.3 times as long as wide (Figure 32A). All coxae black; femora black except tip of fore and mid femora. Squama yellow with black hairs. Male genitalia long, reaching thorax.

Distribution. Satun, krabi, and Phang Nga.

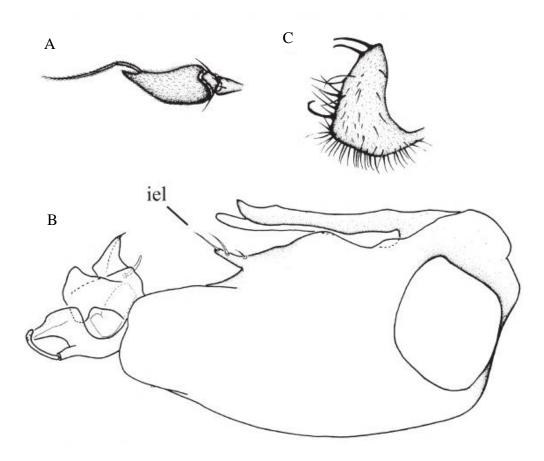


Figure 32. *Hercostomus brevidigitalis*, male: A, antenna; B, male genitalia; C, cercus. iel, inner epandreal lobe (modified from Zhang et al., 2008)



Figure 33. Male habitus, Hercostomus beridigitalis Zhang, Yang and Grootaert

## *Hercostomus lanceolatus*, Zhang, Yang and Grootaert, 2008 (New record) (Figure 34, 35)

## Material Examined.

## THAILAND.

 $9\sqrt[3]}142$ ; Ban Khao Than, Tha Chang district, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh;  $1\sqrt[3]{3}2$ ; Tammalang, Muang, Satun, (6°32'21.7"N 100°04'09.3"E), sweep netting, 2 May 2015, coll. A. Samoh;  $2\sqrt[3]{1}2$ ; Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh;  $2\sqrt[3]{7}2$ ; Prince of Songkla University (PSU), Muang, Pattani, (6°53'04.9"N 101°14'10.1"E), sweep netting and Malaise trap, 11 April 2015, coll. A. Samoh;  $3\sqrt[3]{4}2$ ; Ban Dato, Yaring, Pattani, (6°55'17.1"N 101°19'50.7"E), sweep netting, 14 April 2015, coll. A. Samoh; Pak Phanang Tawantok, Pak Phanang, Nakhon Si Thammarat, (8°22'30.2"N 100°10'00.4"E), sweep netting, 1 May 2015, coll. A. Samoh;  $4\sqrt[3]{3}2$ ; Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), sweep neeting, 17 February 2015, coll. A. Samoh;  $4\sqrt[3]{2}2$ ; Ban Hua Khao, Singha Nakhon, Songkhla, (7°12'03.6"N 100°34'36.8"E), sweep netting, 27 May 2015, coll. A. Samoh.

**Remarks**. Body length 3.7 mm, wing length 3.3 mm. Wing with wide, yellowish brown to grey stigma behind tip of  $R_1$ , stigma reaching the level of thickening of  $R_{4+5}$ .  $R_{4+5}$  thickened from basal quarter, but narrow again before reaching wing border (Figure 34B, and 35).

**Distribution**. Satun, Chumphon, Surat Thani, Nakhon Si Thammarat, Songkhla, Pattani, and Singapore

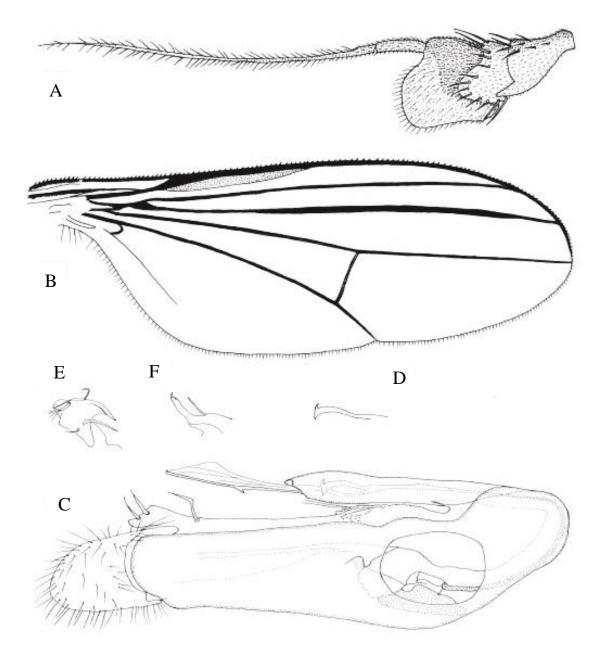


Figure 34. *Hercostomus lanceolatus*, male habitus, (A) wing, (B) antenna, (C) male genitalia, (D) claw-like aedeagus, (E) ventral lobe of surstyli, (F) dorsal lobe of surstyli (modified from Zhang et al., 2008)



Figure 35. Male habitus, Hercostomus lanceolatus Zhang, Yang and Grootaert

# *Hercostomus plumatus* Zhang et al., 2008 (New record) (Figure 36, 37)

**Remarks**. Body length 2.4–2.7 mm, wing length 2.2–2.5 mm. First flagellomere 1.2 times as long as wide (Figure 36A). Fore coxa yellow, mid and hind coxae brownish. Dorsal lobe of surstyli with plumose hair (Figure 36D).

**Distribution**. Nakhon Si Thammarat, Surat Thani, Songkhla (Gulf of Thailand), Krabi Province (Andaman Sea)

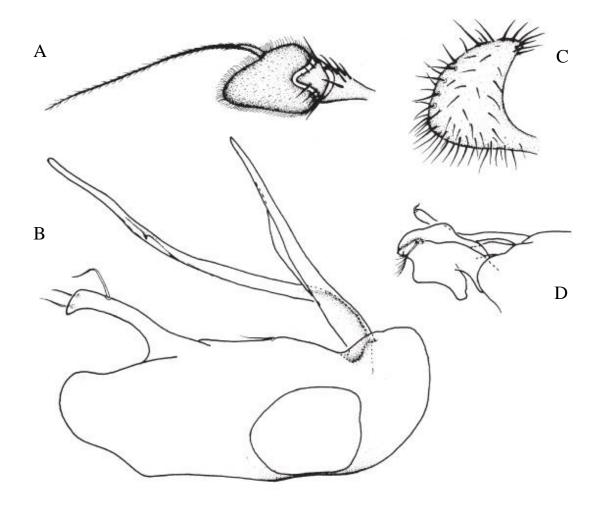


Figure 36. *Hercostommus plumatus*, male habitus; antenna (A); male genitalia (B); cercus (C); surstyli (D) (modified from Zhang et al., 2008)



Figure 37. Male habitus, Hercostomus plumatus Zhang, Yang and Grootaert

#### Hercostomus obtusus sp. nov.

(New record, pending for description by Grootaert)

**Material Examined.**  $3\stackrel{\circ}{}2\stackrel{\circ}{};$  Ban Pakbara, Langu District, Satun Province, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh

**Remarks.** Body length 2.3–2.5 mm, wing length 2.0–2.2 mm. First flagellomere 2.5 times as long as wide. Fore coxa yellow with a little black line at tip, mid and hind coxae brown. In facts, this species was first collected from Singapore mangroves and Grootaert suggested to be a new species.

Distribution. Pakbara and Bakan Toh Thid, Langu, Satun (Andaman Sea)

## Hercostomus propermeieri sp. nov.

(New species, pending for description) (Figure 38)

**Material Examined.**  $2 \stackrel{\wedge}{} 2 \stackrel{\circ}{}$ ; Ban Laemson, Kamphaeng District, Satun Province, (6°56'27.9"N 99°42'12.4"E), sweep netting, 4 May 2015, coll. A. Samoh.

**Remarks.** Body length 2.4–2.5 mm, wing length 2.1–2.2 mm. First flagellomere 3.0 times as long as wide. Fore coxa yellow, mid and hind coxae brown.

Distribution. Ban Laem Son, Kam Phaeng, Langu, Satun (Andaman Sea)



Figure 38. Male habitus, *Hercostomus propermeieri* sp. nov.

#### **Systematic Account**

# Key to species of mangrove *Hercostomus sensu lato* from Thailand (based on male habitus)

1) Wing, with brown colour (sometimes is pale) stigma at the end of vein  $R_1$ ; noticed that vein R<sub>4+5</sub> relatively thickened from the basal quarter onwards (Figure 34B, 35).....2 2) Stigma elongate, exposing beyond start of the thickening of vein R<sub>4+5</sub> (Figure 34B)..... Hercostomus lanceolatus -Stigma short, reaching the as same as level of thickening of vein R<sub>4+5</sub> ..... 3) First flagellomere elongate (at least two times as long as wide) (Figure 30A, 31)..... 4 4) All coxae blackish colour; generally, femora blackish except some part at tip of fore and mid femora; cercus comprising of three strong bristles at outer margin..... - Fore coxae yellowish colour; mid and hind coxae brown; all femora yellow, and cercus - Fore tarsomere relatively shorthened; cercus distinctly large, with short apical tail... 6) Fore and hind coxae yellow; cercus nearly geniculate, aedeagus with two inner denticles ...... Hercostomus brevicornis - Only fore coxae yellow; cercus triangular in shape, aedeagus with one inner 

## 3.2.2) Molecular (COI) Based Identification

## Maximum Likelihoods (ML) and Bayesian Analyses

In this study, five representative species of mangrove *Hercostomus* Loew were selected, namely, *Hercostomus lanceolatus*, *H. plumatus*, *H. obtusus*, *H. brevicornis*, *H. brevidigitalis*. For Maximum Likelihoods (ML) analysis (Figure 39), this revealed the presence of the five separated clades of mangrove *Hercostomus* Loew from peninsular Thailand. 1<sup>st</sup> clade, called *H. lanceolatus* (pink labelled) which has strongly 99 % bootstrap support, comprised 14 individuals which represented a single species of *H.lanceolatus* from several provinces in Thai Peninsula such as Chumphon, Songkhla, Surat Thani and Satun. 2<sup>nd</sup> clade comprised a single species of *H. plumatus* (green labelled) with 99% bootstrap support. 3<sup>rd</sup> clade, also has only one species in this particular clade with high strongly support of bootstrap value, namely, *H. obtusus* (black labelled), 4<sup>th</sup> clade consisted of a single species, namely, *H. brevicornis* (red labelled) with 99% bootstrap support. Lastly, 5<sup>th</sup> clade comprised a species of *H. brevidigitalis* (blue labelled). Despite, this was monophyletic relationship but nonetheless the lowest 44% bootstrap support as shown in Figure 38.

In term of Bayesian Analysis (Figure 40), our findings also revealed the monophyletic relationship of *Hercostomus*. In general, five clades could be recognised from the Bayesian phylogenetic tree.  $1^{st}$  clade (pink labelled) consisted only *H. lanceolatus* collected from Satun, Songkhla, Chumphon, Pattani, and Surat Thani province. Moreover, this clade also showed high probability value (0.91) indicating that they separately evolved from their closer alley *H.plumatus*. Additionally, the subclade also showed the same pattern with probability value (0.9).  $2^{nd}$  clade (green labelled) composed with a single species, *H. plumatus* with high probability score (0.91) indicating on the branch as a strong evident supporting that there were completely separated from *H. lanceolatus*.  $3^{rd}$  clade (black labelled) contained one species known as *H. obtusus* with high value of probability (0.98).  $4^{th}$  clade (red labelled), *H. brevicornis* which has high probability value (0.98) indicating clearly separated from *H. obtusus*. 5th clade (blue labelled) comprised a single species *H. brevidigitalis*, and also showed the highest of probability value (1) within genus (Figure 40).

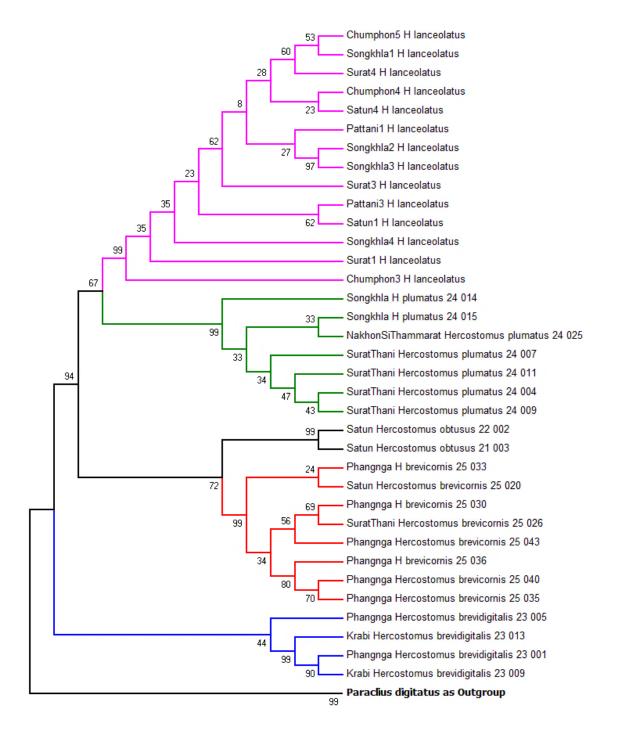


Figure 39. Phylogenetic tree for *Hercostomus* Loew relationships based on Maximum Likelihoods (ML) analysis. Bootstrap supports are indicated on the branches.

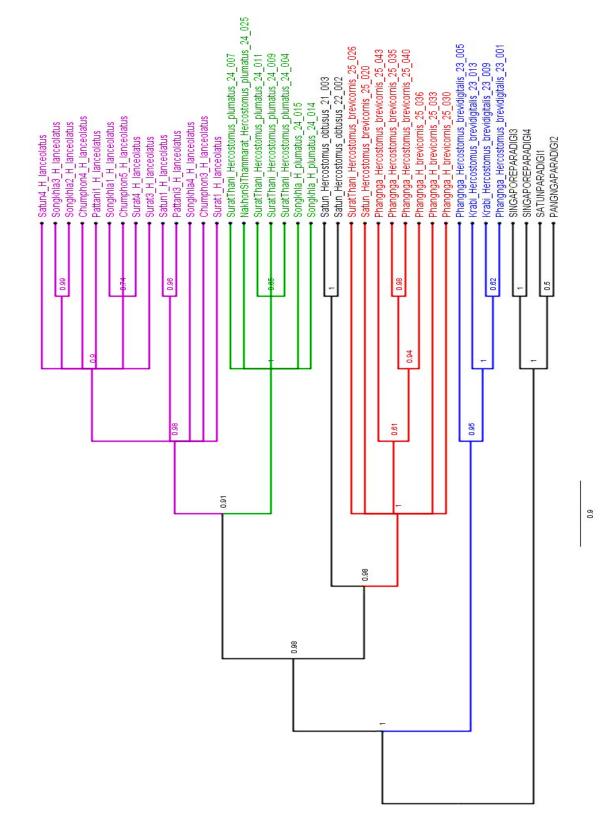


Figure 40. Phylogram of *Hercostomus* Loew based on Bayesian inference analysis performed in Mr.Bayes software.

In the current study, the phylogeny of the mangrove *Hercostomus* Loew was investigated using the cytochrome oxidase subunit I (COI gene). Molecular data analysis greatly provides a clear explanation on the phylogenetic relationships for this genus in Thailand, perhaps due to the fast-evolved rate of this gene.

As demonstrated by Maximum Likelihoods (ML) phylogenetic tree (Figure 39) and Bayesian inference (Figure 40) above indicate that five species; Hercostomus lanceolatus, H. plumatus, H. obtus, H. brevicornis and H. brevidigitalis were distinct species group when analysed using ML-method, and of which four species; H. lanceolatus, H. plumatus, H. obtus, H. brevicornis were strongly supported by 99% bootstrap value, except H. brevidigitalis was 44% supported by bootstrap value (Figure 39), whereas analyzed using Bayesian inferences all species showed high probability scores from 0.98 until 1.0 (Figure 40). The results from all analyses clearly revealed that mangrove Hercostomus sensu lato in Thailand depicted monophyletic relationships (Figure 39, 40) and completely congruent to morphology based identification (Figure 41). Surprisingly, this finding agreed with Zhang et al. (2008) who first recognised seven new species of marine Hercostomus sensu lato from Singapore mangroves using male genital and non-genital features as taxomonic tool for species identification. However, the same authors did not provide any information about phylogenetic relationships. While, a precious genetic data announced by Lim et al. (2010) combined six markers from mitochondrial and nuclear genes found that two mangrove species belong to Hercostomus, namely, H. meieri and H. brevidigitalis were closely related and showed monophyletic relationships (based on ML analysis) and classified as a sister group of the three species of Dolichopodinae such as Tachytrecus tessellatus, Lichtwardtia ziczac, and including Dolichopus bigeniculatus. On the other hand, a previous study by Brooks (2005) mainly adopted external morphology (74 characters) to assess the species phylogenetic relationships and found that the genus Hercostomus sensu lato was paraphyletic descendant.

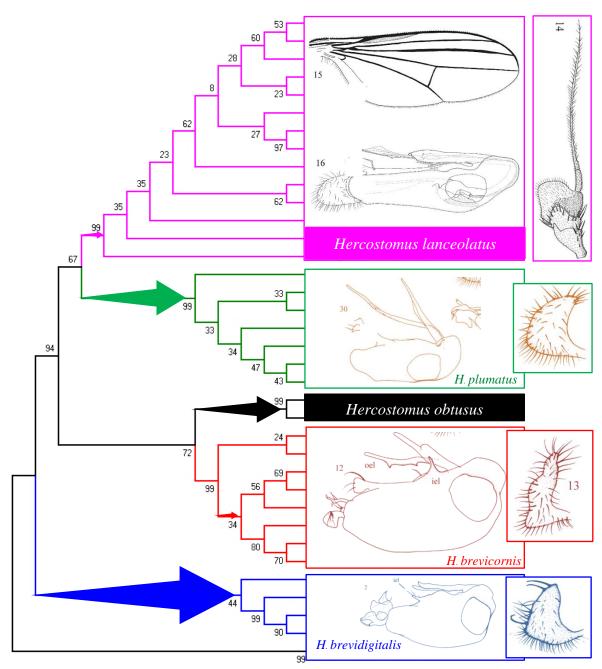


Figure 41. ML phylogenetic tree of Hercostomus Loew in Thailand

## • Further discussion on species identification (external morphology based vs molecular based, COI gene) and monophyly

In general, this study in concordance with numerous evidences of dolichopodid taxonomic studies elucidated that male genital features and some external morphology traits are useful in term of species idetification and recognition. For Ngirhaphium Evenhuis and Grootaert, Hercostomus Loew and other genera belong to marine Dolichopodidae family. For example, marine Paraclius Loew that have been investigated from Singapore mangroves by Zhang et al. (2007a), nine species were erected to new species primarily based on male genital and non-genital morphology triats such as wing pattern, antennae, and chaetotaxy pattern across the legs. Grootaert (2006c) identified nine new species from thirteen species of mangrove Teuchophorus Loew from Singapore merely used male genital features and a part male fore head; such as antenna. Furthermore, not only marine long-legged flies from Singapore, but in Thailand and China species also shown the same pattern that using only male genitalia and other morphology traits to identify species are successfully. To illustrate that statement, some examples are shown as follows: in Thailand, a marine genus, Asyndetus Loew were dissected and studied by Grootaert and Meuffels (2002), they found three species of marine Asyndetus Loew that generally occurred along the beaches in Thailand containing in different male genitalia and external morphological features as follows: in the case of A. ciliatus Grootaert and Meuffels, tip of abdomen with 4 strong macrochetae; cerci small, dark brown, haired; ventral surstylus with a minute dorsal bristle; when A. ciliatus Grootaert and Meuffels, contains a typical male hypopygium with a minute dorsal bristle on ventral surstylus; whereas A. thaicus Grootaert and Meuffels, having an elongated epandrial lobe, with a single minute bristle near tip, and epandrial lobe not so slender as in A. latifrons. However, there are no one tested using molecular data to confirm those species identification.

More than these marine species, genital and none genital morphology is also highly successful in term of freshwater species. For example, Brooks and Ulrich (2012) classified the *Microphorella similimis* from its closely allies, *M. praecox* (Dolichopodidae: Parathalassiinae) mainly based upon male morphological characters and especially male genitalia, they found that *Microphorella similimis* completely differs from *M. praecox* as follows: postpedicel shorter and stylus longer (postpedicel longer and stylus shorter in *M. praecox*), wing vein  $R_{4+5}$  and  $M_1$  sinuous (straight in *M. praecox*), and interestingly male hypopygium was also definitely different; ventral epandreal process lacking hump-like projection on ventral arm of furca, left postgonite lobe with bifurcate apex, phallus bearing pointed process near middle and lacking longitudinal serration, right cercus with basilateral portion enlarged (less developed in *M. praecox*) (Brooks and Ulrich, 2012). Due to those facts, it could be confirmed that external morphology based identification such as male genitalia, wing vein pattern, antennae, and chaetotaxy pattern across the body of marine long-legged flies are good enough to recognise species.

As can be seen above, previous researchers mainly used a single taxonomic tool such as male genital features for species recognition, but nonetheless a remarkable example on using integrated taxonomic tools, genital and non-genital features and COI gene to identify species on mangrove *Thinophilus* Wahlberg from Chinese mangroves by Grootaert et al. (2015) found that the male genital features and a part of male external morphological characters and also COI data could be clearly extended into two new species of mangrove Thinophilus Wahlberg from Shenzhen, southern China. Of the two, T. dongae and T. zhuae, both species consist of unique morphological features as follows: in T. dongae, a small species with entirely yellow fore coxae; mid and hind coxae brown with yellow apex. Legs yellow, only tarsomere 5 of all legs pale brownish. Fore coxa with long black bristles. Wing brownish tinged without spots. Fore femur with short, inconspicuous ventral bristles; fore tibia without ventral spinules, only a row of bristles. Mid and hind femora without long ventral bristles. Mid femur with a long black preapical av and hind femur with 2 ad. Four equally long dc. Antenna yellow, pedicel and postpedicel faintly brownish dorsally. Surstylus pale brown, 1/3rd length of abdomen; cercus shorter than surstylus, yellow with yellowish bristling.while, T. zhuae, different from other hydophorine flies in having this following characters, there is a large species. Legs mainly yellow, but fore coxae with a black lateral streak at base; mid and hind coxae brown except apex; tip of hind femur and base of hind tibia with a faint brownish ring in male, blackish in female; tarsomere 5 of all legs dorsally black. Fore coxa with short, black bristles on apical half. Wing faintly brownish tinged, darker anteriorly between costa and  $R_{2+3}$  and  $R_{3+4}$ ; without spots. Fore femur ventrally widened with a double row of short, strong spine-like bristles; fore tibia with a ventral row of black spinules. Fore tarsomere 1 ventrally with a dense row of thick flattened spinules. Fore tarsomeres 4 and 5 flattened. Five dc lengthening towards scutellum. Antenna yellow, pedicel and postpedicel faintly browned dorsally. Cercus large, black, keel-shaped with dense long black apical bristling (Grootaert et al., 2015). Whereas, the COI sequencing data from the same authors were also completely agreed with external morphology based identification, both species show high different in genetic variation between known mangrove *Thinophilus* from Singapore such as *T. dongae* is definitely separated from *T.simplex* with 98% bootstrap values supported, but nonetheless *T. zhue* is very low segregated from its allies by 14% bootstrap value supported (Grootaert et al., 2015).

Therefore, at present, the age of molecular taxonomy (molecular species concept) or genomic blooming era, plenty of living things have been discovered using this peculiar tool. This tool is widely accepted by modern taxonomists especially the study on molecular taxonomy and phylogenetic relationships of Dolichopodidae. For instance, Bernasconi et al. (2007), a person who adopted DNA sequences data to understand phylogenetic relationships of European Dolichopodidae. While, Germann et al. (2010), analysed the congruency of the two taxonomic tools in term of species identification used 31 characters of external morphology and four markers (COI, 12S, 16S, and nuclear-ITS2) of molecular data from 82 specimens of 49 species belong to the subfamily Dolichopodinae, and found that molecular markers disagree with morphology identification on some species of the genus Dolichopus such as D. plumipes, D. wahlbergi, D. polleti, D. simplex, D. nigricornis. Then, Lim et al. (2010), who used mitochodrial and nuclear markers to reveal the monophyly of Dolichopodidae and further suggest the rapid origin and subfamily concepts of this fly (Dolichopodidae), they confirmed that several subfamilies of Dolichopodidae were monophyletic relationships, such as, Sympicninae, Sciapodinae, Dolichopodinae, Hydrophorinae, and including Neurigoninae. In addition, they also restoring the tribe Aphrosylini (previously classified into subfamily Hydrophorinae) as a distinctive subfamily called Aphrosilinae.

This findings strong support that short COI sequences such DNA barcodes has led an interesting idea and very successfully identified species of marine *Ngirhaphium* Evenhuis and Grootaert and mangrove *Hercostomus* Loew. Additionally, it also support the definition of molecular species concept that DNA is one of the basis tool for recognising species of living things on earth. However, due to nature of COI-DNA barcode contains very short of sequences (~500 bp or lower) or little informative characters (Hajibabaei et al., 2006) may lead to conclude that this tool is not fit to use in order to understand the species phylogenetic relationships (molecular phylogenetics) of long-legged flies, eventhough both data are completely obtained from the same nucleotides of DNA. But nonetheless, DNA barcode maybe provides a great data for species relationships or molecular phylogenetic relationships when increasing a number of genes or doing multigenes analysis (Hajibabaei et al., 2007; Meier et al., 2006, 2016).

# **3.3**) Preliminary analysis on population genetic of marine long-legged flies with referenced to *Hercostomus lanceolatus* in peninsular Thailand

## **3.3.1)** Haplotype diversity and network

The 28 haplotypes (appendix B) of *Hercostomus lanceolatus* were chosen for understanding the genetic structure and variation. In this study, the the average number of nucleotide differences (K-value), nucleotide diversity  $(\theta_1)$ , and nucleotide diversity (Jukes and Cantor) ( $\theta_2$ ) analyses indicated that *Hercostomus lanceolatus* haplotype diversity differences among populations in the Gulf of Thailand (South China Sea). The population from Surat Thani province were the lowest genetic variation (Kvalue=4.000;  $\theta_1$ =0.01278;  $\theta_2$ =0.01291) comparing to other populations such as population from Chumphon province (*K*-value=7.333;  $\theta_1$ =0.02343;  $\theta_2$ =0.02385), Pattani province (*K*-value=6.600;  $\theta_1$ =0.02109;  $\theta_2$ =0.02140), Songkhla province (*K*-value=5.833;  $\theta_1$ =0.01864;  $\theta_2$ =0.01893), and also including population from Singapore (K-value=6.095;  $\theta_1$ =0.01974;  $\theta_2$ =0.01979) (Table 10). Regarding all results (Table 10 and 11), it was clearly indicated that the gene flowed among populations from Surat Thani province (Figure 42) to the nearest bay such as Chumphon, and moved down to Songkhla bay, Pattani bay until Singapore island (located in between West and Eastcoasts of the Thai-Malay Peninsula) (Figure 43). Conversely, for the Andaman Sea (Westcoast, Satun population), the average number of nucleotide differences (K-value=10.000), nucleotide diversity ( $\theta_1$ =0.03195), and nucleotide diversity (Jukes & Cantor) ( $\theta_2$ =0.03265) (Table 10) of *H. lanceolatus* clearly expressed that haplotype diversity was noticeably high or low in term of gene flow. For this event, it could be said that genetic differentiation was recognised between the population from Westcoast and Eastcoast or, in other word, gene flow from parental population (Surat Thani province) in Eastcoast to Satun mangrove in Westcoast was low. This could be cleared up by mangrove geographical distance in the Andaman Sea that is far from parental population (Surat Thani population) which may lead to high genetic variation. Beyond this point, the mangrove in Eastcoast is clumped patches and may able to limit dispersal ability of *H. lanceotus* from parental population to Tammalang mangrove in Satun province (Figure 43).

Table 10. Haplotype relationships within species of *Hercostomus lanceolatus* in Thailand and Singapore Island

Population	N	Polymorphic site (S)	Average number of nucleotide difference (K)	Nucleotide diversity (θ1)	Nucleotide diversity (Jukes & Cantor) (θ <sub>2</sub> )
Chumphon	3	11	7.333	0.02343	0.02385
Pattani	5	15	6.600	0.02109	0.02140
Satun	2	10	10.000*	0.03195*	0.03265*
Songkhla	4	11	5.833	0.01864	0.01893
Surat Thani	7	12	4.000**	0.01278**	0.01291**
Singapore	7	14	6.095	0.01947	0.01979
All Population	28	37	6.870	0.20195	0.02233

Note that; \*\* is the highest value, \* is the lowest value

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Table 11. Genetic distances between six populations (Surat Thani, Patani, Chumphon, Songkhla, Satun, and Singapore) of mangro           Hercostomus lanceolatus (COI marker).         1         2         3         4         5         6         7         8         9         10         11         11         2         23         24         5         6         7         8         9         10         11         12         14         15         16         17         18         19         20         21         23         24         25         28         23         24         25         23         24         25         23         24         25         23         24         25         6         7         8         9         10         11         12         14         15         16         17         18         19         20         21         23         24         5         6         7         8         9         10         11         12         14         15         16         17         18         19         20         21         22         23         24         25         23         24         25         23         24         25         23         24

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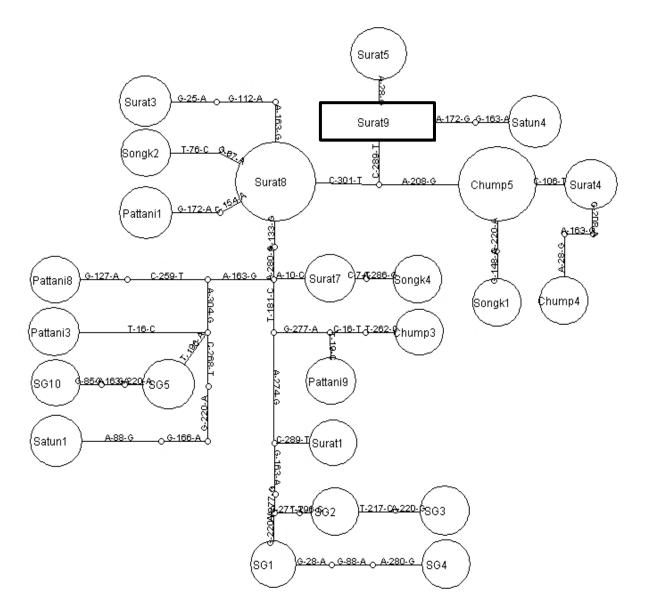


Figure 42. Haplotype network dendrogram of *Hercostomus lanceolatus* in peninsular Thailand estimated with statistical parsimony. Note that retangle is parental population.

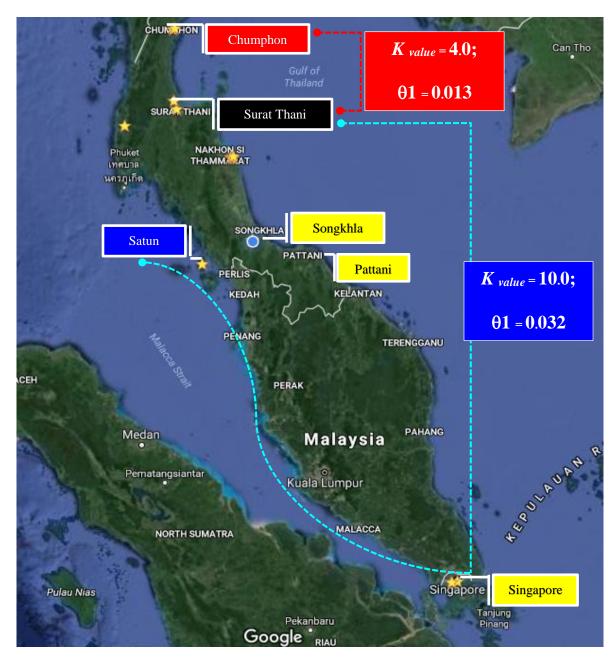


Figure 43. Comparison of averrage number of nucleotide differences (*K*-value) and nucleotide diversity ( $\theta_1$ ) among population of *H. lanceolatus* in the region of peninsular Thailand and Singapore.

Table 12. Comparison of averrage number of nucleotide differences (*K*-value) among population of *Hercostomus lanceolatus* in the region of peninsular Thailand and Singapore

Populations	1	2	3	4	5	6
1) Surat Thani	-					
2) Chumphon	5.095*	-				
3) Songkhla	5.107	6.417	-			
4) Pattani	5.571	6.267	6.365	-		
5) Satun	6.857	8.000	8.250	7.200	-	
6) Singapore	8.621	8.333	9.036**	7.457	8.286	-

Note that; \*\* is the highest value, \* is the lowest value

In addition, as it has been pointed out (Table 12, Figures 45, 46) the averrage number of nucleotide differences (K-values) among population of mangrove H. lanceolatus in the Gulf of Thailand Sea, including Singapore (South China Sea) showed relatively high differrent of K-values from 5.095 to 9.036. Perhaps because of mangrove geographical distances between upper and lower Gulf of Thailand (South China Sea) may lead to the genetic differentiation. For the population in the mangrove of Phanangtak, Chumphon (Chumphon bay, Gulf of Thailand) and in the mangrove of Tha Chang, Surat Thani (Surat Thani bay, Gulf of Thailand) was lowest (K-value = 5.095) (Table 12). This could be explained by a very short mangrove geographic distances (about 160 km) between Surat Thani bay to Chumphon bay in the Gulf of Thailand (Figure 45). In addition, the short geographic distances and no geographic barrier found between two populations might be the main factors leading to the high genetic similarity between two populations. However, both populations sampled in Songkhla province and Pattani province showed high of the averrage number of nucleotide differences (K-value=6.365) or lower gene flow than both Chumphon and Surat Thani province, although mangrove distance between two population is less than 110 km (Figure 45). As a matter of fact that there are completely differences in the context of marine environmental conditions from two populations, in the lagoon (Songkhla province) and in the open sea (Pattani province). It could be assumed by the salinity difference of sea water which is low in the lagoon (Songkhla), but high in the open sea (Pattani)

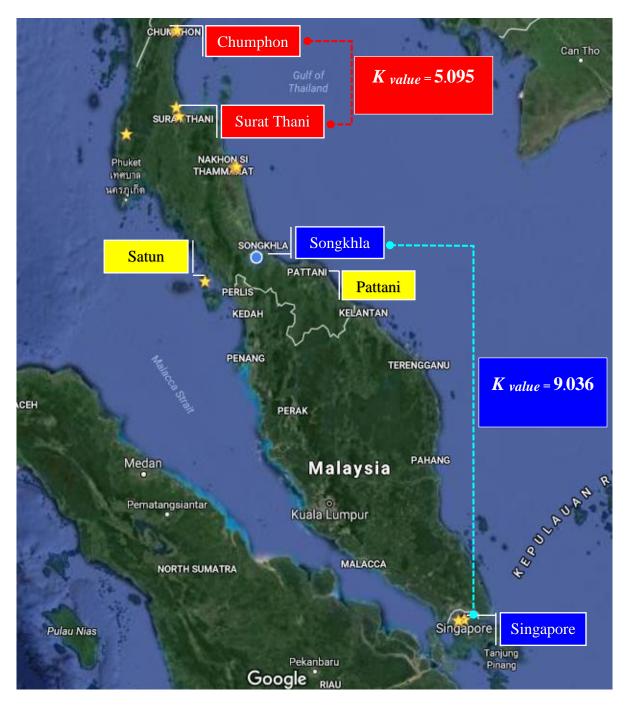


Figure 44. Comparison of averrage number of nucleotide differences (*K*-value) among populations of *H. lanceolatus* in the region of peninsular Thailand and Singapore



Figure 45. Geographical distances between the population from Phanangtak, Chumphon province and Tha Chang, Surat Thani province (Gulf of Thailand, South China Sea)

Further results from haplotype analaysis also revealed that the averrage number of nucleotide differences (*K*-values) between population from Songkhla province, Thailand (Gulf of Thailand, South China Sea) and Pulau Ubin, Eastern Singapore (South China Sea) was highest. Due to this fact, it could be explained by the very long mangrove geographical distances (about 992 km) (Figure 46) and together with habitat differences. In Na Thab, Songkhla, is a big mangrove creeks which is mainly surrounded by brackish water as well as the influence of human anthropogenic threat (Figure 47) such as, land use and aqautic farms that be able to separate mangrove into several small patches.

Similarly, the population from Laem Pho, Hat Yai, Songkhla province sampled from innermost of the lagoon were also low in salininy. Meanwhile, Singapore population mainly collected from back mangroves from the Chek Jawa, Pulau Ubin, open sea were encompassed with high salinity of sea water (Figure 48). Even though this island is known as tourism island, it was less disturbed than Songkla population.



Figure 46. Geographical distances between the population from Laem Pho and Na Thab, Songkhla, Thailand and the Chek Jawa, Pulau Ubin, Singapore.

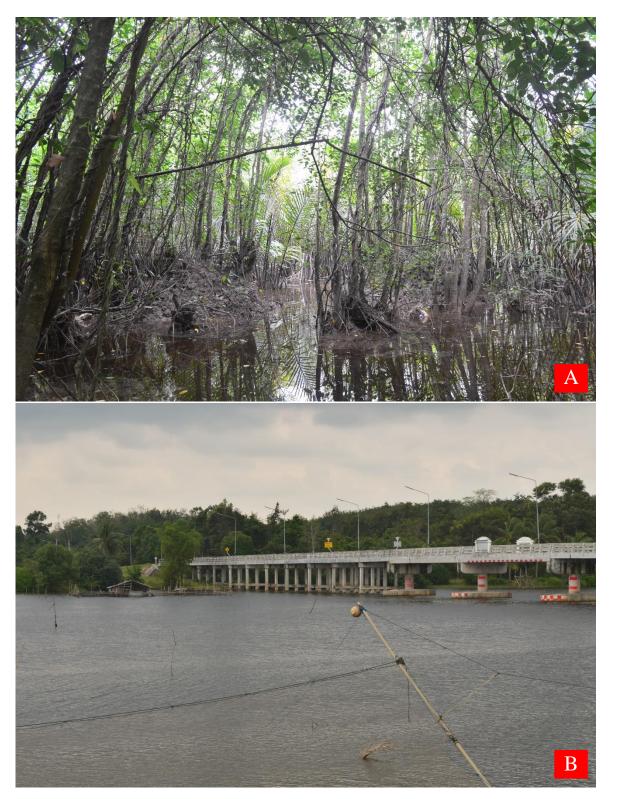


Figure 47. Back mangrove in Na Thab, Chana, Songkhla with brackish water surrounded (A) and local fishing gears or fish traps for shrimp and fish farm purposes (B).

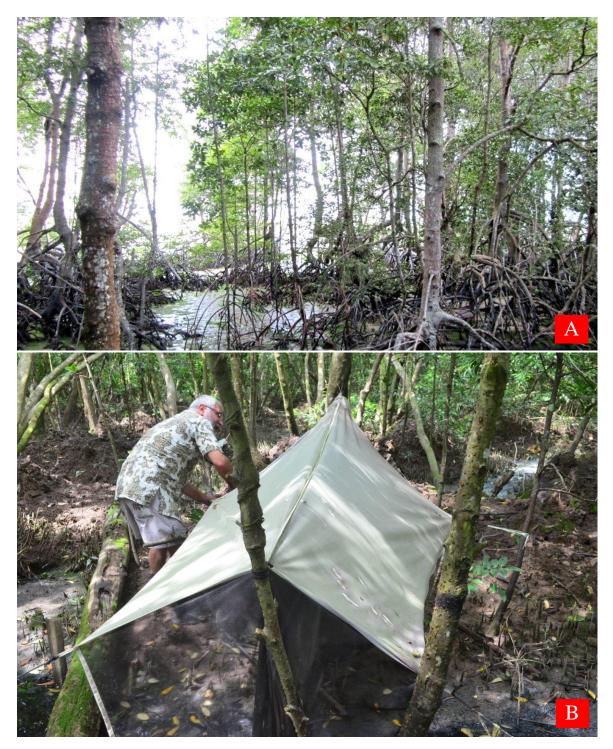


Figure 48. Front sea mangrove with high salinity (A) and back mangrove (B) at the Tanjong Chek Jawa, Pulau Ubin, Singapore.

According to this major finding has led to the conclusion that the genetic variation and diversity of marine long-legged flies (Dolichopodidae), especially mangrove Hercostomus lanceolatus was largely influenced by mangrove geographical distances and habitat complexity. Furthermore, the results clearly supports the theory of island biogeography of organisms predicted by MacAthur and Wilson in 1967, that closer island may easy to disperse and establish new colony (or recolonize), whereas the more isolated island (high degree of isolation from mainland or very distance to nearest neighbour) may limit the dispersal ability or decrease the migration rate of organisms. Similarly, with reference to insects, it was remarkably tested in several mangrove islands in the Florida Keys by E. O. Wilson (a person who coined the term of island biogeography). The various representative mangrove islands were chemical fumigated to eliminate the insect populations in order to understand species richness. The immigration of insect species onto the island were then surveyed and investigated, and it was found that the species thoroughly recolonized within a year. The islands that were closer to the mainland recovered at a faster rate, which follows the theory of island biogeography (MacArthur and E. O. Wilson, 1963).

## 4. CONCLUDING REMARKS

#### 4.1) Species Composition

In summation, the present survey shows notably different results from the previous survey by Grootaert and Meuffels (2001) nearly two decades ago, and also reveals important basic information such as species composition, distribution pattern and range, habitat preferences of marine long-legged flies in Thailand. First and foremost, the species number, this survey clearly shows that the species number is drastically increased (from fifteen species to sixty species or accounted for 79.49%) from previous studies by Grootaert and Meuffels (2001). Of the sixty species of marine long-legged flies which were found from this region, seventeen species were recognised as species new to science, and include a remarkably surprising new genus, namely, *Ornamenta siamese* sp. nov., gen. nov. Additionally, further results indicate that the *Thinophilus* Wahlberg was the most diverse among them; at most sixteen species belonging to this genus have been identified (deeply detailed in published paper 2).

In terms of distribution pattern, most species of marine long-legged flies were largely distributed along the Andaman seacoasts (Indian Ocean) rather than the Gulf of Thailand (South China Sea), to illustrate this, twenty three species were strictly dwelling in various kinds of marine habitats on the western coast (Andaman Sea), whereas only fifteen species were mainly distributed along the eastern coast (Gulf of Thailand Sea). This could be interpreted by the size and abundance of mangroves that might affect species composition of the two sides of peninsular Thailand and also confirm the "island biogeography" theory proposed by MacArthur and Wilson in 1967. Interestingly, a species of mangrove-dwelling long-legged fly, Hercostomus Loew (H. lanceolatus) shows a striking result, it is widely dispersed on both sides of peninsular Thailand, while Ngirhaphium Evenhuis and Grootaert provides notable evidence for this issue: lots of species are strictly found in particular mangroves or very clumped such as N. chutamasae sp. nov. is solely found in Tammalang, Satun province, whereas N. meieri sp. nov. is observed from Takuapa, Phang Nga Province on the Andaman Sea side, and N. caeruleum only occurs in Surat Thani Bay, Surat Thani province and Chumphon Bay, Chumphon province in Gulf of Thailand Sea side.

For habitat preference, mangroves seem to be the most favoured habitat for marine long-legged flies in Thailand, where forty-four species were recognised from several mangroves in peninsular Thailand. Conversely, rocky shores seemed to be the less preferred domicile, only two species were investigated. This could be a result of less humidity and moist places than other marine habitats.

#### 4.2) Species Identification and Molecular Phylogeny

The results generally told us that Thai marine long-legged fly identification using two taxonomic approaches are completely congruent species number. For instance, in mangrove *Ngirhaphium* Evenhuis and Grootaert, five species were recognised by genital and non-genital morphological features and in concordance with COI barcode results (Maximum Likelihoods [ML] Analysis), it provides five distinctive clades and monophyletic relationships of *Ngirhaphium* Evenhuis and Grootaert in Thailand, including, *N. caeruleum*, *N. chutamasae* sp. nov., *N. meieri* sp. nov., *N murphyi*, and *N. sivasothii*. Besides, genetic distances of the latest member, *N. meieri* sp. nov. is closely related to *N. chutamasae* with 6% difference and completely agreed with external morphology based, found that both species as similar as of male genital features but dorsal is definitely different (mushroom-like dorsal surstylus in *N. meieri* sp. nov., whereas boot-like in shape for *N. chutamasae* sp. nov.). Furthermore, the range of distribution is also different. In the case of *N. merieri* sp. nov. it mainly lives in Takuapa, Phang Nga Province, but *N. chutamasae* sp. nov. is solely found from Tammalang mangroves in Satun Province.

While, *Hercostomus* Loew, six species were perceived by traditional way of identification, namely, *H. brevicornis*, *H. brevidigitatus*, *H. lanceolatus*, *H. obtusus*, *H. plumatus*, and *H. propermeieri* sp. nov. In the context of modern way of identification, also known as molecular analysis (based on the Maximum Likelihoods and Bayesian Analyses), only five representative species were examined, and found that they were entirely congruent to traditional way of identification. The results revealed five clusters and monophyletic relationships of mangrove *Hercostomus* Loew in this region. They included *H. brevicornis*, *H. brevidigitatus*, *H. lanceolatus*, *H. obtusus*, and *H. plumatus*.

## 4.3) Preliminary Analysis on Population Genetic

Regarding *Hercostomus lanceolatus*, genetic variation and haplotype diversity from several populations in Thailand and Singapore were tested in order to understand the population genetic relationships. The present result clearly disclosed that there are slight differences of genetic distances among the population of *H. lanceolatus* that is presented by the highest values of *K* (average number of nucleotide differences, 10.000),  $\theta_1$ (Nucleotide diversity, 0.032),  $\theta_2$  (Nucleotide diversity with Jukes and Cantor, 0.033) in Satun Province population, on the other hand, the lowest values of *K* (average number of nucleotide differences, 4.000),  $\theta_1$  (Nucleotide diversity, 0.013),  $\theta_2$  (Nucleotide diversity with Jukes and Cantor, 0.013) in Surat Thani population. In addition to what has been said the average number of nucleotide differences (*K*-value) between Surat Thani and Chumphon populations (*K*-value = 5.095) are low or closely related and conversely, the *K*-value of Songkhla and Singapore populations are high or it could be interpreted that there is low rate of gene flow between the two populations. This could be explained by geographical distances of mangroves and human anthropogenic threat (habitat fragmentation) might be affecting gene flow of marine *H. lanceolatus* in this region.

## **5. RECOMMENDATION FOR FURTHER STUDY**

According to my research experience, there is a distinct lack of knowledge in the areas of number of specimens and molecular markers and female identification, so this would make a strong case for further research in these areas by:

5.1) The area of sampling sites and sampling techniques should be paid attention to in the contexts of obtaining higher numbers of specimens, including various kinds of target fly species in order to obtain enough samples and clearly understand in a molecular phylogenetic study.

5.2) Cytochrome C Oxidase Subunit I (COI) is one of the most suggested markers for marine long-legged fly species identification and confirmation. However, for higher level classification and phylogenetic relationships, combined genes and multigene analysis are highly recommended.

5.3) Entomologists should be wary of the female identification due to female long-legged flies may not contain a distinct sexual morphological feature such as a male fly and then female terminalia study should be investigated and measured in order for precise identification.

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# 8. APPENDICES

# Appendix A

# Nucleotide sequence alignment of the COI gene of mangrove *Ngirhaphium* Evenhuis and Grootaert

>Chumphon\_Ngirhaphium\_caeruleum\_27\_002\_ABDO07

\*CTATCCGCAGGAATCGCTCACGGTGGAGCATCAGTAGACTTAGCAATTTTTTCCCTA CACTTGGCAGGAGTCTCATCAATTCTAGGAGCAGTTAACTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTCCTTCTCCCCTACCAGTATTAGCAGGAGGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>SuratThani\_Ngirhaphium\_caeruleum\_27\_009\_ABDO07

\*CTATCCGCAGGAATCGCTCACGGTGGAGCATCAGTAGACTTAGCAATTTTTTCCCTA CACTTGGCAGGGGTCTCATCAATTCTAGGAGCAGTTAACTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTCCTTCTCCCCTACCAGTATTAGCAGGAGGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>SuratThani-Ngirhaphium-caeruleum-27-011-ABDO07

\*CTATCCGCAGGAATCGCTCACGGTGGAGCATCAGTAGACTTAGCAATTTTTTCCCTA CACTTGGCAGGGGTCTCATCAATTCTAGGAGCAGTTAACTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTCCTTCTCCCCTACCAGTATTAGCAGGAGGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>SuratThani-Ngirhaphium-caeruleum-27-013-ABDO07

\*CTATCCGCAGGAATCGCTCACGGTGGAGCATCAGTAGACTTAGCAATTTTTTCCCTA CACTTGGCAGGGATCTCATCAATTCTAGGAGCAGTTAACTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTCCTCCTCTCCCTACCAGTATTAGCAGGAGGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>SuratThani-Ngirhaphium-caeruleum-27-018-ABDO08

\*CTATCCGCAGGAATCGCTCACGGTGGAGCATCAGTAGACTTAGCAATTTTTTCCCTA CACTTGGCAGGGATCTCATCAATTCTAGGAGCAGTTAACTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTCCTCCTCTCCCTACCAGTATTAGCAGGAGGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>ckq-DOL-ZRCBDP00001462-Ngirhaphium-caeruleum-M-PT3

 $CCTATCCGCAGGAATTGCACACGGCGGGGGGCATCAGTAGACTTAGCAATTTTTTCCCTG\\CACTTAGCAGGAATTTCATCAATTCTAGGAGCAGTTAACTTCATCAACAGTAATTA$ 

130

ATATACGATCCACAGGAATTACATTTGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTTCTTCTCCCCCCCAGTATTAGCAGGAGCTATTACAATA TTATTAACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>ckq-DOL-ZRCBDP00001670-Ngirhaphium-caeruleum-M-SMN1

CCTATCCGCAGGAATTGCACACGGCGGGGGGCATCAGTAGACTTAGCAATTTTTTCCCTG CACTTAGCAGGAATTTCATCAATTCTAGGAGCAGTTAACTTCATCACAACAGTAATTA ATATACGATCCACAGGAATTACATTCGATCGAATACCACTATTTGTATGATCAGTAGT TATTACAGCAATTCTGCTTCTTCTATCCCTCCCAGTATTAGCAGGAGGCTATTACAATA TTATTGACAGACCGAAACTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGAC CCAATCCTTTACCAACACTTATTC

>ckq-DOL-ZRCBDP00002037-Ngirhaphium-caeruleum-M-SMN1

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>Krabi-Ngirhaphium-murphyi-28-013-ABDO08

>Krabi-Ngirhaphium-murphyi-28-014-ABDO08

>Satun-Ngirhaphium-murphyi-28-006-ABDO08

>Satun-Ngirhaphium-murphyi-28-007-ABDO08

 TACTAACAGATCGAAATTTAAATACTTCATTCTTTGACCCAGCAGGAGGAGGTGACC CTATTCTTTACCAACATTTATTC

>Krabi-Ngirhaphium-murphyi-28-008-ABDO08

>ckq-DOL-ZRCBDP00001291-Ngirhaphium-murphyi-M-MM1

>ckq-DOL-ZRCBDP00001351-Ngirhaphium-murphyi-F-MM2

>ckq-DOL-ZRCBDP00001292-Ngirhaphium-murphyi-M-MM1

>Krabi-Ngirhaphium-sivasothii-26-030-ABDO07

TCTATCTGCAGGAATTGCCCATGGAGGAGCCTCAGTAGACCTAGCAATCTTCTCTTTG CATTTAGCAGGTATTTCATCAATTCTAGGAGCAGTAAATTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACCTTTGATCGAATACCCCTATTTGTATGATCAGTAGT AATTACTGCAATCCTACTTCTTCTTTCTCTCCCCAGTTTTAGCCGGAGCTATTACAATAC TCCTAACAGACCGAAACTTAAATACCTCATTTTTTGACCCAGCAGGAGGAGGAGGAGATC CAATCCTTTACCAACATCTATTC

>Satun-Ngirhaphium-sivasothii-26-004-ABDO07

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>Krabi-Ngirhaphium-sivasothii-26-024-ABDO07

TCTATCTGCAGGAATTGCCCATGGAGGAGCCTCAGTAGACCTAGCAATCTTCTCTTTG CATTTAGCAGGTATTTCATCAATTCTAGGAGCAGTAAATTTCATTACAACAGTAATTA ATATACGATCCACAGGAATTACCTTTGATCGAATACCCCTATTTGTATGATCAGTAGT AATTACTGCAATCCTACTTCTTCTTCTCTCCCCAGTTTTAGCCGGAGCTATTACAATAC TCCTAACAGACCGAAACTTAAATACCTCATTTTTGACCCAGCAGGAGGAGGAGGAGATC CAATCCTTTACCAACATCTATTC

>Phangnga-Ngirhaphium-sivasothii-26-008-ABDO07

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>Satun-Ngirhaphium-sivasothii-26-003-ABDO07

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>Krabi-Ngirhaphium-sivasothii-26-005-ABDO07

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>ckq-DOL-ZRCBDP00001199-Ngirhaphium-sivasothii-F-SR3

>ckq-DOL-ZRCBDP00001204-Ngirhaphium-sivasothii-M-SR2

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>Phangnga-Ngirhaphium-meieri-26-005E-ABDO07

>Phangnga-Ngirhaphium-meieri-26-007G-ABDO07

>Satun-Ngirhaphium-chutamasae

Nucleotide sequence alignment of the COI gene of mangrove *Hercostomus* Loew >Phangnga\_H\_brevicornis\_25\_030

TTTATCGGCTGAAATTGCACATGGTGGAGCTTCAGTTGATTTAGCAATTTTTTCATTA CATTTAGCAGGTATTTCATCAATTTTAGGGGCGGTAAATTTTATTACTACTGTAATTA ATATGCGATCTACTGGTATTACTTTTGACCGAATACCTTTATTTGTGTGATCTGTTGTA ATTACTGCTATTTTATTACTATTATCTCTTCCTGTTTTAGCTGGTGCTATTACTATATTA TTAACTGACCGAAACCTTAATACTTCATTCTTTGATCCTGCTGGAGGTGGAGACCCAA TTTTATATCAACATTTATTT

>Phangnga\_H\_brevicornis\_25\_033

TTTATCGGCTGAAATTGCACATGGTGGAGCTTCAGTTGATTTAGCAATTTTTCATTA CATTTAGCAGGTATTTCCTCAATTTTAGGGGGCAGTAAATTTTATTACTACTGTAATTA ATATGCGATCCACTGGTATTACTTTTGACCGAATACCTTTATTTGTGTGATCTGTTGTA ATTACTGCTATTTTATTACTATTATCTCTTCTTGTGTTTAGCTGGTGCTATTACTATATTA TTAACTGACCGAAACCTTAATACTTCATTCTTTGACCCTGCTGGAGGTGGAGACCCAA TTTTATATCAACATTTATTT

>Phangnga\_H\_brevicornis\_25\_036

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>Phangnga\_Hercostomus\_brevidigitalis\_23\_001

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>Krabi\_Hercostomus\_brevidigitalis\_23\_013

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>NakhonSiThammarat\_Hercostomus\_plumatus\_24\_025

>Phangnga\_Hercostomus\_brevicornis\_25\_040

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>Phangnga\_Hercostomus\_brevicornis\_25\_035

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>Phangnga\_Hercostomus\_brevidigitalis\_23\_005

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>SuratThani\_Hercostomus\_plumatus\_24\_009

>SuratThani\_Hercostomus\_plumatus\_24\_011

>Krabi\_Hercostomus\_brevidigitalis\_23\_009

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>Satun\_Hercostomus\_obtusus\_22\_002

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>Satun\_Hercostomus\_brevicornis\_25\_020

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>Satun\_Hercostomus\_obtusus\_21\_003

>SuratThani\_Hercostomus\_plumatus\_24\_007

>SuratThani\_Hercostomus\_brevicornis\_25\_026

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>Phangnga\_Hercostomus\_brevicornis\_25\_043

TTTATCGGCTGAAATTGCACATGGTGGAGCTTCAGTTGATTTAGCAATTTTTCATTA CATTTAGCAGGTATTTCATCAATTTTAGGGGCGGTAAATTTTATTACTACTGTAATTA ATATGCGATCTACTGGTATTACTTTTGACCGAATACCTTTATTTGTGTGATCTGTTGTA ATTACTGCTATTTTATTACTATTATCTCTTCCTGTTTTAGCTGGCGCTATTACTATATT ATTAACTGACCGAAACCTTAATACTTCATTCTTTGACCCTGCTGGAGGTGGAGACCCA ATTTTATATCAACATTTATT

>SATUNPARADIGI1

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>PANGNGAPARADIGI2

TCTCTCAGCAGGAATTGCCCACGGAGGAGCATCAGTAGATTTAGCAATTTTTTCACTA CACTTAGCTGGTATTTCATCAATTTTAGGAGCAGTAAATTTTATTACAACTGTAATTA ATATACGATCTACAGGTATCACATTTGACCGAATACCTCTATTTGTGTGATCTGTTGT AATTACCGCTATTCTACTTTTACTTTCATTACCAGTATTAGCCGGAGCTATTACTATAC TTCTTACAGATCGAAACTTAAATACGTCATTCTTCGACCCTGCCGGAGGAGGAGAGCC CTATTCTTTACCAACATCTATTT

>SINGAPOREPARADIGI3

TCTTTCAGCAGGAATTGCTCACGGAGGAGCATCAGTAGATTTAGCAATTTTTTCTCTA CACTTAGCTGGTATTTCATCAATTTTAGGAGCAGTAAATTTTATTACAACTGTAATTA ATATACGATCTACAGGTATCACATTTGACCGAATACCTTTATTTGTATGATCTGTAGT AATTACAGCTATTCTACTTTTACTTTCATTACCAGTATTAGCTGGAGCTATTACTATAC TCCTTACAGATCGAAACTTAAATACATCATTCTTCGACCCTGCCGGAGGAGGAGAGCC CTATTCTTTATCAACATCTATTT

>SINGAPOREPARADIGI4

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# APPENDIX C

# **NGS-BARCODING PIPELINE**

# > Paired-end merging and demultiplexing

Paired-end merging with PEAR

Source: https://github.com/xflouris/PEAR

Usage: ./pear -f forward\_read.fastq -r reverse\_read.fastq -o output\_prefix

- PEAR will generate 4 output files.
- $\circ$  The assembled sequences will be in the fastq file with the 'assembled' suffix.
- Unassembled sequences will be in the 'unassembled' files (F and R). Take note that the sequences in the 'unassembled' file have been reverse complemented.
- $\circ$  The discarded file has the reads which did not meet the specified criteria.

# > Demultiplexing with NGS-barcoder

# Source: Code written by Amrita Srivathsan

Prepare a demultiplexing .csv file with the following format:

SpecimenID	F primer tag	R primer tag	F primer	R primer
ABCD	ATCG	ATCG	AATTCCGG	ATCGATCG

(Leave out the headers!)

- First change directory to the script folder
- Usage: python NGSbarcoder\_mult\_1.0.2.py
- The script has a GUI interface:
- o Path to Input Fasta file: Select PEAR assembled fastq file
- Path to Barcode csv file: Select demultiplexing .csv file
- Path to output directory: Create and select empty folder for demultiplexing output files
- Minimum Length: Specify a length cutoff, or leave as 0(200)
- Select Number of mismatches allowed per primer sequence: Specify number of bp mismatches in the primer sequence to account for sequencing errors. Must specify a number. Recommended 2 bp.

\* NGS barcoder will generate a fasta file of sequences for every demultiplexed sample. The summary will be in the all\_stats file, which gives total read count, barcode read count, ratio of second most dominant read to most dominant read, as well as the dominant and second most dominant barcodes.

# Barcode filtering and quality control

# $\circ$ Total count

Open the all\_stats file in Excel, sort by Total Counts. Discard samples with total read count below 50. This filters out low coverage samples.

# • Total barcode count

In the all\_stats file, sort by Total Barcode Count. Discard samples with total barcode counts below 10. This filters out low coverage barcodes.

# $\circ$ $\,$ Ratio of second dominant read count to total barcode count

In the all\_stats file, sort by Ratio of second dominant sequence to dominant sequence. Transfer samples with ratios above 0.2 to a separate spreadsheet. This filters out possibly contaminated samples.

# Converting all\_stats output to fasta format

Copy the remaining header and dominant sequence columns into a separate spreadsheet Insert an empty column to the left of the headers and fill it with '>'

Combine the '>' column with the header column using the formula: =A1&B1

Copy this new column and paste as text/values only

Have this column on the left of the dominant sequence column and copy those 2 columns into a text editor

Replace the tab delimiters (\t) with new lines (\n) and save as .fas

\* The fasta file of sequences can then be used for further downstream analyses.

# Running a Local BLAST

The final dataset should be BLASTed to ensure the reads are not from external contaminants. The local BLAST can be run using a database downloaded from GenBank or a locally curated database. The former requires the "readsidentifier" script to assign taxonomic identities while the latter requires the "makeblastdb" command to generate the database files. Also useful for metabarcoding. The BLAST module has to be installed first.

# A) BLAST against a GenBank database

- Downloading the GenBank database
- Can be done using Bio-Python, or from the NCBI GenBank FTP
- Command: wget(url)

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However, we have the COI and nucleotide (nt) database already downloaded

- Running the BLAST

Usage: blastn

query input.fasta: Specify path to input fasta file

db database: Specify path to database files (inclue file prefix)

out output: Specify path to output file(no extension required)

outfmt 6: Format of output(6: tabular, 0/default: verbose)

evalue 1e-5: Specify quality of blast hits

max\_target\_seqs x: Gives x no. of top matches

num\_threads: Specify number of cores to run the BLAST(optional)

task blastn: To run BLASTN (more dissimilar sequences)(optional)

\*IMPORTANT !!: Must make sure there are no spaces in input headers\*

Remark: The BLAST will return a file with hits to gi-numbers. ReadsIdentifier is required to assign the relevant taxonomic information to those numbers.

# Assigning taxonomic information with ReadsIdentifier

Source: https://github.com/asrivathsan/readsidentifier-1.0

Code written by Amrita Srivathsan

- GenBank Taxonomy and gi\_tax files must be downloaded from the NCBI ftp first

- Change directory to the script folder and open the config.txt
- PathToTaxonomy: Specify path to GenBank Taxonomy files
- PathToGiTaxid: Specify path to GenBank gi\_tax files
- Type: Specify single end (s) or paired end (p) inputs
- blastout: Specify path to the output of BLAST that you wish to assign taxo information to

- Identity: Specify match identity cutoff (lower if genbank has poor coverage of the taxa)

- lencutoff: Specify length cutoff for sequence overlap
- outputfileprefix: Specify path to output folder, as well as the name of the output file
- Usage: python readsidentifier.py config.txt

# B) BLAST against a locally curated database

- Generating a BLAST database

- Prepare a fasta file of sequences with informative headers
- Usage: makeblastdb
- in: Specify path to input fasta file
- out: Specify path to output, along with chosen prefix
- dbtype: nucl for nucleotide database

# Running the BLAST

Refer to previous section, but now specify -db as the newly created database.

# > Dealing with samples with multiple signals

This is applicable for metabarcoding analyses, samples with shared barcodes or a batch that is suspect to have a strong contaminant.

In these cases, the dominant/second most dominant signal is no longer reliable. Hence the individual demultiplexed fasta files have to be processed instead.

Obtaining unique reads

Source: Code written by Amrita Srivathsan

Each demultiplexed fasta file has data for every single sequence assigned to that primer combination. To reduce the size of the dataset, only the unique sequences should be considered.

- First place all the demultiplexed fasta files of interest into a folder.

- Then change directory to the one containing files allmerge.py and allmerge\_mult.py.

- Open allmerge\_mult.py and ensure the path to allmerge.py is correct. This is because allmerge\_mult.py is a generic script that can run any particular script on every file in a directory.

- Usage: python allmerge\_mult.py input\_directory

The script output gives 3 files per fasta file:

uniq: Compiles the unique sequences into a single line and gives the read counts

uniq.10: Gives all the unique sequences above 10 (> 10) read counts

merged: Gives all unique sequences above 10 read counts and compiles the length variants

\* There might be an error for some files with low coverage sequences (<10). These files will have an empty *uniq.10* file and hence the *merged* file cannot be generated.

# 2. Adding the SpecimenID to the sequence headers

Source: Code written by Amrita Srivathsan

In each *uniq*, *uniq*.10 and *merged* fasta file, the headers of each sequence do not have the Specimen ID present. This makes it impossible to trace the sequence back to the sample after concatenation.

The script that adds information to the sequence headers has 2 variants: fixheaders\_tab.py and fixheaders\_list.py. The former is for adding multiple fields of sample information (eg. locality, date, etc.) while the latter just adds the SpecimenID.

First prepare a .txt file containing the SpecimenID information. This can be done in Excel (save as tab delimited .txt). If can add additional fields of information in the subsequent columns. The Excel file should look as such:

SpecimenID	Locality (optional)	Date (optional)
ABCDE	Singapore	1Jan15

# IMPORTANT!!!:

The SpecimenID has to correspond EXACTLY to the fasta file name. Given the above example, the filename can be ABCDE.fa.merged or ABCD.fa.uniq.

Do not have spaces in any fields.

- The headers must be there.

- Usage: python fixheaders\_list.py input\_directory header\_info.txt output\_directory

- Usage is the same for fixheaders\_tab.py

- The script will then prompt for a prefix and suffix. If your fasta files have a prefix or suffix (eg. fa.merged), specify it in the command line. If not, just press enter.

\* The script will return each fasta file with the appended sequence headers in the specified output directory

- Concatenating the fasta files

- Change the directory to where the fixed fasta files are stored

- Command: cat \* > output.fas

\* This will create a fasta file of all the unique sequences to be processed.

- Removing the spaces

- Open the fasta file in a text editor and use search and replace ' ' to remove the spaces in the headers.

# Running the BLAST

Refer to the previous instructions on running a local BLAST.

Troubleshooting:

- If much fewer sequences are returned, there might be a problem with the database coverage.

-Try running -task blastn or reducing the identity cutoff in readsidentifier.

Retrieving the desired barcodes

Source: Code written by Amrita Srivathsan

- Open the final blast output in Excel and select the headers which have the desired taxonomic identities. Copy and paste them into a new column. Copy this column into a text editor and save this as a .txt file. Then change the directory to the one containing retrieve2.py.

- Usage: python retrieve2.py headers.txt input.fasta output.fasta

- Further filtering and quality control

- These steps help to ensure that there is no further secondary signal in the retrieved barcodes.

- Open the fasta file in Excel and ensure the first row is not empty.

- Type the following formula in the  $2^{nd}$  column: =INDEX(A:A,1+(2\*ROW()-2))

- Type the following formula in the  $3^{rd}$  column: =INDEX(A:A,1+(2\*ROW()-1))

- Apply these formulae to every row of data. This gives you all the headers in one column and all the sequences in another.

- Copy these cells and paste as text/values only.

-Split the headers by ";" using the Text to Columns function.

Eg. Merged\_0;100;SpecimenID=1234 will become:

Merged_0	100	SpecimenID=1234
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\* You can then filter by read count, as well as look at the ratios of the multiple signals in a sample (eg. if Sample 1234 had Merged\_0, Merged\_1, Merged\_2, etc. lines).



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

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Zootaxa 3946 (1): 125–132 www.mapress.com/zootaxa/

http://dx.doi.org/10.11646/zootaxa.3946.1.6

http://zoobank.org/urn:lsid:zoobank.org:pub:013418D0-B239-4837-9D9C-39616349AEF3

# *Ngirhaphium* Evenhuis & Grootaert from southern Thailand (Diptera: Dolichopodidae) with the description of a new species

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# Abstract

The genus *Ngirhaphium* Evenhuis & Grootaert, 2002 is reported for the first time from Thailand in particular from mangroves on the coast of the Andaman Sea in southern Thailand. Three species were found: *N. murphyi* Evenhuis & Grootaert, 2002, *N. sivasothii* Grootaert & Puniamoorthy, 2014 and *N. chutamasae* **sp. nov.** The latter species is described and illustrated and a key to all four known species is provided. COI barcode data showed that the new species is most closely related to *N. murphyi* with a genetic distance of 7%. The distance with the other species is 11 to 12%.

Key words: Dolichopodidae, Ngirhaphium, new species, mangrove, Thailand

# Introduction

The genus *Ngirhaphium* Evenhuis & Grootaert, 2002 is a genus of large dolichopodid species that occurs only in the front mangrove and along creeks in mangroves (Grootaert & Puniamoorthy 2014). Hitherto three species were known exclusively from Singapore and it is the first time that the genus is reported from another country.

Here we report on three species found in mangroves along the coast of the Andaman Sea in southern Thailand: *N. murphyi* Evenhuis & Grootaert, 2002, *N. sivasothii* Grootaert & Puniamoorthy, 2014 and a new species for science that is described, illustrated and barcoded.

# Material and methods

**Study sites and sampling techniques**. The present study is based on a survey of the marine dolichopodids in southern Thailand done by the first author (AS). Both Malaise traps and sweep netting techniques were used to collect fresh specimens in various mangroves in the provinces of Nakhon Si Thammarat, Songkhla, Pattani and Satun (Tammalang subdistrict) and Tarutao Island, all in southern Thailand. Terminology following Grootaert & Puniamoorthy (2014).

**Specimen storage**. The holotype and paratypes of the new species and other species are preserved in 70% ethanol to prevent the degradation of DNA and deposited in the collections of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkhla University, Hat Yai, Thailand (PSU). Voucher specimens are also stored in the collection of the Royal Belgian Institute of Natural Sciences, Brussels (RBINS).

**Genetic analysis.** Total DNA was extracted from a pair of middle legs. The remaining portions of the specimens sampled were kept as voucher in PSU Natural History Museum. The sample tissues were placed into 1.5 ml sterile tubes and pulverized by adding 50  $\mu$ l of tissue lysis buffer; eventually incubated at 65°C for 30 minutes. Subsequently, 2  $\mu$ l of Proteinase K was added and incubated at 60°C overnight (24 hr.). The mixture was agitated with 7  $\mu$ l of 8M potassium acetate for 5 minutes and incubated at -20°C for 30 minutes before extraction of the

aqueous supernatant. This procedure was repeated once before an equal volume of 95% ethanol was added to precipitate the DNA pellet. The pellet was washed subsequently in 70% ethanol and 30  $\mu$ l of TE buffer was applied to dissolve the DNA before storage at -20°C.

Universal primers amplifying portion of the mitochondrial cytochrome *c* oxidase I (COI) gene (LCOI1490 and HCO2198, Folmer *et al.* 1994) were as previously used by Grootaert & Puniamoorthy (2014). Thermocycling conditions were as follows: initial denaturation at 95°C for 3 mins, followed by 40 cycles of denaturation at 94°C for 1 min., annealing at 48°C for 1 min., and extension at 72°C for 1.5 min. A final extension of 5 mins at 72°C was used. Gel electrophoresis was performed to verify the PCR success in a 1% agarose gel using 5  $\mu$ l of the reaction mix. All PCRs were purified and sequenced commercially by First BASE Sequencing Company (Malaysia).

The evolutionary history was inferred using the Neighbor-Joining method (MEGA6, Tamura *et al.* 2013). The optimal tree with the sum of branch length = 0.20749235 is shown. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The analysis involved 13 nucleotide sequences. Codon positions included were 1st+2nd+3rd+Noncoding. All positions containing gaps and missing data were eliminated. There were a total of 494 positions in the final dataset.

# Observations

# Family DOLICHOPODIDAE

#### Subfamily RHAPHIINAE

#### Genus Ngirhaphium Evenhuis & Grootaert

Ngirhaphium Evenhuis & Grootaert, 2002: 310. Type species by original designation: Ngirhaphium murphyi Evenhuis & Grootaert, 2002.

**Diagnosis**. Medium to large sized species (4.5–8 mm) with a metallic green or blue ground-colour. Antenna very long in males, a little shorter in females. Arista apical, basal article long. Rostrum in male small with well-developed labellae. Large rostrum in female. Vertex excavated (cf. Sciapodinae).

Mid and hind coxae without exterior bristle. Femora with inconspicuous bristling. All tibiae with strong bristles. Fore leg in male with tarsomere 4 bearing an asymmetrical, apical dorsal forked protuberance (absent in females); terminal segment with a pair of normal claws and a thickened claw-like structure beneath the posterior claw. Females with the claws as usual, but the terminal segment bears a long dorsal protuberance. Mid and hind legs with tarsomeres 1–4 with an apical comb of spinules ventrally.

Wing with tip of  $M_{1+2}$  sharply bent upwards just before reaching the wing border and ending near tip of  $R_{4+5}$ .

#### Ngirhaphium chutamasae sp. nov.

(Figs 1–6)

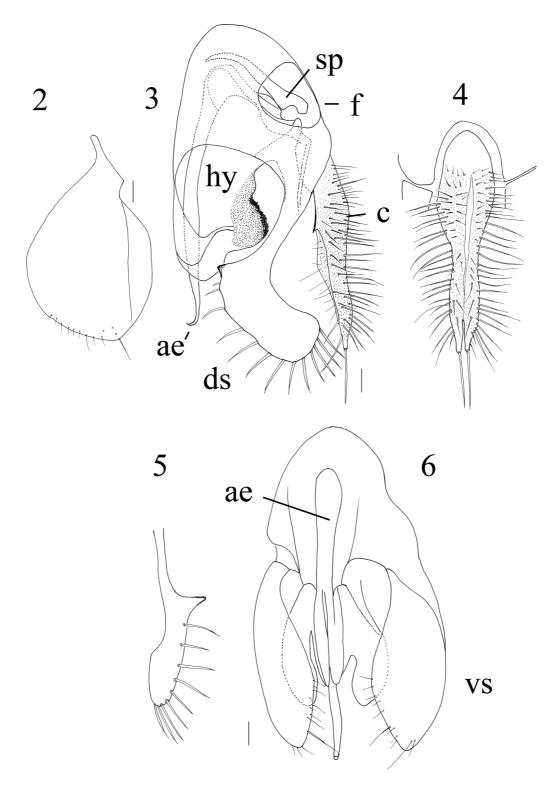
**Diagnosis.** A large species differing from the other *Ngirhaphium* species mainly in the structure of the male genitalia. Cercus in lateral view slightly shorter than dorsal surstylus. Cercus brown, tip pointed bearing a single yellow bristle. Dorsal surstylus brown, bordered with short, stout yellow bristles. Outer branch of apical fork on the fore tarsomere 4 slightly longer than inner branch.  $M_{1+2}$  with a short stub on apical bend.

**Material examined. HOLOTYPE** ♂, labelled: "THAILAND: Satun prov., Tammalang (6°32'21.05"N, 100°04'9.42"E), 3.x.2014 (reg. 34030, leg. P. Grootaert)" (PSU); **PARATYPE**: 1 ♂, Tammalang (6°32'21.05"N, 100°04'9.42"E), 6.viii.2014 (leg. A. Samoh) (RBINS).

**Etymology**. The species is dedicated to Associate Professor Dr. Chutamas Satasook, director of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkhla University, Hat Yai as a token for her dynamic support of our research.



**FIGURE 1**. *Ngirhaphium chutamasae* **sp. nov.**, male habitus; inset: apical tarsomeres on fore leg, showing the large fork-like extensions on tarsomere 4 and the additional claw-like structure on tarsomere 5 (photo: J. Brecko). Scale = 1 mm.



FIGURES 2–6. *Ngirhaphium chutamasae* **sp. nov.**, holotype male genitalia: 2. Left ventral surstylus; 3. Lateral view of genital capsule with left ventral surstylus removed; 4. Cerci dorsally; 5. Dorsal surstylus in ventral view; 6. Ventral view of genital capsule. Abbreviations: ae: aedeagus; c: cercus; ds: dorsal surstylus; f: foramen; hy: hypandrium; sp: sperm pump; vs: ventral surstylus. Scale = 0.1 mm.

**Description**. **Male**. Length body: 7 mm; wing: 5.6 mm. **Head**. Frons shining metallic green (not dusted). Face greenish brown in ground-colour; apex of face and clypeus yellowish brown in ground-colour, wide, nearly as wide as front of frons, parallel-sided, grey dusted with very short clypeus (less than 0.1 length of face). Eyes pass beyond border of face; eyes densely set with white hairs. Ocellar callus globular protruding from frons with 2 very long

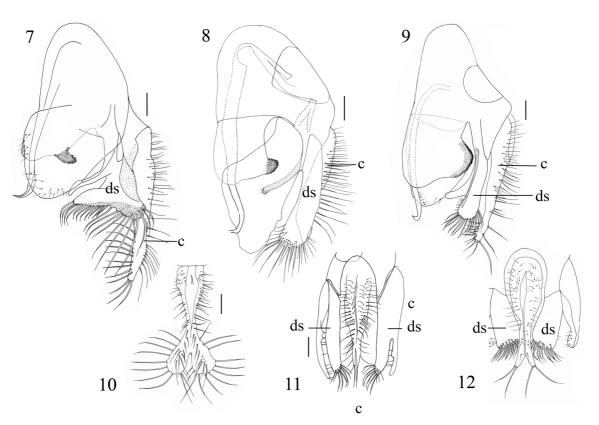
ocellars, directed backward, divergent. Vertical bristles long, half as long as ocellars, rather anteriad on frons at level of ocellar callus, close to eye border, long, black, directed forward and cruciate. Pair of long black postverticals directed backward and crossing. Postoculars above strong, black in single row, below white and mixed with very long white hairs below mouth; postcranium greenish in ground-colour but grey dusted. Palpus long, strap-shaped, yellowish with few short black hairs, no bristles. Labella brown with black hairs. Antenna very long, completely black. First segment long, 3X as long as second segment; second segment short, apically with crown of short black bristles. Third segment very long strap-shaped, laterally flattened, about 6X as long as width at base. Arista apical, apical article longer than basal article, gradually tapered towards tip. Length of scape: 0.52; pedicel: 0.13; postpedicel: 0.95; basal aristal article: 0.34; apical aristal article: 0.4 (in mm). Thorax and scutellum dark metallic green in ground-colour (bluish when seen from in front), covered with fine grey dusting. All hairs and bristles black. Pleura more densely grey dusted than mesonotum. Acrostichals biseriate, about 7 pairs, rows widening slightly behind. Presutural dorsocentrals multiseriate; 6 postsutural dorsocentrals: 4 short and 2 long prescutellars; 1 pair of strong scutellars. One long humeral with shorter bristle in front; 1 strong posthumeral, 2 strong notopleurals, 1 postsutural, 1 supraalar, 1 very strong postalar. Propleural bristles black, 6 short upper and 2 longer lower propleural bristles (lower one twice as long as upper). Legs (Fig. 1) yellow, all bristles black. All coxae greenish black in ground-colour, covered with fine greyish dusting. All trochanters brown. Tip of hind tibia annulated brown at tip. All tarsi yellowish, becoming darker towards tip. Apical tarsomeres completely black. Fore leg. Coxa with short black bristles. Fore femur slightly swollen on basal half; row of minute posteroventrals in apical half. Short preapical posterior bristle and 1 stronger anterior preapical bristle directed forward. Fore tibia with 4 strong ad, 4 strong pd and crown of 4 apicals. All tarsal segments densely set with black hairs and short black bristles. Tarsomere 4 with dorsal asymmetrical fork, extended over tarsomere 5; fork about 1.5X length of tarsomere; outer branch of fork slightly longer than inner branch, tips pointed (Fig. 1, inset). Terminal segment with pair of long normal claws and thicker claw-like structure beneath posterior claw. Two well-developed pulvilli and empodium present. Length of femur, tibia and tarsal segments (in mm): 2: 1.96: 0.98: 0.56: 0.28: 0.28: 0.28. Mid leg. Coxa with short bristles anteriorly; no exterior bristle. Mid femur as wide as fore femur; ventrally with inconspicuous bristles; 1 strong anterior preapical and 2 tiny posterior preapicals. Tibia with 6 ad, 6 pd (might be considered as dorsal), 8 longer av and crown of long apicals. Tarsomeres 1-4 ventrally at tip with pair of short spine-like bristles as well as comb of shorter black spinules. Apical tarsomere dorsally thickly set with long black squamiform bristles. Length of femur, tibia and tarsal segments (in mm): 2:3.08:1.68:0.84:0.77:0.35:0.42. Hind leg. Coxa bare. Hind femur thicker than mid femur, as wide as fore femur; ventrally almost bare; 1 strong anterior preapical, 1 fine posterior preapical. Tibia stronger bristled than mid tibia with 7 long av, 7 ad, 7 pd and crown of long apicals. Tarsomeres 1-4 ventrally at tip with pair of short spine-like bristles as well as comb of shorter black spinules. Length of femur, tibia and tarsal segments (in mm): 2:3.78:1.68:0.98:0.77:0.42:0.35. Wing mostly tinged brownish, but anteriorly between costa and R<sub>4+5</sub> with yellowish brownish tinge. Tp brown seamed. Veins dark brown, yellowish at base. M<sub>1+2</sub> sharply bent upwards and ending in costa closely near tip of  $R_{4+5}$ . Tp straight, about as long as apical part of  $M_{3+4}$ . Anal vein reaching wing border. Halter with white knob. Squama white with long white cilia. Abdomen shiny dark metallic green; tips and sides of tergites with greyish dusting. Sternites greyish dusted. Tergites densely set with quite long black bristles; hind-marginal bristles slightly longer than other bristles. Only tergite 5 with very long marginal bristles. Sternites with very short hairs except for longer marginals on sternite 4. Genital capsule black. Cercus brown, slightly shorter than dorsal surstylus (Fig. 3). Ventrally at base with black sclerotisation. Both cerci fused for almost entire length, only tips free (Fig. 4). Tip of cercus pointed, with single yellow apical bristle, dorsally set with long black bristles. Dorsal surstyli brown, much enlarged, forming clasper transverse on cercus, bordered with short, stout yellow bristles, shorter than surstylus is wide (Fig. 3). Ventrally with dark spur-like apex (Fig. 5). Ventral and dorsal surstyli not fused. Ventral surstylus yellowish, large, rounded (Fig. 2); tip with short hair-like bristles; inner carina running parallel to dorsal border. Hypandrium dorsally with large rounded black protuberance set with spinules (Fig. 3). Female. Indistinguishable from females of *N. sivasothii* (see Remarks section).

**Remarks**. The new species is morphologically almost identical to *N. sivasothii*, except for the larger forked extension on fore tarsomere 4 in male and the very different male genitalia. The outer branch of the apical fork on fore tarsomere 4 is slightly longer than the inner branch, the outer branch is slightly shorter than the inner branch in *N. sivasothii*.

The wing is brownish tinged and only the Tp is dark seamed. In N. sivasothii the wing is darker and the

longitudinal veins as well as Tp are generally black seamed. Vein  $M_{1+2}$  includes a short stub on the apical bend in the new species. Such a stub was never observed in the large populations of *N. sivasothii* in Singapore.

Presently females of the new species are indistinguishable from females of *N. sivasothii*. Both species have been collected together. For this reason, no females were included in the material examined section.



FIGURES 7–12. Lateral view genital capsule: 7. *Ngirhaphium sivasothii*; 8. *N. caeruleum*; 9. *N. murphyi*; dorsal view cerci: 10. *N. sivasothii*; 11. *N. caeruleum*; 12. *N. murphyi*. Abbreviations: c: cercus, ds: dorsal surstylus (modified after Grootaert & Puniamoorthy 2014). Scale = 0.1 mm.

# Ngirhaphium murphyi, Evenhuis & Grootaert

(Figs 9, 12)

*Ngirhaphium murphyi* Evenhuis & Grootaert, 2002: 310. Type locality: SINGAPORE: Kranji mangrove. *N. murphyi*: Grootaert & Puniamoorthy, 2014: 147 (figs 1–3, 4, 5, 18).

**Diagnosis**. A large species (5.7–7.3 mm), generally with clear wings. Mesonotum and tergites metallic green. Apical aristal article quite thick, nearly half as long as basal aristal article. Male with cerci longer than surstyli and thus the tips are visible outside the surstyli (Figs 9, 12).

**Material examined**. THAILAND: 1 ♂, 4 ♀, Satun province, Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 11.viii.2014 (leg. A. Samoh) (PSU)

**Remarks**. The pedicel is yellowish-brown in females and black in males. The pedicel is always black in both sexes in Singapore populations (Grootaert & Puniamoorthy 2014).

#### Ngirhaphium sivasothii Grootaert & Puniamoorthy

(Figs 7, 10)

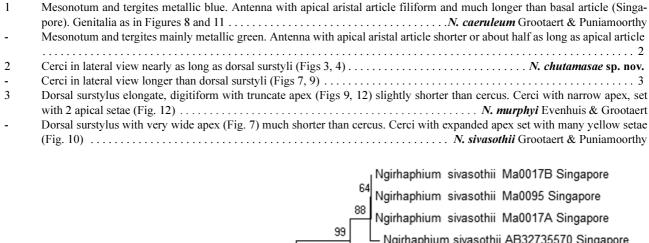
Ngirhaphium sivasothii Grootaert & Puniamoorthy, 2014: 150 (figs 6-8, 9-10, 17). Type locality: SINGAPORE: Semakau Island.

**Diagnosis**. A medium-sized species (4.5–5.5 mm), generally with dark infuscate wing and with longitudinal veins and Tp (posterior cross vein) brownish seamed. Mesonotum and tergites metallic green. Apical aristal article shorter, but nearly as long as basal article. Male with dorsal surstylus half as long as cerci, with a rectangular bend, set with very long bristles (Fig. 7). Cercus much longer than dorsal surstylus, tip wide, rounded, set with many long yellow bristles (Fig. 10). Outer branch of apical fork on the fore tarsomere 4 slightly shorter than inner branch.

**Material examined**. THAILAND: Satun province:  $1 \[3mm]{\circ}, 2 \[2mm]{\circ}$ , Tammalang (6°32'21.05" N, 100°04'9.42" E); 4  $\[3mm]{\circ}, 7 \[2mm]{\circ}$ , 6.viii.2014 (leg. A. Samoh); 1  $\[3mm]{\circ}, 2 \[2mm]{\circ}$ , 3.x.2014 (reg. 34030, leg. P. Grootaert & A. Samoh); 7  $\[3mm]{\circ}, 20 \[2mm]{\circ}$ , Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 12.viii.2014 (leg. A. Samoh); 2  $\[3mm]{\circ}, 1 \[2mm]{\circ}$ , Tanjong Po (6°36'57.43" N, 99°57'25.66" E), 3.x.2014 (leg. A. Samoh) (PSU).

**Remarks**. Some specimens had quite clear wings without the brown of black seams along the longitudinal veins and the Tp (posterior cross vein).

#### Key to males of Ngirhaphium



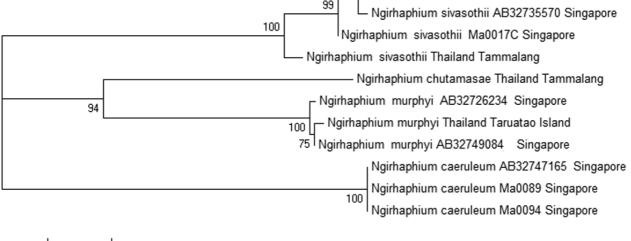


FIGURE 13. Neighbour-Joining tree of the COI barcodes of *Ngirhaphium* with bootstrap values indicated at the nodes. Scale of genetic distance is 1%.

#### Discussion

With three species, *Ngirhaphium* appears quite diverse on the coast along the Andaman Sea in southern Thailand. Although several mangroves along the coast of the South China Sea were investigated, we failed to collect any specimens. This might be due to differences in microhabitat that we failed to recognize, or simply that the genus is not present there.

0.01

*Ngirhaphium murphyi* and *N. sivasothii* also occur in Singapore. The geographic distance between the Singaporean populations and those in the Satun province (Tammalan, Tarutao Island) is about 800 km and the genetic distance between the two populations is less than 1% for *N. murphyi* and about 1% for *N. sivasothii* (Fig. 13). This is very low compared to another mangrove species, *Teuchophorus simplicissimus* Grootaert & Meuffels, which differed by 6.5% for COIb with a geographic distance of only 240 km between Singapore and Pulau Tioman (Lim *et al.* 2009).

*Ngirhaphium chutamasae* **sp. nov.** is most closely related to *N. murphyi* and they cluster with a bootstrap of 94 while the genetic distance is 7% (Fig. 13) The genetic distance between the new species and *N. sivasothii* and *N. caeruleum* is 11 % and 12 % respectively.

#### Acknowledgements

The authors are very grateful for the support of Associate Prof. Dr. Chutamas Satasook (Prince of Songkhla University, Hat Yai). The staff of the Nature Reserve at Tammalang is thanked for their help in the field. Our thanks also to Mr. Rueangrit Promdam and the entomology research unit members, Department of Biology, Faculty of Science, PSU for assistance with fieldwork, to Mr. Phuripong Meksuwan, Ms Sakiyah Morlor and Ms Bongkot Wichachoochert for assistance with the molecular work and highly appreciated suggestions, and to the Princess Maha Chakri Sirindhorn Natural History Museum staff for their help. This study was in part supported by funds from the National Research Universities grant (NRU), Government of Thailand. We also thank Dr. Brad Sinclair and two anonymous referees for many crucial comments.

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European Journal of Taxonomy 329: 1-40 https://doi.org/10.5852/ejt.2017.329 (cc) BY

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# **Research** article

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# Eight new species of marine dolichopodid flies of *Thinophilus* Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand

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Abstract. Eight new species of marine dolichopodid flies from southern Thailand belonging to the genus Thinophilus Wahlberg, 1844 are described and illustrated: Thinophilus boonrotpongi sp. nov., T. langkawensis sp. nov., T. minutus sp. nov., T. parmatoides sp. nov., T. parvulus sp. nov., T. spinatus sp. nov., T. spinatoides sp. nov. and T. variabilis sp. nov. A key is provided to the species of the Thai-Malay Peninsula.

Keywords. Marine Dolichopodidae, Thinophilus, peninsular Thailand.

Samoh A., Satasook C. & Grootaert P. 2017. Eight new species of marine dolichopodid flies of Thinophilus Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand. European Journal of Taxonomy 329: 1-40. https://doi.org/10.5852/ejt.2017.329

# Introduction

The present paper is part of a recent inventory of the marine dolichopodid flies from southern Thailand. In a previous survey (Grootaert & Meuffels 2001) 15 species belonging to seven genera of Dolichopodidae were found. Samoh et al. (2015) added the genus Ngirhaphium Evenhuis & Grootaert, 2002 with three species, resulting in 18 known species from southern Thailand to date. Only three species of Thinophilus have been recorded from peninsular Thailand until now: T. nitens Grootaert & Meuffels, 2001, T. parmatus Grootaert & Meuffels, 2001 and T. setiventris Grootaert & Meuffels, 2001.

# European Journal of Taxonomy 329: 1-40 (2017)

The genus *Thinophilus* Wahlberg, 1844 belongs to the subfamily Hydrophorinae Lioy, 1864 and is one of the most diverse groups of dolichopodid flies inhabiting coastal environments (Grootaert *et al.* 2015). They are adapted to and survive excellently in marine habitats such as front, mid and back mangroves, tide pools, mudflats, sandy beaches and rocky shores.

At the moment 31 species of *Thinophilus* are known from Southeast Asia. None of these species, however, correspond to the species reported in the present paper.

In the extreme northern part of the South China Sea, four species occur on the coast of continental China: *T. clavatus* Zhu *et al.*, 2006 (Hainan), *T. dongae* Grootaert *et al.*, 2015 (Shenzhen), *T. lamellaris* Zhu *et al.*, 2006 (Shenzhen) and *T. zhuae* Grootaert *et al.*, 2015 (Shenzhen). These robust species have not yet been reported from other parts of Southeast Asia and a key to these species can be found in Grootaert *et al.* 2015.

Various species were described in the past from Taiwan. Becker (1922) was the first to describe five species from Taiwan: *T. formosinus* Becker, 1922, *T. insertus* Becker, 1922, *T. integer* Becker, 1922, *T. seticoxis* Becker, 1922 and *T. tesselatus* Becker, 1922. In addition, Becker (1922) also reported *T. indigenus* Becker, 1902 from Taiwan, a species he had described earlier from Egypt. However, having examined the holotype from Egypt and compared it with the specimens from Taiwan, we doubt their conspecificity (Grootaert, unpubl.). Later, Parent (1935) reported *T. indigenus* Becker, 1902 from Port Dickson (peninsular Malaysia), but since he did not give any characteristics regarding the identification and because we have not found any specimens during our inventory that fit the description given by Becker's (1902), we consider this record as doubtful. Finally Parent (1941) added *T. hilaris* Parent, 1941, so that now seven species of *Thinophilus* are known from Taiwan. In fact none of these have been reported from the rest of the South China Sea and so they are provisionally considered as endemic to Taiwan.

*Thinophilus aequalichaetus* Parent, 1941 is the only species of *Thinophilus* reported from the Philippines (Luzon). Labelled 'Atimonan S.O. Luzon', it is probably a marine species since this locality is situated near the sea. We studied the holotype and paratype males, with missing heads, and found that there are a few characters typical of this species: the legs are yellow, including the fore coxa, but mid and hind coxae are black. Tarsomere 5 of the fore leg is brownish. The fore coxa is anteriorly set, with yellowish bristles and a few brown bristles at the tip. Fore, mid and hind femora lack ventral bristles. We consider *T. aequalichaetus* as a species *inquirenda* for the moment (Grootaert, unpubl.).

In 1935, Parent described eight species from Northeast Borneo (now Sabah, Malaysia): *T. amoenus* Parent, 1935, *T. chetitarsis* Parent, 1935, *T. ciliatus* Parent, 1935, *T. duplex* Parent, 1935, *T. egenus* Parent, 1935, *T. pallidipes* Parent, 1935, *T. valentulus* Parent, 1935 and *T. varicoxa* Parent, 1935. All these species seem to be endemic to Sabah for the moment and none of them correspond to the species of southern Thailand.

Up to now, only four species of *Thinophilus* have been reported from Indonesia: *T. androegenus* Hollis, 1964 and *T. phollae* Hollis, 1964, both described from Fort de Kock (now Bukittingi on Sumatra), *T. cuneatus* De Meijere, 1916 and *T. pectinipes* De Meijere, 1916, having both Wonosobo, Java as type locality (De Meijere 1916). All four are fresh water species and the descriptions do not fit those of our marine species from southern Thailand.

At the moment, only three marine species have been published from Singapore: *T. asiobates* Evenhuis & Grootaert, 2002, *T. longicilia* Evenhuis & Grootaert, 2002 and *T. murphyi* Evenhuis & Grootaert, 2002. Since they occur at the tip of the Malay Peninsula, the three species are included in the key given below.

Concerning the fauna of peninsular Malaysia, Parent (1935) described *T. peninsularis* based on one male and five females from Port Dickson on the coast of peninsular Malaysia, not far from Kuala Lumpur. In the description, Parent also included specimens from Langkawi Island, but did not indicate how many and whether they were males or females. Being sympatric, this species is of special concern in our study since it occurs very close to our study area. According to Parent's description (1935) it is a very small species of about 2 mm without particular characters, and it seems to be related to one of our new species.

In the present paper we describe an additional eight new species of *Thinophilus* found in mangroves along the seacoast from both sides of peninsular Thailand (Andaman Sea and Gulf of Thailand). The new species from the Andaman Sea side also represent the first records of *Thinophilus* for the Andaman Sea, which proves to be more diverse than the Gulf of Thailand, as will be demonstrated herein. A key is given for all of the presently known species from the Thai-Malay Peninsula.

# Material and methods

# Study sites and sampling techniques

This study was mainly conducted in eight provinces of peninsular Thailand namely, Chum Phon, Surat Thani, Nakhon Sri Thammarat, Songkhla, Pattani, Satun, Krabi and Phang Nga (Fig. 41). Sweep netting and Malaise trapping were used to collect fresh specimens of marine dolichopodids in various types of mangroves, tide pools and mudflats. Ethyl acetate was used to relax all specimens collected by sweep netting.

# Collection preservation and deposition

All specimens were preserved in 70% ethyl alcohol and stored in a refrigerator to prevent DNA degradation. All type material is deposited in the collections of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkla University (NHM-PSU), Hat Yai, Songkhla, Thailand, unless otherwise indicated. In addition, a few voucher specimens are also kept in the collections of the Royal Belgian Institute of Natural Sciences (RBINS), Brussels, Belgium.

# Revision of the oriental types of Thinophilus

The third author (P.G.) revised the material described by Becker (Museum für Naturkunde, Berlin; Deutsches Entomologisches Institut, Müncheberg) and de Meijere (Naturalis, Leiden) as well as the types deposited by Parent in the Muséum national d'Histoire naturelle (Paris) (Grootaert, in preparation).

# Terminology and abbreviations

Fly terminology is used as in Grootaert & Puniamoorthy (2014). The following abbreviations are used in text and figures:

- acr = acrostical bristles
- ad = anterodorsal bristles
- av = anteroventral bristles
- c = cercus
- dc = dorsocentral bristles
- ds = dorsal surstylus
- hy = hypandrium
- pd = posterodorsal bristles
- pv = posteroventral bristles
- T 1-5 = tarsomeres 1-5
- Tp = posterior cross vein (dm-Cu)
- vs = ventral surstylus

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Measurements are presented as mean values. Scales on drawings are 0.1 mm.

# Photography

A focus stacking technique (see Brecko *et al.* 2014) was used to photograph all specimens. The high resolution pictures were stacked using Zerene Stacker software. Scales on photos are 1 mm.

# Results

Class Insecta Linnaeus,1758 Order Diptera Linnaeus,1758 Superfamily Empidoidea Latreille, 1804 Family Dolichopodidae Latreille, 1809 Subfamily Hydrophorinae Lioy, 1864

Thinophilus Wahlberg, 1844

*Thinophilus* Wahlberg, 1844: 37. Type species: *Rhaphium flavipalpe* Zetterstedt, 1843 (monotypy). *Parathinophilus* Parent, 1932: 161. Type species: *Parathinophilus expolitus* Parent, 1932 (monotypy).

*Thinophilus boonrotpongi* sp. nov. <u>urn:lsid:zoobank.org:act:21C6EEB3-B179-432F-8D83-A863422A249D</u> Figs 1–5

#### Diagnosis

A medium-sized species with black fore coxa bearing long white bristles. Apical half of fore tibia pale, almost white with black tip. Tip of all apical tarsomeres black.



Figs 1–2. *Thinophilus boonrotpongi* sp. nov. 1. ♂, habitus. 2. ♀, habitus.

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# Etymology

This species is dedicated to Dr Singtoe Boonrotpong, promoter of the PhD thesis of the first author, in recognition of his help and support during the current project.

# Type material

# Holotype

THAILAND: ♂, Sai Thai, Muang, Krabi Province, 8°03′23.5″ N, 98°53′38.2″ E, sweep netting, 27 Feb. 2015, A. Samoh leg. (NHM-PSU).

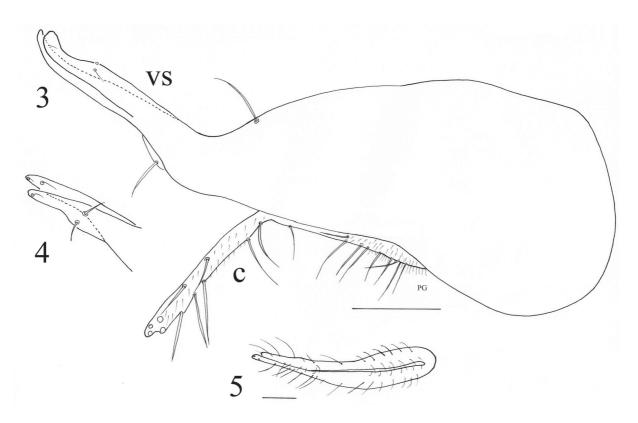
# Paratypes

THAILAND: 7  $\Im$ , 10  $\bigcirc$  , same collection dat os for holotype; 1  $\Im$ , 7  $\bigcirc$  , Khlong Phon, Khlong Thom, Krabi Province, 7°48'11.2" N, 99°10'11.9" E, sweep netting, 13 Jun. 2015, A. Samoh leg.; 1  $\Im$ , 1  $\bigcirc$ , Ban Bakan Tohtid, Langu, Satun Province, 6°47'29.8" N, 99°48'53.5" E, sweep netting, 3 Jun. 2015, A. Samoh leg.; 1  $\Im$  (with yellow femora), Ban Bakan Tohtid, Langu, Satun Province, 6°47'29.8" N, 99°48'53.5" E, sweep netting, 4 Jun. 2015, A. Samoh leg. (RBINS); 3  $\Im$ , 2  $\bigcirc$ , Bo Sane, Thappud, Phang Nga Province, 8°27'29.7" N, 98°36'17.8" E, sweep netting, 13 Feb. 2015, A. Samoh leg.

# Description

Male (Fig. 1) Length. Body 3.5 mm; wing 2.8 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. Face as wide as length of postpedicel. Clypeus about one-third of epistoma, protruding. A pair of long divergent black ocellars. Two very



**Figs 3–5.** *Thinophilus boonrotpongi* sp. nov., ♂, terminalia. **3**. Genital capsule, lateral view. **4**. Apex of surstylus, dorsal view. **5**. Cerci, dorsal view.

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short postocellars. A pair of convergent proclinate verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars. Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna brownish at tip and above, yellowish below. Arista dorsal, twice as long as antenna, brown, bare. Basal article short. Palpus yellowish to brown, with black bristly hairs. Proboscis dark brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 equally long dc in one row, preceded by a short bristle and a longer prescutellar outside the row. Scutellum with 2 marginals, without lateral hairs. Two short white upper propleural bristles and 2 longer lower propleural bristles.

LEGS. Brownish, but tibiae and tarsi pale. Fore coxa completely black; mid and hind coxae entirely black. All femora generally black. All tibiae with basal half brownish, becoming whitish towards tip. Fore tibia with black spot on tip ventrally. All tarsomeres whitish, but tip of terminal tarsomere black. Coxa anteriorly with long white bristles in apical half. Trochanter with long white bristles. Fore femur thickened in basal two-thirds. Ventrally at base with 2 rows of white bristles, longer than femur is wide, apical two-thirds with few short black bristles; with 3 strong equally long posterior preapical bristles. Fore tibia shorter than femur, ventral bristles short; posteroventral bristles of tibia on basal third longer than following bristles. Tarsomere 1 densely set with spine-like bristles. Mid coxa: exterior bristles white and longer than coxa; anterior bristles long and white. Mid femur thinner than fore femur; with row of black ventral bristles, longer at base. Mid tibia with a long anterodorsal at apical quarter; 2 dorsal and 2 pd; crown of apicals, ventral bristles longest. Hind coxa with short white exterior bristles. Hind femur a little thicker than mid femur; a long dorsal and anterodorsal bristle at apical third; row of black ventral bristles about as long as femur is wide. Hind tibia with 2 anterodorsal and 2 shorter dorsal bristles and a crown of long apicals. Hind tarsomere 1 long but shorter than tarsomere 2.

WINGS. Uniformly brownish tinged, without spots. Tp straight, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short, black. Sternites with short white bristles.

TERMINALIA (Figs 3–5). Phallus long, strap-shaped. Cerci pale brownish, with pale hairs; epandrium black. Cerci not fused (Fig. 5).

**Female** (Fig. 2) LENGTH. Body 3.6 mm long; wing 3 mm long. Larger than male.

BODY. Similar to male except following characters: clypeus  $\frac{1}{4}$  length of face, bulging; fore coxa with short white bristles only, fore femur with minute bristles, mid and hind femora also with minute ventral bristles; sternites with short white bristling.

# Distribution

Southern Thailand, only known from Andaman Sea coast.

# Remarks

*Thinophilus boonrotpongi* sp. nov. is quite unique in having a black fore coxa bearing long white bristles, combined with the apical half of the fore tibia almost white with a black apex. All apical tarsomeres are also darkened. Only *T. nitens* Grootaert & Meuffels, 2001 has white bristles on the fore coxa, with a single black bristle among them, but the fore coxa itself is yellow. Among the material examined

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was one male specimen with all femora and tibiae yellow that we attribute to *T. boonrotpongi* sp. nov. The tarsi are yellowish and not whitish (cf. Fig. 1). Other characters, such as the fore femur with long white soft bristles at the base, the general bristling of the legs and the male genitalia, also suggest that it represents *T. boonrotpongi* sp. nov. A future molecular analysis should ascertain if there is a genetic difference.

# Thinophilus langkawensis sp. nov.

urn:lsid:zoobank.org:act:EE41F65A-641F-4D96-87FE-9228A57D4155

Figs 6-11

# Diagnosis

A large species. Antenna completely yellow. Tibiae and tarsomeres completely yellowish white. Hypopygium elongate, more than half length of abdomen. Cerci in male reaching almost to thorax. Surstyli are movable and out-folding with a veil-like membrane.

# Etymology

The specific epithet refers to the island of Langkawi (Malaysia), where the species was found for the first time.

# **Type material**

# Holotype

THAILAND: ♂, Ko Tarutao, Molae Bay, Satun Province, 6°40′21.0″ N, 99°38′20.9″ E, sweep netting, 9 Jan. 2015, A. Samoh leg. (NHM-PSU).

# Paratypes

THAILAND: 5 33, 7 99, same collection data as for holotype (1 3 and 1 9 in RBINS).

# Description

Male (Fig. 6) Length. Body 6.4 mm; wing 5 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. Face twice as wide as length of postpedicel. Clypeus a third of length of face. Ocellar tubercle pronounced but sunken between the eyes, not surpassing eye borders (Fig. 6). A pair of long divergent black ocellars. No postocellars. A pair of convergent proclinate verticals, a little shorter than ocellars. Vertex excavated; postcranium metallic green. Two converging postverticals, stronger and longer than, and not in row with, upper postoculars. Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna yellow; pedicel and postpedicel hardly darkened dorsally. Arista dorsal, 2.5 times as long as antenna, not pubescent. Basal article short, yellowish brown; arista white, base a little browned. Palpus yellow, with short white bristly hairs. Proboscis brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 5 equally long dc, prescuttelar a litle longer and dc row preceded by a short bristle. Scutellum with 2 marginals and a short lateral bristle. Four short white propleurals above and 7 longer white propleural bristles below.



**Fig. 6.** *Thinophilus langkawensis* sp. nov.,  $\mathcal{C}$ , habitus.

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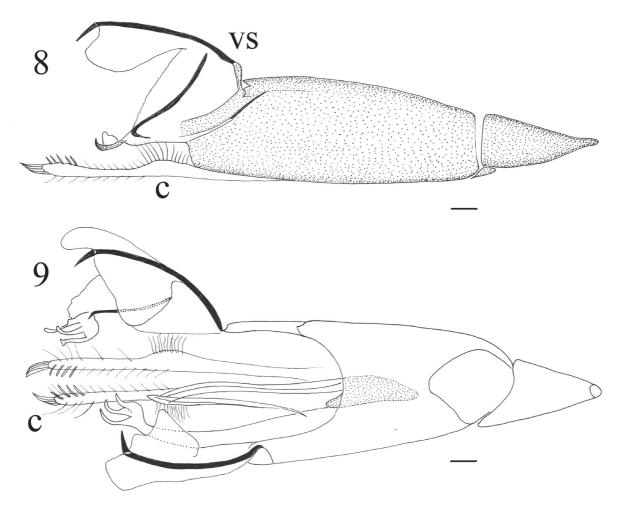
**Fig. 7.** *Thinophilus langkawensis* sp. nov., ♀, habitus.

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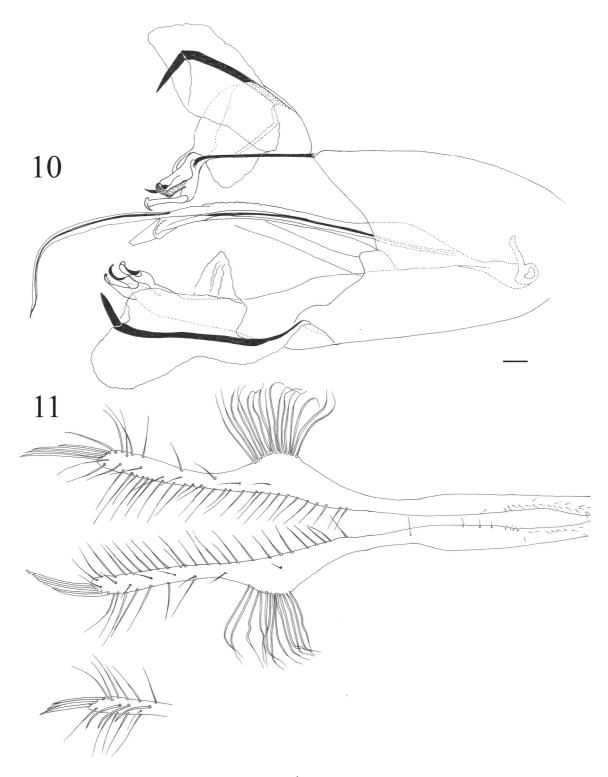
LEGS. Yellowish white including all tarsomeres. Fore coxa black on basal two-thirds, yellowish on apical third; mid and hind coxae brownish, apices pale. Fore coxa anteriorly with short white bristles. Trochanter bare. Fore femur narrower than mid femur. Ventrally almost bare, except for some minute white hairs; 2 short posterior preapical bristles. Fore tibia shorter than femur, with only minute ventral bristles. Mid coxa with a long, black exterior bristle near middle, with short, white anterior bristles at tip. Mid femur wider than fore femur; ventrally with an anterior row of 3 short brown bristles and a posterior row of 5 bristles. Mid tibia as long as femur, with 3 short ad, 2 longer ad and 2 pd. Hind coxa with black exterior bristle and minute white anterior bristles. Hind femur only a little wider than mid femur; ventrally on apical <sup>2</sup>/<sub>3</sub> with a row of long white bristles, twice as long as femur is wide; in addition a few minute ventral bristles on basal third; 2 long black ad bristles on apical third. Hind tibia with 3 ad, 2 very long pd; a row of short black pd on basal third as long as tibia is wide; 2 somewhat recurved ventral bristles at basal third.

WINGS. Clear, without spots. Tp straight, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites with short pale hairs.



**Figs 8–9.** *Thinophilus langkawensis* sp. nov., ♂, terminalia. 8. Genital capsule, lateral view. 9. Genital capsule, dorsal view.



**Figs 10–11.** *Thinophilus langkawensis* sp. nov.,  $\mathcal{Z}$ , terminalia. **10**. Genital capsule and surstyli, ventral view. **11**. Detail of apex of cerci, dorsal view.

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TERMINALIA (Figs 8–11). Elongate, more than half length of abdomen, with surstyli reaching tip of sternite 3 but cerci almost reaching to base of thorax. Cerci pale yellowish (Fig. 6), ventrally not fused. Apex cercus with remarkable pattern of bristling (Fig. 11). Surstylus movable, connected by a veil-like membrane to epandrium, suspended by black, rod-like structures. Phallus long, strap-shaped, but not coiled (Fig 10). Epandrium elongate, brown.

**Female** (Fig. 7) LENGTH. Body 6.4 mm long; wing 5.6 mm long.

BODY. Stouter than male, otherwise similar except following characters: hind femur lacking long white ventral bristles; sternites with minute pale bristling.

#### Distribution

Southern Thailand and northern Malaysia (Andaman Sea coast).

#### Remarks

The male of this robust species with yellow legs has very long terminalia, which in rest position are partly hidden in a cavity formed by the sternites 4 to 6. When the terminalia are extended, the surstyli move and open a veil-like lined cavity (Figs 8, 10). This phenomenon was not previously observed in *Thinophilus*. This large species was found on the adjacent islands of KoTarutau in Thailand and Langkawi Island in Malaysia.

#### Thinophilus minutus sp. nov.

urn:lsid:zoobank.org:act:150A1E50-9F4E-466B-9765-2BF976ACECE4 Figs 12–15

### Diagnosis

A small species with completely yellow antenna, yellow fore coxa, brown mid and hind coxa and legs further completely yellow. Fore tibia without a ventral row of spine-like bristles. Only mid and hind femur with distinct black ventral bristles.

### Etymology

The specific epithet refers to the small size of the species.

### **Type material**

### Holotype

THAILAND: ♂, Ban Laem Son, Langu, Satun Province, 6°56′27.9″ N, 99°42′12.4″ E, sweep netting, 27 Feb. 2015, A. Samoh leg. (NHM-PSU).

#### **Paratypes**

THAILAND: 1 Å, same collection data as for holotype; 1 Å, Phanang Tak, Muang, Chumphon Province, 10°30′23.9″ N, 99°13′55.6″ E, sweep netting, 17 Feb. 2015, A. Samoh leg.; 1 Å, Bang Yai, Bang Nai Si, Takuapa, Phang-Nga Province, 9 Feb. 2015, A. Samoh leg.

#### Description

Male (Fig. 12) LENGTH. Body 2.4 mm; wing 2 mm.

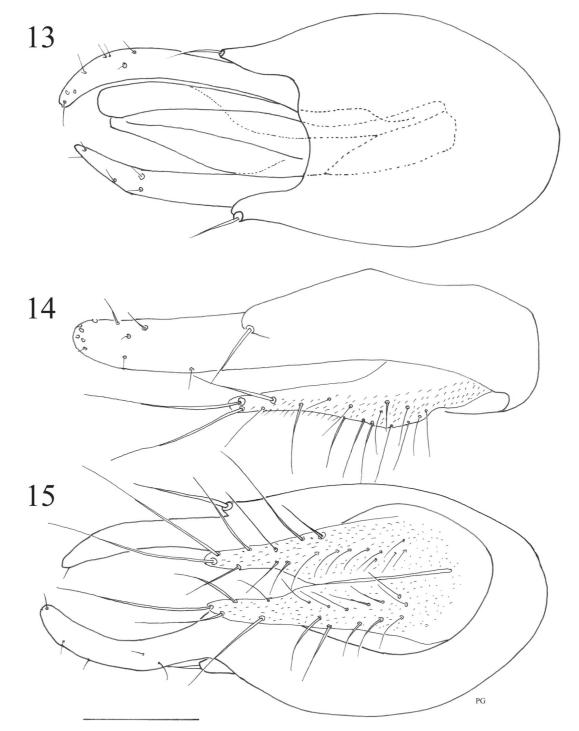
HEAD. Frons and face with shiny dark metallic green ground colour. A pair of long, divergent, black ocellars. Two very short postocellars. A pair of convergent, proclinate, long verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars. Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna pale brownish. Arista dorsal, 2.5 times as long as antenna, shortly pubescent. Basal article short. Palpus yellow, with short, black bristly hairs, only anteriorly. Proboscis dark brown.



**Fig. 12.** *Thinophilus minutus* sp. nov., ∂, habitus.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 equally long dc in one row, preceded by a short bristle and prescutellar outside the row and hardly longer than preceding bristles. Scutellum with 2 marginals, without lateral hairs. Three short lower pale brownish propleural bristles.

LEGS. Yellow including all tarsomeres. Fore coxa yellowish white; mid and hind coxae entirely brownish. Fore coxa anteriorly with short brown bristles. Trochanter bare. Fore femur club-shaped, a



**Figs 13–15.** *Thinophilus minutus* sp. nov., ∂, terminalia. **13**. Genital capsule, ventral view. **14**. Genital capsule, lateral view. **15**. Genital capsule, dorsal view.

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little thickened in basal half, apical half thin. No ventral bristles; 3 distinct posterior bristles on apical third. Fore tibia shorter than femur, no ventral bristling. First tarsomere densely set with spine-like bristles. Mid coxa with a long black exterior near middle and a long anterior bristle at tip. Mid femur slightly thinner than fore femur; with row of short ventral bristles in basal half. Mid tibia with a short ad and pd in basal quarter and a short ad and pd near middle; a crown of short apical bristles. Hind coxa with a black exterior bristle. Hind femur wider and longer than mid femur; short ventral bristles, short, upright anterior bristles near middle. Hind tibia with 1 ad and 2 dorsal bristles, a crown of long apicals.

WINGS. Yellowish brown, without spots. Tp straight, brownish seamed, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites with short brown hairs.

TERMINALIA (Figs 13–15). Phallus long, strap-shaped (Fig. 13). Cercus whitish, with long brown apical bristles (Figs 14–15), epandrium brown.

## Female

Unknown

## Distribution

Southern Thailand (Andaman Sea and Gulf of Thailand).

## Remarks

*Thinophilus minutus* sp. nov. is quite unique among *Thinophilus* in southern Thailand by having only a few distinct bristles on the legs. Only mid and hind femora have distinctly longer ventral bristles. It is similar to *T. peninsularis* Parent, 1935, a sympatric species that also exhibits only a few distinctive characters on the legs. The latter species, however, has a dorsal bristle on the basal quarter of the fore tibia, lacking in *T. minutus* sp. nov. Further, it has the fore coxa darkened on the basal two-thirds and the apical tarsomere darkened as well. The fore coxa and even the apical tarsomere of all legs are yellow in *T. minutus* sp. nov. Finally, in *T. peninsularis* the first tarsomere of the fore leg is as long as the following tarsomeres together, while in *T. minutus* sp. nov. the first tarsomere is half as long as the following four tarsomeres together. Both species share a brownish tinged wing. In *T. minutus* sp. nov. the Tp and M are brownish seamed.

*Thinophilus minutus* sp. nov. should also be compared with *T. dongae* Grootaert *et al.*, 2015, known from southern China. The latter species also has yellow fore coxae, no ventral bristles on the fore femur, no ventral spinules or bristles on the fore tibia. It has, however, the apical tarsomere of all legs black and mid and hind femora without ventral bristles. In *T. minutus* sp. nov. all tarsomeres are yellow and the mid and hind femora have short but distinct bristles. Both species are likely related in a species-group characterized by the similar shape of the cerci and surstyli.

## Thinophilus parmatoides sp. nov.

urn:lsid:zoobank.org:act:74D863DC-E1F0-4BF9-80FC-8F5339E26D42 Figs 16, 18-20

### Diagnosis

A medium-sized species with a shield-like protuberance on mid tarsomere 2. Mid femur with a cluster of about 10 short spine-like ventral bristles at base.

# Etymology

The specific epithet refers to the resemblance with *T. parmatus* Grootaert & Meuffels, 2001, also described from southern Thailand.

# **Type material**

# Holotype

THAILAND: ♂, Pak Phanang Tawantok, Pak Phanang, Nakhon Sri Thammarat Province, 8°24′09.4″ N, 100°11′29.9″ E, sweep netting, 30 Apr. 2015, A. Samoh leg. (NHM-PSU).

# Paratypes

THAILAND: 7  $\Im \Im$ , 10  $\Im \Im$ , same collection data as for holotype (2  $\Im \Im$ , 2  $\Im \Im$  at RBINS).

## Description

Male (Fig. 16) Length. Body 2.6 mm; wing 2.4 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. Face above as wide as length of postpedicel, near middle half as wide as postpedicel. A pair of long divergent black ocellars. Two very short postocellars. A pair of minute verticals at level of ocellar tubercle. Vertex a little sunken. A pair of minute postverticals. Four black upper postoculars, followed by a row of yellowish uniseriate lower postoculars. Antenna yellowish; only postpedicel dusky above. Arista subdorsal, 3.5 times as long as antenna, brown, with short pubescence. Basal article very short. Palpus yellowish brown, with a few fine black bristles along sides, centrally only minute bristles.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 short dc of equal length, prescutellar twice as long as preceding dc. Scutellum with 2 long crossing marginals, and a short lateral bristle. No upper propleurals and a few very short lower propleurals.

LEGS. Yellow, but fore coxa completely black, densely set with black bristles; mid and hind coxae brown. Fore and mid trochanters yellow, ventrally brown. Fore femur a little wider than mid femur, especially on basal half; ventrally near base with a few short bristles. Fore tibia longer than femur, with a ventral row of bristles, over entire length, all longer than tibia is wide; bristles near middle longest. Mid coxa with a long, black exterior bristle, half as long as coxa is high; anterior bristles very dense, black. Mid femur with spindle-shaped base; at base a cluster of about 10 black bristles (shorter than femur is wide). Mid tibia much longer than femur; without prominent bristles; ventrally in apical quarter with long hair-like bristles. Mid tarsomere 2 bearing a black shield-like dorsal extension; tarsomere 3 shorter than tarsomere 2, white (Fig. 16). Hind coxa with black exterior bristle. Hind femur a little spindle-shaped at base; ventrally in apical half with only 2 short black bristles. Hind tibia with a short ad near middle.

WINGS. Brownish tinged, without spots. Tp straight, longer than apical part of  $M_{3+4}$ . Anal vein not reaching wing margin.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites 2 and 3 with minute hairs; sternite 4 with a few longer black apical bristles.

TERMINALIA (Figs 18–20). Phallus long, strap-shaped. Cerci pale brownish, with pale hairs, dorsally fused (Fig. 20); surstyli and epandrium a little darker than cerci.

## Female

LENGTH. Body 2.9 mm long; wing 2.6 mm long.

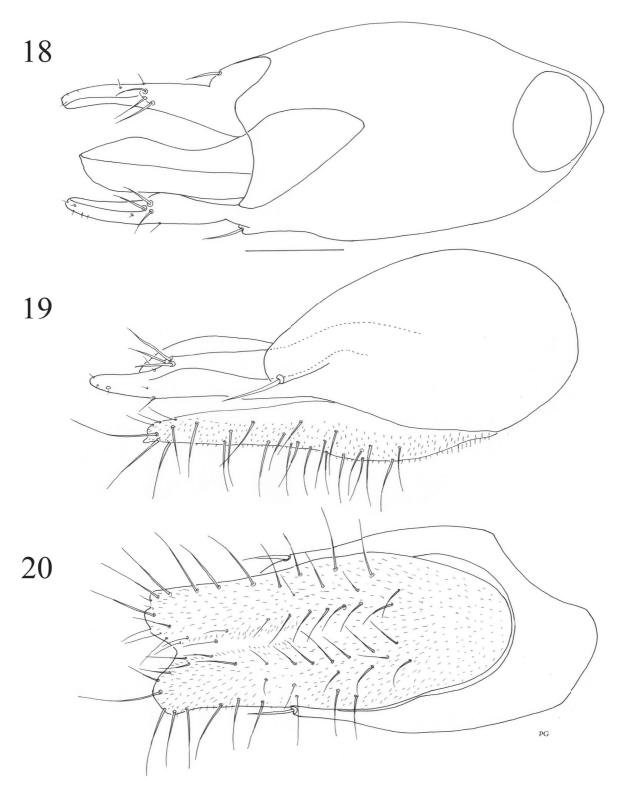
BODY. Similar to male, except for following characters: mid femur without cluster of ventral bristles at base, mid tarsomere 2 without shield-like protuberance.



**Fig. 16.** *Thinophilus parmatoides* sp. nov.,  $\mathcal{E}$ , habitus.



Fig. 17. Thinophilus parmatus Grootaert & Meuffels, 2001, *A*, habitus.



Figs 18–20. *Thinophilus parmatoides* sp. nov., ♂, terminalia. 18. Genital capsule, ventral view. 19. Genital capsule, lateral view. 20. Cerci, dorsal view.

## Distribution

Southern Thailand (Gulf of Thailand).

#### Remarks

This species is similar to *T. parmatus* in having a black shield-like protuberance on tarsomere 2 of the mid leg. There are a few black bristles at the base of the fore femur, a thick tuft of black bristles at the base of the mid femur, long hair-like bristles on the tip of the mid tibia and only short ventral bristles on the hind femur. In *T. parmatus*, there is a single long bristle at the base of the fore femur, the mid femur has only 4 thin bristles at its base and the hind femur has longer bristles in the apical half. The shield on tarsomere 2 of the mid leg is rounded in *T. parmatoides* sp. nov., but elongated in *T. parmatus* (Fig. 17). The shape of the male genitalia is very similar in both species.

#### Thinophilus parvulus sp. nov.

urn:lsid:zoobank.org:act:96F9E2AB-6AC4-43FF-99DE-CB949D3DAE00

Figs 21-24

#### Diagnosis

A small species with fore tibia bearing 1 short and 1 long black posterodorsal bristle near base.

#### Etymology

The species name is derived from the Latin 'parvulus', referring to the very small size of the species.

#### **Type material**

#### Holotype

THAILAND: ♂, Muang, Pattani Province, Prince of Songkhla University, Pattani campus, 6°53′04.9″N, 101°14′10.1″E, Malaise Trap, 11 Apr. 2015, A. Samoh leg. (NHM-PSU).

#### Description

Male (Fig. 21) LENGTH. Body 1.8 mm; wing 1.7 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. Face at narrowest point wider than postpedicel. Clypeus about a quarter as long as face. A pair of long divergent black ocellars. No postocellars. A pair of convergent verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars. Postoculars uniseriate, black above and white below. Antenna brownish. Arista dorsal, 3 times as long as antenna, brown, bare. Basal article short. Palpus yellow, with pale bristly hairs. Proboscis dark brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 dc, anterior 3 dc equally long, prescutellar twice as long. Scutellum with 2 marginals, without lateral hairs. Two very short lower white propleurals.

LEGS. Yellow, tarsomeres 4 and 5 brown. Fore coxa yellowish white, mid and hind coxa entirely brown, extreme tips yellowish. Fore coxa anteriorly with short white bristles. Trochanter bare. Fore femur a little thickened in basal half. Anteroventrally with a row of whitish to pale brownish, long, hair-like bristles, up to three times as long as femur is wide; a little coiled at tip and with a posteroventral row of white bristly hairs, also 3 times as long as femur wide. Fore tibia as long as femur, with 2 remarkable posteroventral bristles in basal half. Mid coxa without exterior bristle. Mid femur thickened in basal <sup>2</sup>/<sub>3</sub>, a little thicker than fore femur; with a row of 4 brownish ventral bristles in basal third, half as long as

femur is wide, anteriorly with row of 4 tiny preapicals; a stronger preapical pv. Mid tibia with a short ad and pd. Hind coxa without exterior bristle. Hind femur thickened in basal half, a little thicker than mid femur; double row of pale ventral bristles in apical half, as long as femur is wide, dorsally near base with a few erect bristles, anteriorly with 2 fine preapical bristles, posteriorly with 1 preapical bristle. Hind tibia with a row of ventral bristles, near middle as long as tibia is wide.

WINGS. Without spots. Tp straight, apical part of  $M_{3+4}$  2 times as long as Tp. Anal vein not reaching wing margin.

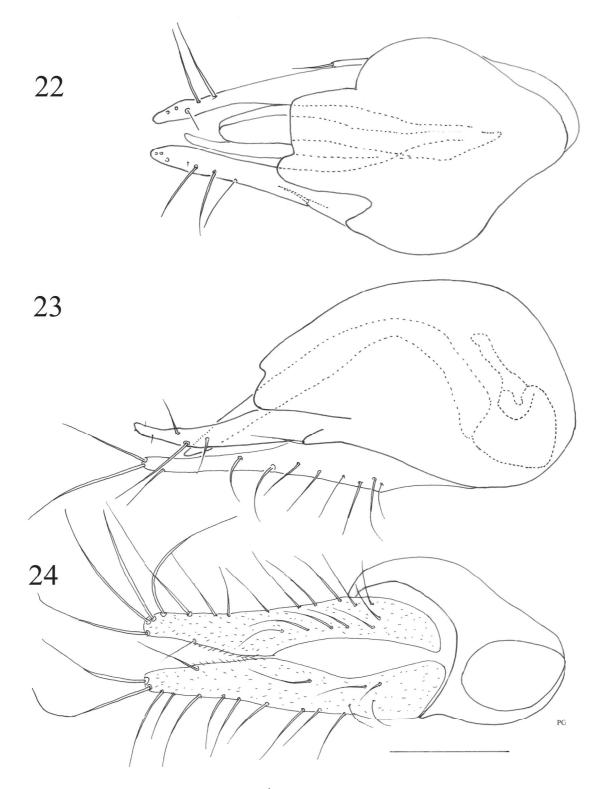
ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short and pale. Sternites with brownish, inconspicuous bristles.

TERMINALIA (Figs 22–24). Phallus long, strap-shaped. Cerci yellowish, not fused and with long apical bristles (Figs 23–24).

**Female** Unknown.



Fig. 21. Thinophilus parvulus sp. nov., ♂, habitus.



**Figs 22–24.** *Thinophilus parvulus* sp. nov., ♂, terminalia. **22**. Genital capsule, ventral view. **23**. Genital capsule, lateral view. **24**. Genital capsule, dorsal view.

## Distribution

Southern Thailand (Gulf of Thailand).

# Remarks

*Thinophilus parvulus* sp. nov. is a very small species characterized by the yellowish white fore coxa and the 2 long posteroventral bristles near the base of the fore tibia.

## *Thinophilus spinatoides* sp. nov. urn:lsid:zoobank.org:act:6B1C7B5E-A676-4A5D-A67A-07CF3E6356D1 Figs 25–29

## Diagnosis

A medium-sized species with very long yellow legs. Both male and female with a set of 4–5 long, stiff brown ventral bristles on fore femur. Fore femur spindle-shaped, basal quarter much dilated. Fore tarsomere 1 very long and slender, twice as long as fore tibia. Tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 widened, black.

# Etymology

The specific epithet refers to the resemblance with *T. spinatus* sp. nov., also described from southern Thailand.

## **Type material**

## Holotype

THAILAND: ♂, Bakan Tohtid, Langu, Satun Province, 6°47′29.8″ N, 99°48′53.5″ E, sweep netting, 3 Jun. 2015, A. Samoh leg. (NHM-PSU).

# Paratypes

THAILAND: 2  $\bigcirc \bigcirc$ , 1  $\bigcirc$ , same collection data as for holotype.

# Description

Male (Fig. 25) LENGTH. Body 4.5 mm; wing 3.8 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. A pair of long divergent black ocellars. No postocellars. A pair of tiny proclinate verticals at level of front ocellars. Postcranium dark metallic green. Postverticals not differentiated from upper postoculars. Upper postoculars uniseriate, short, black; with a few yellow lower postoculars. Antenna yellowish. Arista dorsal, 2.5–3 times as long as antenna, brown, not pubescent. Basal article short, brown; rest of arista paler. Palpus yellow, with few black bristly hairs. Proboscis dark brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 7 rather short dc, gradually growing longer toward scutellum, ending in a very long prescutellar. Scutellum with 2 long marginals with a tiny hair at outside. 2 short black propleural bristles.

LEGS. Yellow, with apical 2 tarsomeres of all legs black. Fore coxa with basal quarter darkened; mid and hind coxae black, tip yellow. Coxa anteriorly with a short bristle near base and a long bristle at basal third. Trochanter with short white bristles. Fore femur club-shaped, very thickened in basal quarter; apical <sup>3</sup>/<sub>4</sub> very thin. Ventrally with 4 long black bristles; longest bristle nearly twice as long as femur is

wide; others shorter. Fore tibia much longer than femur, without ventral bristling. Fore tarsomere 1 very long and slender, twice as long as fore tibia. Tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 widened, black. Mid coxa with a short black exterior bristle above middle; anterior bristles short, black. Mid femur ventrally without bristles; no preapical av. Mid tibia longer than mid femur, with a crown of short apical bristles and 2 minute ad. Mid tarsomere 1 almost twice as long as following tarsomeres. Hind coxa without exterior bristle. Hind femur without ventral bristles; no preapical anterodorsal bristles. Hind tibia with 2 short ad and crown of apical bristles. Hind tarsomere 1 a little longer than tarsomere 2.

WINGS. Uniformly yellowish tinged, without spots. Tp straight, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.



**Fig. 25.** *Thinophilus spinatoides* sp. nov.,  $\mathcal{J}$ , habitus.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short, black. Sternites without bristles, except sternite 4 with tuft of short black bristles.

TERMINALIA (Figs 27–29). Phallus long, strap-shaped. Cerci pale brownish, with pale hairs, dorsally fused (Fig. 29).

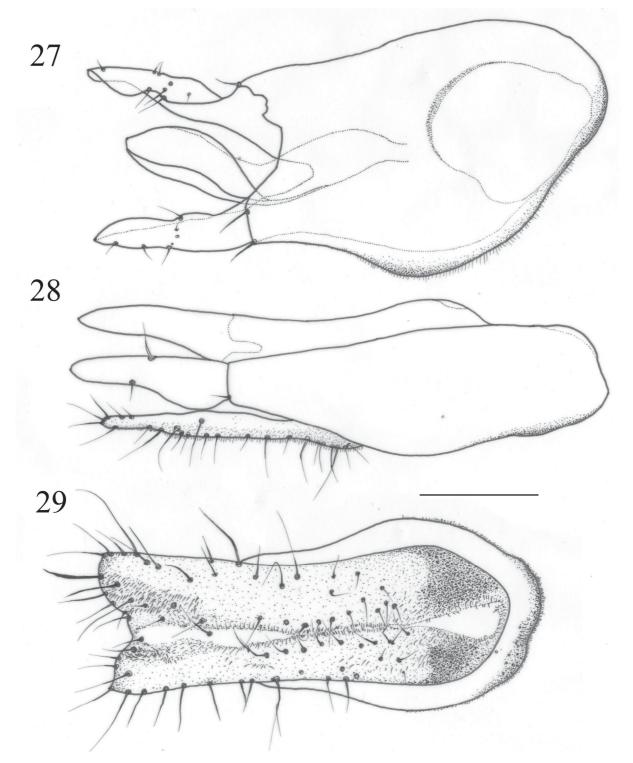
#### Female (Fig. 26)

LENGTH. Body 4.5 mm long, wing 4.2 mm long.



**Fig. 26.** *Thinophilus spinatoides* sp. nov.,  $\mathcal{Q}$ , habitus.

BODY. Similar to male except for following characters: fore femur basally not so strongly swollen as in male and with 5 strong black ventral bristles up to 3 times as long as femur is wide; tarsomere 1 of fore and mid legs more than twice as long as following tarsomeres together; sternites 3, 4, and 5 with pale bristles.



Figs 27–29. *Thinophilus spinatoides* sp. nov., ♂, terminalia. 27. Genital capsule, ventral view. 28. Genital capsule, lateral view. 29. Genital capsule, dorsal view.

## Distribution

Southern Thailand (Andaman Sea coast).

## Remarks

*Thinophilus spinatoides* sp. nov. is particular in that it has the fore femur with the basal quarter very spindle-shaped and dilated. It is less dilated in *T. spinatus* sp. nov. Fore tibia much longer than fore femur; shorter in *T. spinatus* sp. nov. Fore tibia slender and without ad in male, present in female; fore tibia stouter and with 2 long ad in *T. spinatus* sp. nov. Fore tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 much widened, black. Fore tarsomere 3 has the same pale yellowish colour as tarsomeres 1 and 2. Tarsomeres 4 and 5 black, not widened in *T. spinatus* sp. nov. Only base of fore coxa brown; basal <sup>2</sup>/<sub>3</sub> of fore coxa brown in *T. spinatus* sp. nov. Lower postocular bristles yellow; black in *T. spinatus* sp. nov. Anal vein distinct in basal <sup>2</sup>/<sub>3</sub>; anal vein not distinct at all in *T. spinatus* sp. nov.

#### Thinophilus spinatus sp. nov.

urn:lsid:zoobank.org:act:14EBE5D8-334A-4838-9510-8EBB664BC0BD

Figs 30–35

## Diagnosis

A medium-sized, slender-legged species with yellow legs, but fore coxa black except for apical third. The femora are spindle-shaped and the fore femur in male as well as in female bear long, brown spine-like bristles.

## Etymology

The specific epithet refers to the ventral bristles on the fore femur that are present in both male and female.

### **Type material**

## Holotype

THAILAND: ♂, Phang Nga Province, Muang, Bang Phat, 8°21′48.8″ N, 98°34′38.8″ E, Malaise trap, 13 Feb. 2015, A. Samoh leg. (NHM-PSU).

### Paratypes

THAILAND: 1  $\stackrel{\frown}{\supset}$ , 1  $\stackrel{\bigcirc}{\rightarrow}$ , same collection data as for holotype.

## **Additional material**

SINGAPORE: 1  $\bigcirc$ , Sarimbun (SR3), mangrove, 21 May 2014, J (leg. J. Puniamoorthy; Lee Kong Chian Natural History Museum, Singapore).

### Description

Male (Fig. 30) LENGTH. Body 4.3 mm; wing 3.75 mm.

HEAD. Frons and face with shiny dark metallic green ground colour. Face half as wide as length of postpedicel. Clypeus about one third of epistoma, hardly protruding. A pair of long divergent black ocellars. No postocellars. A pair of tiny proclinate verticals at level of front ocellars. Postcranium dark metallic green. Postverticals not differentiated from upper postoculars. Upper and lower postoculars uniseriate, short, black, with a few white bristles behind mouth. Antenna pale brownish. Arista dorsal, 2.5–3 times as long as antenna, brown, not pubescent. Basal article short, brown; rest of arista paler. Palpus yellow, with few black bristly hairs. Proboscis dark brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 7 rather short dc, gradually growing longer toward scutellum, ending in a very long prescutellar. Scutellum with 2 long marginals with a tiny hair at outside. Two short black propleural bristles.

LEGS. Yellow but sometimes pale brownish; apical tarsomere 2 of all legs brownish. Fore coxa black, but apical third yellowish brown; mid and hind coxae entirely black. Coxa anteriorly with a short bristle near base and a long bristle at apical third. Fore femur club shaped, thickened in basal half, apical half thin. Ventrally with 4 long black bristles; longest bristle twice as long as femur is wide. Fore tibia about as long as femur, without ventral bristling; tarsomere 1 much longer than following tarsomeres together. Mid coxa with a tiny black exterior bristle near middle; anterior bristles very short, black. Mid femur ventrally without bristles; no preapical av. Mid tibia as long as mid femur; with a crown of short apical bristles; 2 distinct ad. Mid tarsomere 1 twice as long as following tarsomeres together. Hind coxa with a very short black exterior bristle. Hind femur without ventral bristles; no preapical arcown of apical bristles. Hind tarsomere 1 as long as tarsomere 2.

WINGS. Uniformly brownish tinged, without spots. Tp straight, apical part of  $M_{3+4}$  almost twice as long as Tp. Anal vein not reaching wing margin.



**Fig. 30.** *Thinophilus spinatus* sp. nov., ∂, habitus.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short, black. Sternites without bristles, except sternite 4 with a tuft of short black bristles in apical half.

TERMINALIA (Figs 32–35). Phallus long, strap-shaped (Fig. 34 phallus folded). Cerci pale brownish with pale hairs, dorsally fused (Fig. 33).

**Female** (Fig. 31) LENGTH. Body 3.5 mm long, wing 3.1 mm long.

BODY. Similar to male except for following characters: clypeus <sup>1</sup>/<sub>3</sub> length of face, bulging; fore femur with 5 strong black ventral bristles up to 3 times as long as femur is wide.

## Distribution

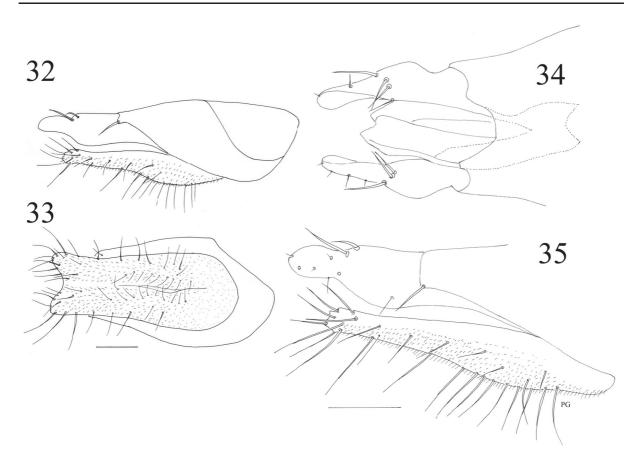
Southern Thailand (Andaman Sea) and Singapore.

## Remarks

The femora are spindle-shaped and the fore femur in male as well as in female bears long, brown stiff bristles as in *T. spinatoides* sp. nov. The main difference is that the fore femur in males of *T. spinatoides* sp. nov. is much more inflated than in *T. spinatus* sp. nov. For further differences, see under Remarks in *T. spinatoides* sp. nov.



**Fig. 31.** *Thinophilus spinatus* sp. nov.,  $\mathcal{Q}$ , habitus.



Figs 32–35. *Thinophilus spinatus* sp. nov., ♂, terminalia. 32. Genital capsule, lateral view. 33. Cerci, dorsal view. 34. Detail of tip of surstyli, ventral view. 35. Detail of surstylus and cercus, lateral view.

## *Thinophilus variabilis* sp. nov. <u>urn:lsid:zoobank.org:act:269414E1-124D-4113-9781-234E3E5340F9</u> Figs 36–40

#### Diagnosis

Medium-sized species with yellowish brown to brown fore coxa bearing black bristles. Fore tibia with a row of long ventral spine-like bristles over entire length of tibia. Wing brownish.

## Etymology

The specific epithet refers to the variable colour of the legs. In some specimens the legs are yellow, in others brown to black.

#### **Type material**

#### Holotype

THAILAND: ♂, Laem Pho, Hat Yai, Songkhla Province, 7°09′15.9″ N, 100°28′03.6″ E, sweep netting, 27 Jun. 2015, A. Samoh leg. (NHM-PSU).

#### **Paratypes**

 SAMOH A. et al., New species of Thinophilus from peninsular Thailand

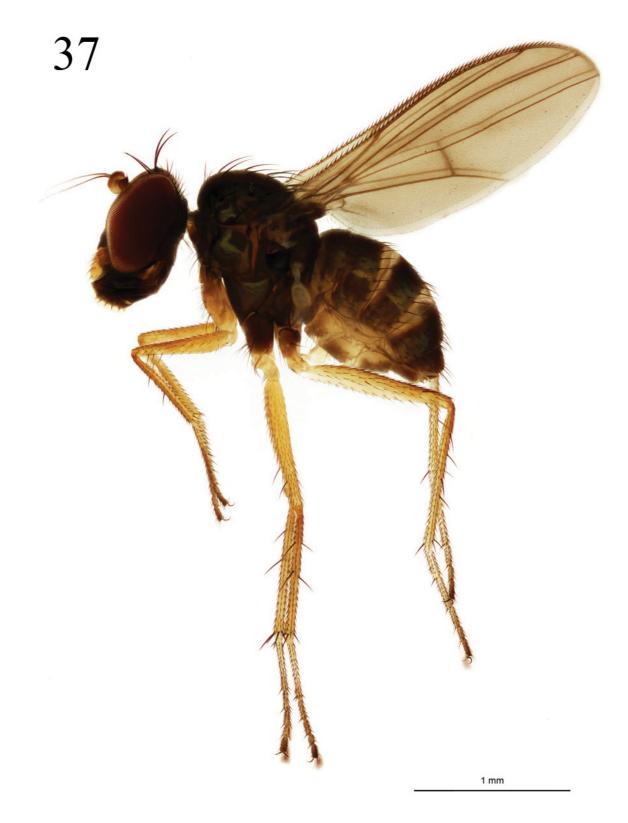
Thani, 9°23'34.0" N, 99°15'24.0" E, sweep netting, 20 Apr. 2015, A. Samoh leg.; 5  $\Diamond \Diamond$ , 18  $\bigcirc \bigcirc$ , Ban Dato, Yaring, Pattani, 6°55'17.1" N, 101°19'50.7" E, sweep netting, 12 Apr. 2015, A. Samoh leg. (NHM-PSU); 2  $\Diamond \Diamond$ , 1  $\bigcirc$ , same collection data as for holotype (RBINS).

## Description

Male (Fig. 36) LENGTH. Body 2.7 mm; wing 2 mm.

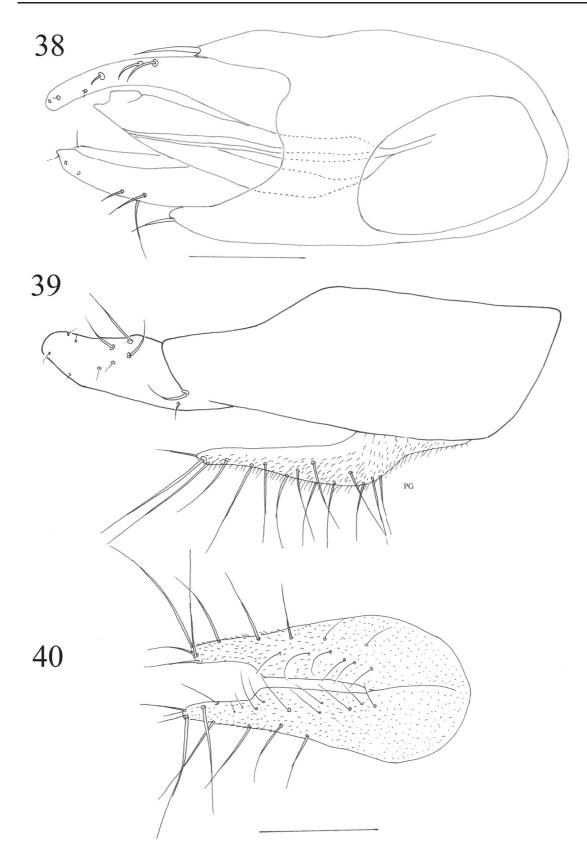


**Fig. 36.** *Thinophilus variabilis* sp. nov.,  $\mathcal{J}$ , habitus.



**Fig. 37.** *Thinophilus variabilis* sp. nov.,  $\mathcal{Q}$ , habitus.





**Figs 38–40.** *Thinophilus variabilis* sp. nov.,  $\eth$ , terminalia. **38**. Genital capsule, ventral view. **39**. Genital capsule, lateral view. **40**. Cerci, dorsal view.

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HEAD. Frons and face with shiny dark metallic green ground colour, but epistoma above with purplish reflections. Face above as wide as length of postpedicel, near middle narrower than postpedicel. Clypeus a third of length of epistoma. A pair of long divergent black ocellars. Two very short postocellars. A pair of convergent proclinate verticals, as long as ocellars. Vertex not excavated, dull. A pair of converging postverticals, only a little longer than postoculars, and not in row with upper postoculars. Postoculars uniseriate and black throughout; below neck with a transverse row of 4 black bristles longer than postoculars. Antenna yellowish; pedicel darker than postpedicel. Arista subdorsal, 3 times as long as antenna, brown, with short pubescence on basal half, longer diverging pubescence on apical half. Basal article short, brown; rest of arista paler. Palpus yellow, with short black bristly hairs. Proboscis brown.

THORAX. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 almost equally long dc, prescutellar one longest and outside row. Scutellum with 2 marginals and a short lateral bristle. One short black propleural above and 2 longer black propleurals below.

LEGS. Yellow to brown, including all tarsomeres. Fore coxa completely yellow, sometimes with sides brownish or completely brown; mid and hind coxae brownish, apices pale. Fore coxa anteriorly with long curved black bristles. Trochanter with a long black bristle. Fore femur a little wider than mid femur, especially on basal half; ventrally near base a few black bristles that are shorter than femur is wide; a posteroventral row of bristles over entire length, near base as long as femur is wide, on apical half longer (Fig. 36). Fore tibia shorter than femur, a ventral row of bristles over entire length, bristles over entire length, bristles as long as tibia is wide only on apical half. Mid coxa with a long black exterior bristle near middle as long as coxa is long; anteriorly with long black bristles. Mid femur with an av bristle at apical quarter; 4 pv bristles on apical quarter. Mid tibia as long as femur; with 2 ad, 2 shorter pd and apical crown of bristles. Hind coxa with a row of black bristles half as long as femur is wide; near middle with an ad and an preapical at apical fifth; 3 preapical pv as long as femur is wide and 3 shorter av.

WINGS. Brownish tinged, without spots. Tp straight, apical part of  $M_{3+4}$  2.5 times as long as Tp. Anal vein not reaching wing margin.

ABDOMEN. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites with black hairs.

TERMINALIA (Figs 38–40). Phallus long, strap-shaped. Cerci pale yellowish, surstyli brown, epandrium brown. Cerci pale yellowish, surstyli brown, epandrium brown. Cerci not fused, with very long subapical bristles.

**Female** (Fig. 37) LENGTH. Body 2.4 mm long, wing 2.3 mm long.

BODY. Stouter than male, otherwise similar except for following characters: fore femur with only a row of pd near tip; tibia with only short ventrals.

#### Distribution

Southern Thailand (Gulf of Thailand).

#### Remarks

*Thinophilus variabilis* sp. nov., a small species, differs from *T. minutus* sp. nov. in having distinct ventral bristles on all femora. Most characteristic in *T. variabilis* sp. nov. is the row of long ventral bristles on

the fore tibia, which is absent in *T. minutus* sp. nov. Coxae and femora can vary in colour from yellow to brown and even dark brown. Such a variation in colour is fairly unusual in *Thinophilus* and might be due to the preservation of the specimens in denaturised ethanol. The species seems to be widespread in peninsular Thailand.

# Key to male Thinophilus from the Thai-Malay Peninsula

1.	Wing with dark spot on middle of apical section of $M_{1+2}$ (level of wing boss), on cross vein and sometimes on vein $R_{4+5}$ , if the clouding on the veins is weak; male with a tuft of long bristles on sternite 3 and 4
2.	Fore femur with long ventral bristles, at least twice as long as femur is wide
3. -	Fore femur in both male and female with 4–5 stiff brown bristles that are more than twice as long as femur is wide (Figs 25, 30). Legs yellow
4.	Fore coxa completely yellow. Fore femur strongly spindle-shaped dilated in basal quarter (Fig. 25). Fore tibia longer than fore femur. Fore tarsomere 1 very long and slender, twice as long as fore tibia. Fore tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 widened, black <i>spinatoides</i> sp. nov. Fore coxa black. Fore femur weakly dilated at base. Fore tibia a little shorter than fore femur (Fig. 30). Fore tarsomere 1 about as long as fore tibia. Fore tarsomere 3 not paler than preceding tarsomeres. Fore tarsomeres 4 and 5 not widened, black
5. -	Fore coxa darkened on basal half or completely darkened ( <i>variabilis</i> sp. nov. usually has yellow fore coxa, but they might be brownish infuscate)
6. —	Tarsomere 2 of mid leg with a shield-like dorsal black protuberance (Figs 16–17), tarsomere 3 white      7      Tarsomere 2 of mid leg without dorsal protuberance
7. —	Mid femur at base with a cluster of distinct black ventral bristles (Fig. 16). Hind femur with ventral bristles in apical half shorter than femur is wide (Fig. 16) <i>parmatoides</i> sp. nov. Mid femur at base without a cluster of black ventral bristles (Fig. 17). Hind femur with ventral bristles in apical half longer than femur is wide (Fig. 17) <i>parmatus</i> Grootaert & Meuffels, 2001
8.	All femora darkened, if femora yellow, fore femur with long white curly bristles at base. Tip of fore tibia and all tarsomeres 5 darkened at tip. Hypopygium short, less than one-third length of abdomen (Fig. 1)
	Fore coxa entirely black. Large robust species with distinctly bristled legs
10.	Fore coxa in male protruding, hump-backed. Hypopygium less than half length of abdomen

Legs entirely yellowish white except for all coxae darkened. Fore coxa not hump-backed swollen.
 Hypopygium elongate, more than half length of abdomen (Fig. 6) ... *langkawensis* sp. nov.

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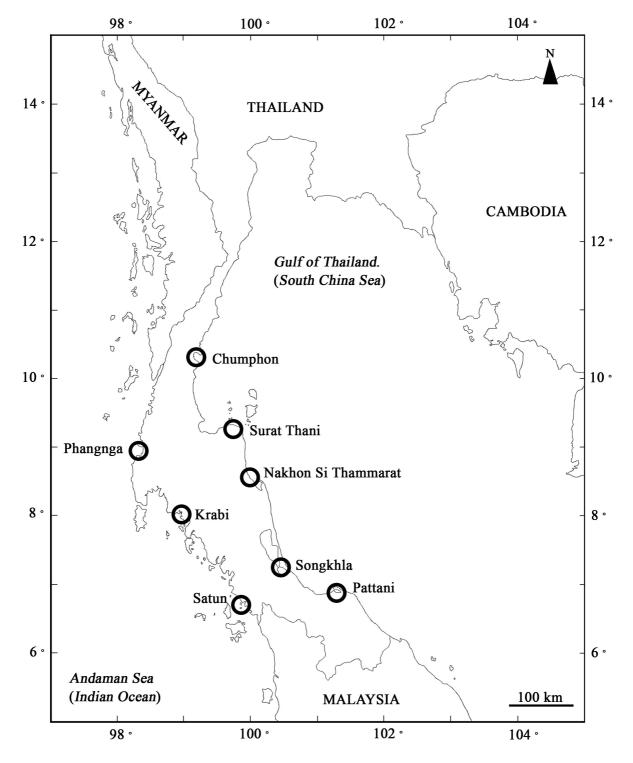
# Discussion

The present study is primarily based on a survey done by the first author to assess the species diversity in mangroves of peninsular Thailand. *Thinophilus* is a very diverse genus that is widely distributed in many littoral marine habitats, including mangroves, mudflats, sandy beaches and rocky shores. The present survey in peninsular Thailand confirms the statement of Evenhuis & Grootaert (2002), that *Thinophilus* is quite common in marine habitats of the Oriental and the Indo-Pacific regions.

Here, we did not compare the marine fauna with freshwater habitats such as streams and marshland. It should be noted that *T. setiventris* and *T. nitens*, described from a dry streambed near Wat Tapotaram in Ranong Province (Thailand) by Grootaert & Meuffels (2001), are primarily freshwater species and might be erroneously interpreted as marine as the title of that paper suggests. Observations in Singapore showed that *T. setiventris* is mainly present in drains and marshland. It rarely invades mangroves, together with *T. nitens*, after periods of heavy rains and flooding from nearby grasslands, where they forage on mosquito and chironomid larvae. Otherwise, these species were never found in mangrove (Grootaert, unpubl.). Most of the marine *Thinophilus* occur in front mangroves or along creeks draining back mangroves (Grootaert *et al.* 2016), where they forage along the water line for insect larvae in the mudflats. However, we also observed specimens foraging on rocky shores.

Although marine fauna is supposed to disperse easily along coasts, the marine *Thinophilus* seem to be rather endemic in the different parts of the South China Sea. The species of the mangroves along the coast of the Chinese mainland differ from those of Taiwan (Becker 1922), Northeast Borneo (Parent 1935) and those of the southern part of the South China Sea, as shown in the present study. Differences in faunal composition between the Gulf of Thailand and the Andaman Sea is more trivial, since the composition of the mangrove flora on either side of the Thai-Malay Peninsula has been proven to be different (Ge & Sun 2001; Huang *et al.* 2008; Minobe *et al.* 2009; Liao *et al.* 2009). We do not yet have information on the insect fauna in general. Three species of the nine true marine *Thinophilus* from peninsular Thailand, *T. parmatoides* sp. nov., *T. parvulus* sp. nov. and *T. variabilis* sp. nov., are actually known from the Gulf of Thailand, the southern part of the South China Sea, only; while *T. minutus* sp. nov. and *T. parmatus* occur on both sides. The remaining four newly described species are so far known only from the side of the Andaman Sea (*T. boonrotpongi* sp. nov., *T. langkawensis* sp. nov., *T. spinatus* sp. nov. and *T spinatoides* sp. nov.). Moreover, the type of mangroves is different. Along the

Gulf of Thailand, the mangroves are less extended, with smaller and less dense trees and under higher anthropogenic pressure. The mangroves along the coast of the Andaman Sea are much more extended, with higher trees and more pristine overall. The land barrier between the Andaman Sea and the Gulf of Thailand is important, implying that the flies cannot cross them easily (Fig. 41). First contact between the two seas is only in the extreme South of the Peninsula at the level of Singapore. Genetic studies



**Fig. 41.** Map of peninsular Thailand indicating the provinces respectively on the side of the Andaman Sea and the Gulf of Thailand (southern part of the South China Sea).

may demonstrate how large the genetic differences are in species common to both sides and how old the separation is. On the other hand there is a yearly crossing of fishing boats over land from one side to the other that might explain that some species are found on both sides of the peninsula. Pupae or larvae sticking on the hull of the boats could be transported this way.

Although nearly 40 species of *Thinophilus* are known hitherto from the Oriental region, it is not yet practical to classify them into species-groups. In the present study, a tight relationship is seen between *T. parmatus* and *T. parmatoides* sp. nov. They share a modified mid tarsus with a shield-like black protuberance, being a male secondary sexual character often found in other dolichopodid genera. *Thinophilus spinatus* sp. nov. and *T. spinatoides* sp. nov. share the presence of a pair of very long ocellar bristles and minute vertical bristles on the head (the forward shifted vertical bristle in a fronto-orbital position), combined with long, slender legs with club-shaped fore femora swollen near the base. All four species also have dorsally fused cerci over the entire length. It is likely that these four species can be united into a species-group that, however, will need to be confirmed by molecular support. Although Lim *et al.* (2009) used six genes, the relationship of fourteen species was not resolved at all, with very low bootstraps between the nodes. Similar poor resolution between various species from Singapore and China was found by Grootaert *et al.* (2015). All this points to an early origin of *Thinophilus* that cannot be resolved by non-conservative molecular markers. Delineating further species-groups is highly speculative, since the polarities of key morphological characters are unclear and sound molecular data are not yet available.

# Acknowledgements

This research was supported by the Higher Education Research Promotion and National Research University (NRU) Project of Thailand (No. SCI 540531 M), Office of the Higher Education Commission, and the Prince of Songkla University graduate school research funding. We also thank a scholarship for an overseas thesis research study from the Faculty of Science for financial support and the opportunity to conduct our research at the Royal Belgian Institute of Natural Sciences (RBINS), Brussels, Belgium. The authors thank Dr Singtoe Boonrotpong and the members of the Entomology Research Unit for their highly appreciated guidance and generous help in the field. We also thank Julien Caudron (Entomology, RBINS) for his kind and skillful help and fruitful suggestions in fly photography. The third author (P.G.) obtained a grant from the FPVII European-funded Integrated Infrastructure Initiative Synthesys to study the dolichopodid collections in Naturalis (Leiden), the Muséum national d'Histoire naturelle (Paris) and the Museum für Naturkunde (Berlin). The help in tracing type material received from Prof. Cees van Achterberg (Naturalis), Mr Ben Brugge (Amsterdam), Dr Pjotr Oosterbroek (Amsterdam), Dr Christophe Daugeron (Paris), Dr Joachim Ziegler (Berlin) and Dr Frank Menzel (DEI, Müncheberg) is much appreciated.

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Manuscript received: 15 April 2016 Manuscript accepted: 19 September 2016 Published on: 12 June 2017 Topic editor: Gavin Broad Desk editor: Kristiaan Hoedemakers European Journal of Taxonomy 329: 1-40 (2017)

Printed versions of all papers are also deposited in the libraries of the institutes that are members of the *EJT* consortium: Muséum national d'Histoire naturelle, Paris, France; Botanic Garden Meise, Belgium; Royal Museum for Central Africa, Tervuren, Belgium; Natural History Museum, London, United Kingdom; Royal Belgian Institute of Natural Sciences, Brussels, Belgium; Natural History Museum of Denmark, Copenhagen, Denmark; Naturalis Biodiversity Center, Leiden, the Netherlands; Museo Nacional de Ciencias Naturales-CSIC, Madrid, Spain; Real Jardín Botánico de Madrid CSIC, Spain.

# Manuscript I

# AN ANNOTATED CHECKLIST OF THE MARINE LONG-LEGGED FLIES (DIPTERA: DOLICHOPODIDAE) FROM THAI PENINSULA

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## ABSTRACT

Sixty species of Dolichopodidae were listed from the peninsular of Thailand, with 27 of these constituting new record for the country. General discussion and keys to species of some genera are provided and together with short remarks in each species.

## **INTRODUCTION**

Dolichopodidae or long-legged flies (Diptera: Dolichopodidae) is one of the most diverse dipteran family in the world. With more than 7000 species have been recognized and are also known as cosmopolitan flies that can be distributed in all zoogeographical regions (Robinson 1970a,b; Dyte 1975; Dyte and Smith 1980; Bickel and Dyte 1989; Negrobov 1991; Pollet et al. 2004, Young et al., 2006). Unluckily, there are poorly sampled and reported from Oldworld. This flies can be extremely occurred in moist environments such as waterfall, saltmashes, water seepages, canals, but nonetheless there is little knowledge from studies on these flies in their marine habitats. In general, both adult and larvae of long-legged flies are considered as predaceous dipteran flies and mostly predated on soft-bodied arthropods and annelids. Besides, due to its highly sensitive to environmental alterations that making them inherently useful as bioindicator for site quality assessment (Pollet 1992, 2001; Pollet and Grootaert 1991, 1996).

Up to this time, lacking basic knowledge in various aspects of the Dolichopodidae in Thailand is evident. Few reports have been published from this country. For example, three new species of genus *Nanothinophilus*, namely, *N. armatus*, *N. dolichurus*, and *N. pauperculus* (Grootaert and Meuffels, 1998) from the Andaman seacoast were discovered. A year later, one peculiar genus such *Terpsimyia* was rediscovered from Gulf of Thailand (south China sea). A briefly data, including species composition, description, and regional generic key have been provided by Grootaert and Meuffels (2001). Fifteen species in seven genera were found, of seven species were claimed as species new to science (Grootaert and Meuffels, 2001). However, the species number of previously surveyed is too low and have not been conducted all marine habitats in Southern Thailand which is considered as the richest part in this country. In addition, it is not covered the standard methods of flying insects sampling such as Malaise trap and yellow pan trap. Moreover, in terms of species recognition, only traditional way of species identification has been done and lots of specimens remain unclear. Due to these facts, many gaps of knowledge are open and need to be elucidating and understanding.

Interestingly, after a year of fly surveyed from several kinds of marine habitats (including mangroves, rocky shores, sandy beaches, mudflats) throughout peninsular Thailand. The preliminary results revealed that the numbers of species were drastically increased and plenty of specimens were pending to described as a new species and new genus. Furthermore, the marine long-legged fly of Thailand has not been studied in other comprehensive contexts such as molecular taxonomy, biogeography, distribution pattern, ecology, and etc. Regading to this annotated checklist, we gladly provided an update of the species composition, species distribution, habitat preferences, and including a primarily discussion on the status of marine long-legged fly in Thailand.

## MATERIALS AND METHODS

**Study sites, sampling techniques, and duration**. This study was mainly conducted in eight provinces of peninsular Thailand, namely, Chum Phon, Surat Thani, Nakhon Si Thammarat, Songkhla, Pattani, Satun, Krabi, and Phang Nga. Sweep netting, Yellow pan traps and Malaise traps were adopted to collect fresh marine long-legged fly specimens in several mangroves, tide pools and mudflats by author during November 2014 until May 2015.

**Collection preservation and deposition**. All fresh specimens were preserved in 70% ethyl alcohol and deposited in the collections of the Princess Maha Chakri Sirindhon Natural History Museum (MNHM) of the Prince of Songkla University (PSU), Hat Yai, Songkhla, Thailand. In addition, voucher specimens were also preserved in the collections of the Royal Belgian Institute of Natural Sciences (RBINS), Brussels, Belgium.

Male genitalic features analysis. In order to investigate male genital features, the last part of the abdomen was dissected and immediately macerated in 70% ethanol. Dark sclerotized male terminalia were macerated in 10% KOH which was gently heated on a hot plate for 15-20 minutes. While a lighter colour of sclerotized male genitalia were soaked in 85% lactic acid and heated in oven (Brooks, 2005).

**Photography.** A focus stacking technique (see Brecko et al., 2014) is used to take a photo of all the marine long-legged fly specimens. The high-resolution pictures are stacked using Zerene Stacker software. The scale on these photos are 1 mm.

**Abbreviations used in text and on figures.** acr: acrostical bristles; ad: anterodorsal; ae: aedeagus; av: anteroventral; c: cercus; dc: dorsocentral bristles; pd: posterodorsal; ds: dorsal surstylus; pv: posteroventral; hy: hypandrium; Tp: posterior cross vein (dm-Cu); vs: ventral surstylus.

#### **RESULTS AND DISCUSSION**

With 3,870 (1,556 males, 2,314 females) of marine long-legged flies (Insecta: Diptera: Dolichopodidae) specimens from peninsular Thailand, during November 2014 until May 2015 have been sorted out and identified into species level using external morphology characters. This leads to the conclusion that 60 morphospecies, 23 genera, seven subfamilies, and one unplaced subfamily were recognised from both sides of Southern Thai seacoasts (Table 1), and clearly shown that there were drastically increased (account for 79.49% or four time expanded) in term of species number when comparing to previous report by Grootaert and Meuffels (2001).

Furthermore, the results divulged that seventeen "new species" of marine longlegged flies were discovered from Thailand (Table 1). There were two species belong to Diaphorinae, namely, *Asyndetus* sp. nov and *Diaphorus* sp. nov., three species belong to subfamily Dolichopodidae such as *Hercostomus propermeieri* sp. nov., *Paraclius* sp. nov., *Phoomyia* sp. nov., and nine extended species for Hydrophorinae were identified and were considered the most diverse subfamily that containing the highest number of species new to science, there were *Cymatopus mayakunae* sp. nov., *Thinophilus boonrotpongi* sp. nov., *T. langkawensis* sp. nov., *T. minutus* sp. nov., *T. parmatoides* sp. nov., *T.s parvulus* sp. nov., *T. spinatoides* sp. nov., *T. spinatus* sp. nov., *T. variabilis* sp. nov., then, two species classified belong to subfamily Rhaphiinae, namely, *Ngirhaphium chutamasae* sp. nov. and *Ngirhaphium meieri* sp. nov., and a single new species and new genus of unplaced subfamily, which we have named *Ornamenta siamese* sp. nov.

In the context of "new record" for the country, 27 new records could be considered from this region (Table 1). In the manner that there were composed with a species of subfamily Diaphorinae; *Chrysotus dot*. While 22 species classified belong to the subfamily Dolichopodinae; were included *Argyrochlamys impudicus*, *Hercostomus brevicornis*, *H. brevidigitalis*, *H.s lanceolatus*, *H. obtusus*, *H. plumatus*, *Lichtwardtia ziczac*, *Paraclius adligatus*, *P. asiobates*, *P. digitatus*, *P. obtus*, *P. serratus*, *P. serratus*, *Tachytrecus tessellatus*, *Phoomyia singaporensis*, *Thinophilus apicatus*, *Thinophilus chaetulosus*, *T. melanomerus*, *T. simplex*, *T. superbus*, and *T. yeoi*; one minute species belongs to subfamily Parathalassiinae was *Microphorella malaysiana*; three species of the subfamily Rhaphiinae, namely, *Ngirhaphium caeruleum*, *N. murphyi*, and *N. sivasothii*; whereas a single species such *Sympycnus* sp. of the subfamily Sympycninae was also reported as a new record for Thailand (Table 1). Here below is annotated checklist and remarks of marine long-legged flies from Thai peninsula:

## SYSTEMATICS ACCOUNT

#### **SUBFAMILY DIAPHORINAE SCHINER, 1864**

**Remarks.** The Diaphorinae are cosmopolitan distribution long-legged flies (Diptera: Dolichopodidae) and be considered as a complex rich subfamilies (Bickel, 2005).

#### Asyndetus Loew, 1869

Type species: Asyndetus interruptus Loew, 1861

**Remarks.** The genus *Asyndetus* Loew, 1869 belongs to subfamily Diaphorinae, is presumed commensal flies of crab burrows, and is commonly found along beaches of seacoast in southern hemisphere (Grootaert and Meuffels, 2002). Both sexes have very peculiar wing, a broad with the costal ending at the tip of third longitudinal vein ( $R_{4+5}$ ) and not reaching to the tip of the fourth vein (M) like the most dolichopodidae. According to the list of Grootaert (1993), he listed 11 species from Papua New Guinea. In Thailand, four species were recorded (Grootaert and Meuffels, 2002), including, *Asyndetus ciliatus*, *A. aciliatus*, *A. thaicus*, and *A. latifrons*. While, three species were identified from current study.

#### Key to the male Asyndetus Loew from Thailand

2) Hind tibiae yellowish brown, and containing a series of long posteroventral hairs; lower postoccular bristles white, at most pale yellowish....... *A. ciliatus* Grootaert and Meuffels

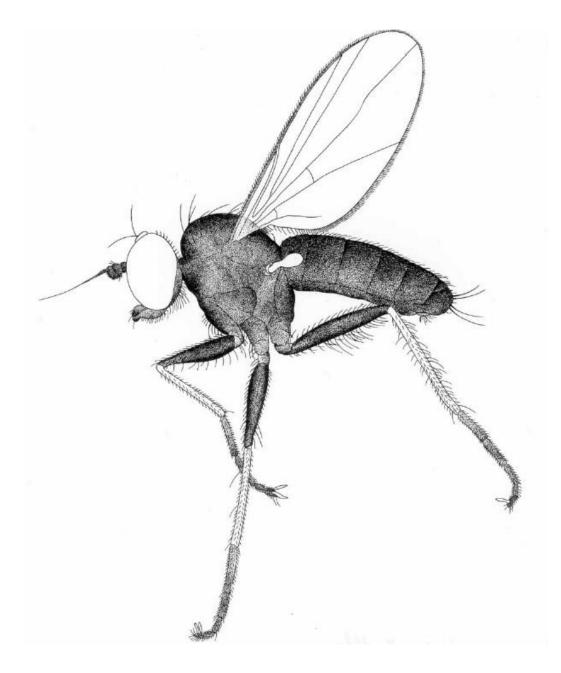


Figure 1. Male habitus, *Asyndetus ciliatus* Grootaert and Meuffels, 2002 (Grootaert and Meuffels, 2002)

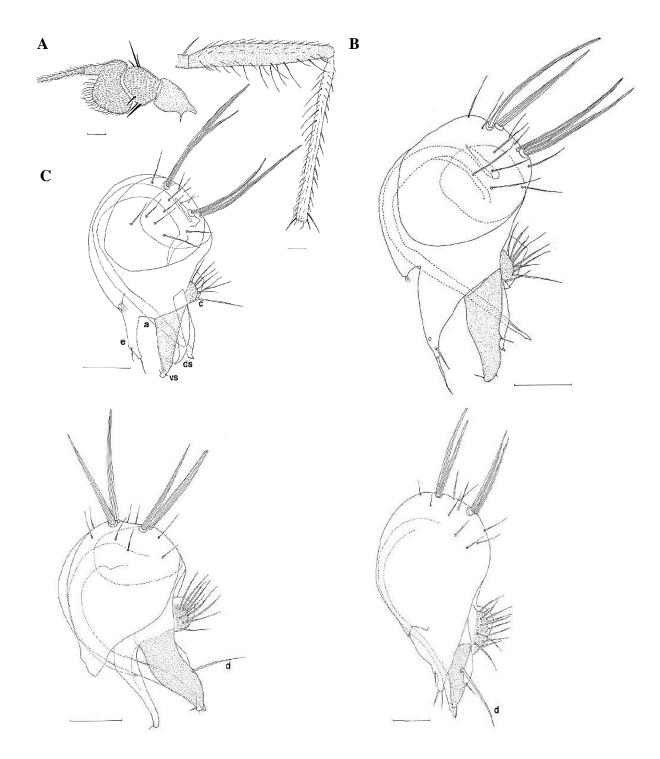


Figure 2. *Asyndetus ciliatus*, male. (A). Antenna; (B). Hind femur and tibia from behind (arrow points to the long posteroventral hairs); (C). Hypopygium (a: aedaeagus; c: cercus; d: dorsal bristle on ventral surstylus; ds: dorsal surstylus; e: epandrial lobe; vs: ventral surstylus). Bar scale = 0.1 mm (modified from Grootaert and Meuffels, 2002).

(Figure 3)

**Material Examined.** 13; Thailand, Talumpuk Cape, Pak Phanang, Nakhon Si Thammarat, (8°31'06.1"N 100°06'51.6"E), sweep netting, 30 April 2015, coll. A. Samoh.

**Remarks**. There is a small diaphorine species (body length 2.5-2.6 mm; wing length 2.2-2.3 mm). In fact, in Thailand, *A. aciliatus* was firstly collected from Na Haeo, Loei province in 2001 (Grootaert and Meuffels, 2002) at the altitude of 500 m from sea level (non-marine habitat). On the contrary, this study, we mainly collected from sandy beaches with *Ipomoea* entirely covered and high sun-exposed. Femora black with yellow knees; fore and mid tibia yellow; and hind tibia completely brown; dorsal bristle on ventral surstylus very minute are taxonomic characters to indicate the species.

**Distribution.** Nakhon Si Thammarat (Gulf of Thailand)

# Asyndetus thaicus Grootaert and Meuffels, 2002

(Figure 4)

**Material Examined.**  $7 \stackrel{\circ}{\supset} 2 \stackrel{\circ}{\ominus}$ ; Thailand, Tarutao Island, Langu, Satun, (6°44'19.2"N 99°38'45.4"E), sweep netting, 9 January 2015, coll. A. Samoh;  $1\stackrel{\circ}{\supset}$ ; Ban Thong Tom Yai, Muang, Chumphon, (N 10°12'39.2', E 99°12'21.4'), sweep netting, coll. A. Samoh.

**Remarks**. There is a small species but a bit larger than *A. aciliatus* (body length 2.7-2.8 mm; and wing length 2.4 mm). They used to report from creek near the sea form Ko Samed, Rayong province. Interestingly, this species mostly observed at the same biotopes as *A. aciliatus* that was sandy beaches with little *Ipomoea* covered. However, other biological information is lacking off and badly needed. In the context of morphological features, it is remarkably recognised by completely black legs, fore femur on basal half with a row of ventral bristles, half as long as femur as deep. Hind femur with 2-3 strong preapical anteroventral bristles; and epandrial lobe not so slender when comparing to *A. latifrons*.

Distribution. Satun (Andaman Sea), Chumphon (Gulf of Thailand)



Figure 3. Male habitus, Asyndetus aciliatus Grootaert and Meuffels, 2002



Figure 4. Male habitus, Asyndetus thaicus Grootaert and Meuffels, 2002

# Chrysotus Meigen, 1824

**Remarks.** – The *Chrysotus* comprises more than 440 species have been identified throughout the world (Negrobov et al., 2015; Wei et al., 2015). Moreover, this genus has been revised many times by Negrobov and colleagues (Negrobov 1980; Negrobov and Maslova, 1995; Negrobov et al., 2000, 2003). In Asia, China is the leading country for species recorded of this long-legged fly genus, with more than a hundred species are known (Wei 2010; Wei et al., 2015; Liu et al., 2013, 2015). In general, they are few morphological differences between the species of the genus, but most species can be distinguished by the distinctive male hypopygium (Figure 6, 7) with reference to the apex of the phallus (Figure 8) (Negrobov et al., 2016).

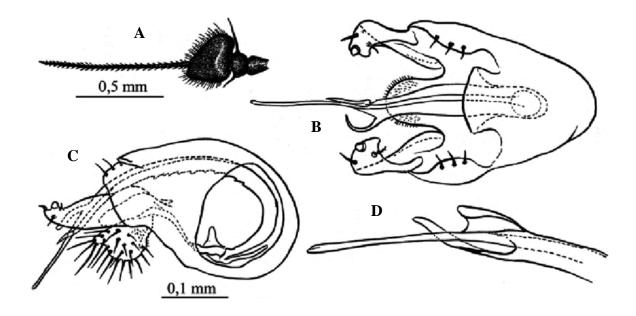


Figure 5. *Chrysotus tagoi* Negrobov, Maslova and Fursov, 2015, A – antenna; B – hypopygium, ventral view; C – hypopygium, lateral view; D – phallus, ventral view (Negrobov et al., 2016)

#### Chrysotus sp. A (dot)

**Material Examinaed.**  $1 \circlearrowright 1 \circlearrowright 1$ ; Thailand, Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), sweep netting, 17 February 2015, coll. A. Samoh;  $1 \circlearrowright$ ; Hua Khao, Singha Nakhon, Songkhla, (7°12'03.6"N 100°34'36.8"E), sweep netting, 27 May 2015;  $1 \circlearrowright$ ;

Laem Pho, Khu Tao, Hat Yai, Songkhla, (7°09'15.9"N 100°28'03.6"E), sweep netting, 27 May 2015, coll. A. Samoh.

**Remarks.** – This species was firstly collected from Singapore mangrove and pending for describing as a new species to science (Grootaert, P., personal communication). This species has not been recoded from Thailand and here is the first record for this country.

**Distribution.** Chumphon, Songkhla, Satun, and Phang Nga.

# Diaphorus Meigen, 1824

**Remarks.** – The genus *Diaphorus* was firsly recognised by Fallen in 1823 belonging to the genus *Dolichopus*. But, a year later, it was designated out from *Dolichopus* by Meigen (Hollis, 1964). The type species of this genus, *Diaphorus flavocinctus* Meigen [at present, a synonym of *D. oculatus* (Fallen)], was designated by Westwood (1840) (Hollis, 1964). Meigen (1824) also described the species *D. tuberculatus* in the genus *Dolichopus* and *Diaphorus nigricans*, *D. wintemi*, and *D. lavocinctus* in the genus *Diaphorus*, *D. tuberculatus* and *D. flavocinctus* being later lowered to synonyms of *D. oculatus*.

The genus *Diaphorus* Meigen, 1824 (Diptera: Dolichopodidae) can be distinguished from other relative genera of the subfamily Diaphorinae by the following characters: body small to medium, metallic green at grounded of body; eyes usually contiguous or narrowly separated on frons, rarely with parallel margins; face rather wide with parallel margins; arista long, with dorsal insertion; 4–6 strong dorsocentral bristles, acrostichal bristles biserial; legs with small claws or absent, usually with well-developed pulvilli; male sternum 8 with 2–8 strong bristles. This genus has 267 known species distributed worldwide (Yang et al. 2011).

# Diaphorus sp. A

#### (Figure 6)

**Material Examinaed.** 53; Thailand, Ban Bang Yai, Takuapa, Phangnga, (8°54'27.5"N 98°23'59.6"E), sweep netting, 11 February 2015, coll. A. Samoh; 23; Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), sweep netting, 17 February 2015, coll. A. Samoh; 2312; Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh; 13, Ban Nua Nam, Phumriang, Chaiya, Surat Thani, (9°23'34.0"N 99°15'24.0"E), sweep netting, 20 April 2015, coll. A. Samoh.

**Remarks.** – This species has not previously been recorded from this country. This recorded species was recently collected from mangroves used Malaise traps and probably represent new species.

Distribution. Phang Nga, Satun, Surat Thani, and Chumphon



Figure 6. Male habitus, Diaphorus sp. A

## SUBFAMILY DOLICHOPODINAE LATREILLE, 1809

#### Type genus: Dolichopus Latreille, 1796

**Remarks**. The Dolichopodinae are recognised as one of the most diverse subfamilies of Dolichopodidae. There are cosmopolitan distribution and over 1,700 described species from all zoogeographical regions (Brooks, 2005). Furthermore, this subfamily can be easily distinguished from their allies by these combination characters: scape dorsally setose (Figure 7A); mid and hind femur with 1 or more anterior preapical setose; male segmented 7<sup>th</sup> bare and forming a peduncle; hypopygium folded under the abdomen (Figure 7 C, D, E).

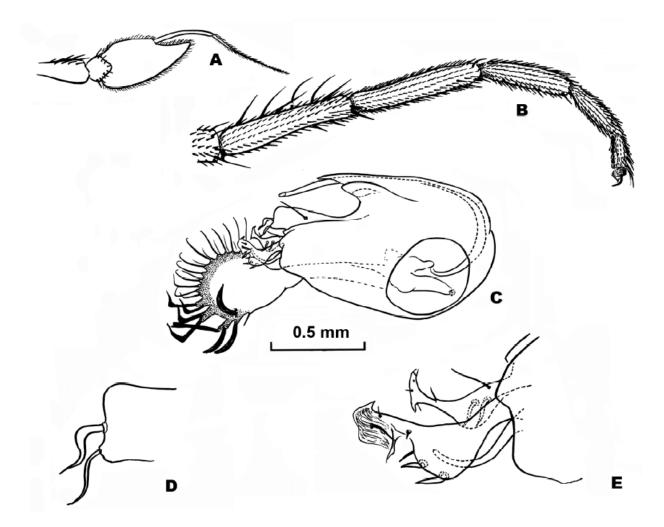


Figure 7. *Dolichopus grootaerti*, laterally: (A) antenna; (B) hind tarsus; (C) hypopygium; (D) surstylus; (E) gonopodes (Negrobov et al., 2014).

# Argyrochlamys Lamb, 1922

# Argyrochlamys Lamb, 1992: 391.

# Type species: Argyrochlamys impudicus Lamb [Oriental], by monotypy

**Remarks**. The generic name of this fly related to place that mostly found. There are generally observed near the entrance zone of the ghost crab burrows and seemed to be restricted to the ocean beaches with high sun-exposed. In addition, they can be recognized by their non-metallic body, pale yellowish brown to dark grey colour, and also contained with 5 pairs of dorsocentrals. Males can be easily identified by the distinctive "*comma shaped-like*" or "*bifurcate projection*" (Brooks, 2005) near the joint of the hind tibia and basitarsus. This genus is mainly record from Afrotropic, for example, Ghana, Mauritius, Somalia, Sudan (Dyte and Smith, 1980) and is also reported from Oriental realm, Chagos Island Srilanka (Dyte, 1975)

# Argyrochlamys impudicus Lamb, 1922

(New record, Figure 8)

**Material Examined.** 223; Tarutao Island, Langu, Satun, (6°44'19.2"N 99°38'45.4"E), sweep netting and hand collecting, 25 December 2014, coll. A. Samoh; 35322; Bulon Island, Langu, Satun, (6°49'44.5"N 99°32'07.7"E), sweep netting, 13 May 2015, coll. A. Samoh.

**Remarks.** Setation of body and legs mostly black. Wing venation aberrant, with  $R_{2+3}$  short, reaching wing margin just beyond middle; apical section of  $M_{1+2}$  in middle with nearly right-angular curvation; ratio of m-cu to distal part of CuA<sub>1</sub>. Female postpedicel as long as high, with distinctly dorsal stylus; stylus longer than postpedicel. Hypopygium with long and narrow curved distal epandrial lobe; cercus long, gradually narrowing apicad.

Distribution. Satun (Andaman Sea), Nakhon Si Thammarat



Figure 8. Male habitus, Argyrochlamys impudicus Lamb, 1922

#### Genus Hercostomus Loew, 1857

(New record)

# **Systematic Account**

# Key to species of mangrove Hercostomus sensu lato from Thailand

1) Wing, with brown colour (sometimes is pale) stigma at the end of vein  $R_1$ ; noticed that vein  $R_{4+5}$  relatively thickened from the basal quarter onwards (Figure 13B, 14)......2 2) Stigma elongate (Figure 14), exposing beyond start of the thickening of vein  $R_{4+5}$ - Stigma short, reaching the as same as level of thickening of vein  $R_{4+5}$  ..... - First flagellomere rather short (at most, one point up to five times as long as wide)..... 5 4) All coxae blackish colour; generally femora blackish except some part at tip of fore and mid femora; cercus comprising of three strong bristles at outer magin......Hercostomus brevidigitalis - Fore coxae yellowish colour; mid and hind coxae brown; all femora yellow, and cercus without strong bristles as above...... Hercostomus meieri (Singapore) - Fore tarsomere relatively shortened; cercus distinctly large, with short apical tail 6) Fore and hind coxae yellow; cercus nearly geniculate, aedeagus with two inner denticles ...... Hercostomus brevicornis - Only fore coxae yellow; cercus triangular in shape, aedeagus with one inner 

#### Hercostomus Loew, 1857: 9.

Type genus: Dolichopus Latreille, 1796

*Hercostomus* Loew, 1857: 9. Type species: *Sybistroma longiventris* Loew [Palaearctic], by original designation.

**Remarks**. Genus *Hercostomus* sensu lato, as defined as, a polyphyletic assemblage of species, related to *Dolichopus*, *Parahercostomus*, and *Poeccilobotrus* (Brooks, 2005), with typical wing vein  $R_{4+5}$  and M gently bent anteriorly beyond crossvein dm-cu. In addition, this genus is remained all major external morphological characters of the subfamily Dolichopodinae with pteropleuron without hairs i.e. no hairs in front of the posterior thoracic spiracle. Fifth pair of dc not or rarely slightly convergent. Male clypleus not bulging, lower margin usually straight and not reaching lower eyes margin, scape and pedicel well developed; arista simple and short with developed pubescent.

Apparently, this group of fly is still largely debatable in term of taxonomic arrangement. Especially one recognized species from Southeast Asia, namely, *Steleopyga* (Grootaert and Meuffels, 2001b) as described a separated genus, by indicated some characters (the possession of a cluster of spines on sternite 8 and one preapical anteroventral row of 4 setae on the hind femur, and the complexes of male genitalia – forming elements of entangled asymmetrical lobes, and Brooks (2005) suggested that to be a homologous with the condition of *Hercostomus longiventris* lineages that is a part of the *"Hercostomus complex"*. In addition, Yang et. al. (2006) reported that these flies contained with over 270 species from the Oriental region and mostly collected from non-marine habitats. However, some species (seven new species) are also recorded from marine habitat such as mangroves in Singapore (Zhang et. al. 2008). Unfortunately, this genus never ever recorded from Thailand. This study is the first reported (with seven known species) from Thai peninsula.

# Hercostomus brevicornis, Zhang et. al., 2008

(New record) (Figure 10)

**Material Examined.**  $4\overset{\circ}{\partial}1\overset{\circ}{\Box}$ ; Thailand, Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh; 5 $\overset{\circ}{\partial}$ ; Laemson Kamphaeng, Satun, (6°56'27.9"N 99°42'12.4"E), sweep netting, 4 May 2015, coll. A. Samoh;  $7\overset{\circ}{\partial}4\overset{\circ}{\Box}$ ; Tammalang, Muang, Satun, (6°32'21.7"N 100°04'09.3"E), sweep netting, 2 May 2015, coll. A. Samoh;  $6\overset{\circ}{\partial}1\overset{\circ}{\Box}$ ; Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh;  $4\overset{\circ}{\partial}2\overset{\circ}{\Box}$ ; Ban Bang Yai, Bang Nai Si, Takuapa, Phangnga, (8°54'27.5"N 98°23'59.6"E), 9 February 2015, coll. A. Samoh;  $6\overset{\circ}{\partial}4\overset{\circ}{\Box}$ ; Khlong Phon, Khlong Thom, Krab,i (7°48'11.2"N 99°10'11.9"E), sweep netting, 13 May 2015, coll. A. Samoh.

**Remarks.** Body length 2.9–3.1 mm, wing length 2.7–2.8 mm. All coxae yellow, but mid coxa at most pale brownish with a narrow black anterior stripe. Cercus strongly curved, nearly geniculate. Aedeagus with 2 small, inner denticles (Figure 9). **Distribution.** Satur, Phangnga, Krabi, and Surat Thani

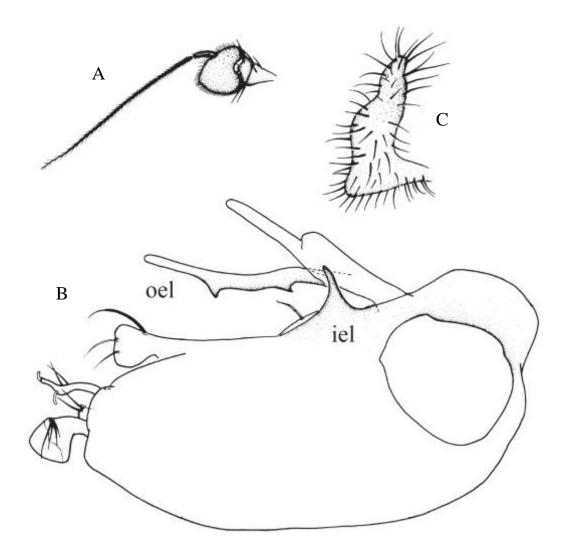


Figure 9, *Hercostomus brevicornis*, Male: A, antenna; B, male genitalia; C, cercus; ile, inner epandreal lobe; oel, outer epandreal lobe (modified from Zhang et al., 2008)

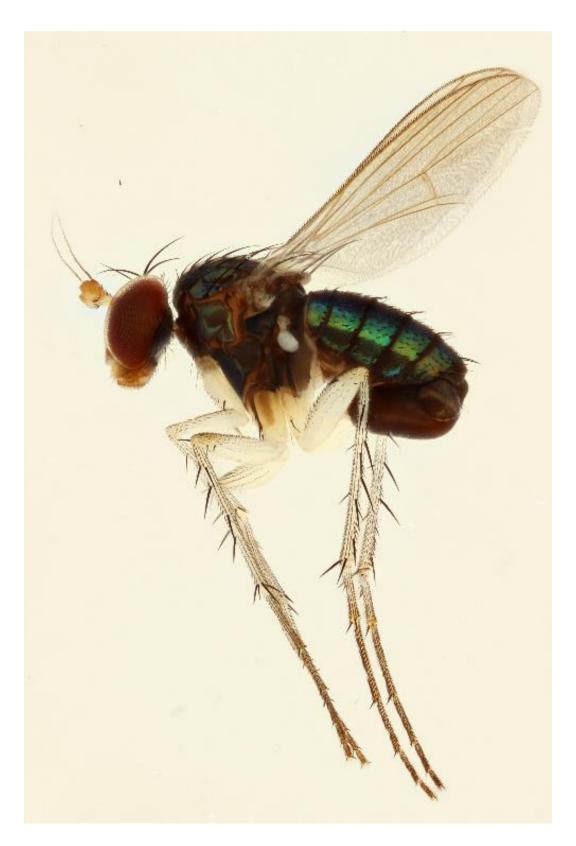


Figure 10. Male habitus, Hercostomus brevicornis Zhang et al., 2008

# Hercostomus brevidigitalis, Zhang et al., 2008

(New record, Figure 12)

**Material Examined.**  $1 \cancel{3} 2 \cancel{2}$ ; Laemson, Kamphaeng, Satun, (6°56'27.9"N 99°42'12.4"E), sweep netting, 4 May 2015, coll. A. Samoh;  $3 \cancel{3} 1 \cancel{2}$ ; Thailand, Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh;  $2 \cancel{3} 1 \cancel{2}$ ; Ban Ramard, Khlong Thom, Krabi, (7°42'17.4"N 99°03'48.4"E), sweep netting, 26 April 2015;  $3 \cancel{3} 1 \cancel{2}$ ; Ban Bang Yai, Takuapa, Phangnga, (8°54'27.5"N 98°23'59.6"E), sweep netting, 11 February 2015, coll. A. Samoh.

**Remarks.** This is a small to medium species (body length 3.2–3.6 mm, wing length 3.2–3.5 mm). First flagellomere elongate, 2.3 times as long as wide. All coxae black; femora black except tip of fore and mid femora. Hind femur with 3 brown ventral bristles near base (a little shorter than width of femur). Fore tarsomere1 without ventral slit. Squama yellow with black hairs. Male genitalia very long, reaching thorax (Zhang et al., 2008).

Distribution. Satun, krabi, and Phangnga

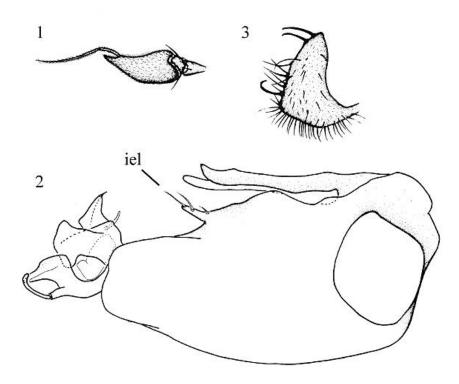


Figure 11. *Hercostomus brevidigitalis*, male: 1, antenna; 2, male genitalia; 3, cercus. iel, inner epandreal lobe (modified from Zhang et al., 2008)



Figure 12. Male habitus, Hercostomus beridigitalis Zhang et al., 2008

# Hercostomus lanceolatus, Zhang et. al., 2008 (New record, Figure 14)

**Material Examined**.  $9 \checkmark 14 \heartsuit$ ; Thailand, Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh;  $1 \checkmark 3 \heartsuit$ ; Tammalang, Muang, Satun, (6°32'21.7"N 100°04'09.3"E), sweep netting, 2 May 2015, coll. A. Samoh;  $2 \checkmark 1 \heartsuit$ ; Thailand, Pakbara, Langu, Satun, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh;  $2 \checkmark 7 \heartsuit$ ; Prince of Songkla University (PSU), Muang, Pattani, (6°53'04.9"N 101°14'10.1"E), sweep netting and Malaise trap, 11 April 2015, coll. A. Samoh;  $3 \And 4 \heartsuit$ ; Ban Dato, Yaring, Pattani, (6°55'17.1"N 101°19'50.7"E), sweep netting, 14 April 2015, coll. A. Samoh; Ban Pak Phanang Tawantok, Pak Phanang, Nakhon Si Thammarat, (8°22'30.2"N 100°10'00.4"E), sweep netting, 1 May 2015, coll. A. Samoh;  $4 \Huge 3 \image$ ; Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), sweep neeting, 17 February 2015, coll. A. Samoh;  $4 \Huge 3 \trianglerighteq$ ; Ban Hua Khao, Singha Nakhon, Songkhla, (7°12'03.6"N 100°34'36.8"E), sweep netting, 27 May 2015, coll. A. Samoh.

**Remarks.** Body length 3.7 mm, wing length 3.3 mm. Wing with wide, yellowish brown to grey stigma behind tip of  $R_1$ , stigma reaching the level of thickening of  $R_{4+5}$ .  $R_{4+5}$  thickened from basal quarter (Zhang et al., 2008), but narrow again before reaching wing border.

**Distribution.** Satun, Chumphon, Surat Thani, Nakhon Si Thammarat, Songkhla, Pattani, and Singapore

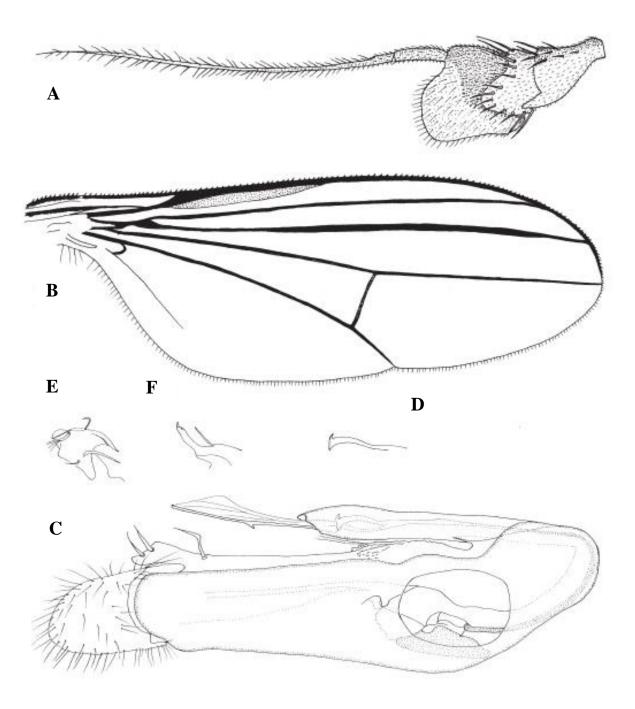


Figure 13. *Hercostomus lanceolatus*, male habitus, wing(A), antenna (B), male genitalia (C), claw-like aedeagus (D), ventral lobe of surstyli (E), dorsal lobe of surstyli (F) (modified from Zhang et. al., 2008).



Figure 14. Male habitus, Hercostomus lanceolatus Zhang et al., 2008

# *Hercostomus plumatus* Zhang et al., 2008 (New record, Figure 16)

**Remarks.** Body length 2.4–2.7 mm, wing length 2.2–2.5 mm. First flagellomere 1.2 times as long as wide (Zhang et al., 2008). Fore coxa yellow, mid and hind coxae brownish. Dorsal lobe of surstyli with plumose hair.

**Distribution.** Nakhon Si Thammarat, Surat Thani, Songkhla (Gulf of Thailand), Krabi Province (Andaman Sea)

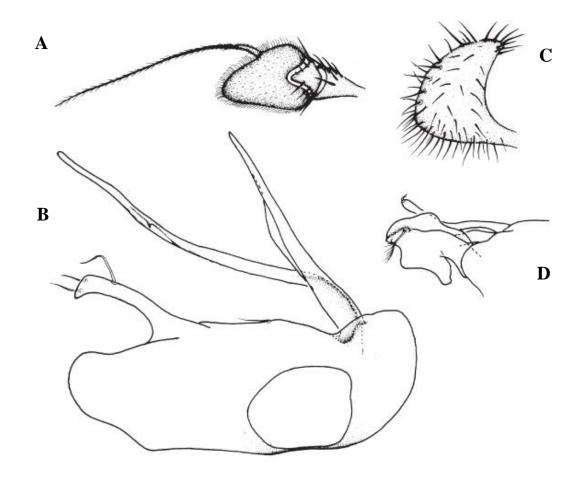


Figure 15. *Hercostommus plumatus*, male habitus; antenna (A); male genitalia (B); cercus (C); surstyli (D) (modified from Zhang et al., 2008)



Figure 16. Male habitus, Hercostomus plumatus Zhang et al., 2008

# Hercostomus obtusus sp. nov.

(New record, pending for description by Grootaert)

**Material Examined.** 332; Pakbara, Langu, Satun Province, (6°50'30.4"N 99°46'32.9"E), sweep netting, 29 April 2015, coll. A. Samoh

**Remarks.** Body length 2.3–2.5 mm, wing length 2.0–2.2 mm. First flagellomere 2.5 times as long as wide. Fore coxa yellow with a little black line at tip, mid and hind coxae brown. In facts, this species was first collected from Singapore mangroves and Grootaert suggested to be a new species.

Distribution. Pakbara and Bakan Toh Thid, Langu, Satun (Andaman Sea)

# Hercostomus propermeieri Zhang et. al., 2008

(New species, pending for description, Figure 17)

**Material Examined.**  $232^{\circ}$ ; Laemson Kamphaeng, Satun, (6°56'27.9"N 99°42'12.4"E), sweep netting, 4 May 2015, coll. A. Samoh.

**Remarks.** Body length 2.4–2.5 mm, wing length 2.1–2.2 mm. First flagellomere 3.0 times as long as wide (Zhang et al., 2008). Fore coxa yellow, mid and hind coxae brown.

Distribution. Ban Laem Son, Kam Phaeng, Langu, Satun (Andaman Sea)



Figure 17. Male habitus, *Hercostomus propermeieri* sp. nov.

#### Genus Lichtwardtia Enderlien, 1921

#### Lichtwardtia ziczac Wiedmann, 1824

(New record)

**Materials Examined.** 43; Ban Hua Khao, Singhanakhon, Songkhla, (7°12'03.6"N 100°34'36.8"E), sweep netting, 19 November 2014, coll. A. Samoh

**Remarks.** Small metallic green long-legged flies with dark yellow palpus and proboscis, fore coxa yellow, mid and hind coxae blackish colour. This species is mostly found at riverbank during high tide in very muddy mangroves near front sea.

**Distribution.** Songkhla province (Gulf of Thailand)

#### Paraclius Loew, 1864

(New genus record for Thailand)

Type species: Pelastoneurus arcuatus Loew, designation by Coquillett, 1910.

Erroneously treated as an emendation of Paracleuis Bigot in Foote et. al. 1965

**Remark and Recognition**. *Paraclius* is polyphyletic assemblages species which can be identified by these following combination characters: arista bare to pubescent (Figure 18A); clypeus flat, lower margin generally straight and ending above lower eye margin; the wing vein M beyond cross vein dm-cu with strong anterior bent near middle, sharply convergent with  $R_{4+5}$  and arcuate (see Figure 18B); hind coxa with strong lateral seta near apex; mid and hind femur usually with 1 anterior to anterodorsal preapical seta; hind basitarsus without dorsal setae.

The *Paraclius* has not been reported yet from Thailand. This study is the first recorded, with seven species are discovered as following. There are ranged in sizes (from 3-6.6 mm), by the way, this dolichopodine fly usually gathered along the river banks, stagnant brackish mangrove, and sun-exposed area of the muddy mangroves. Two sides of peninsular could be found.

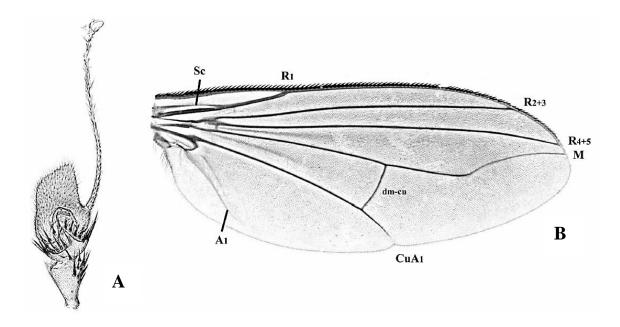


Figure 18. *Paraclius parenti*, **sp. nov.**, paratype. **A.** Male antenna. **B.** Male wing. (Modified from Capellari and Amorim, 2009)

#### Paraclius adligatus Becker, 1922

(New record)

**Materials Examined.**  $1 & 7 \\ \bigcirc$ ; Thailand, Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), sweep netting, 9 January 2015, coll. A. Samoh.

**Remarks.** - The medium marine dolichopodine flies (body length 4.0–4.1 mm, wing length 3.9–4.0 mm.) is easily recognized by some remarkable characters such as antenna black, first flagellomere twice as long as wide; arista with basal segment 0.25 times as long as apical segment (Zhang et al., 2007). All coxae and femora black. Apex of hind tibia and entire hind tarsus black. Mid and hind femora with black ventral bristles (shorter than femur is wide). Interestingly, this species was strictly distributed in muddy and very shady mangroves from Ban Khao Than, Surat Thani, Gulf of Thailand (South China Sea).

**Distribution.** Ban Khao Than, Tha Chang, Surat Thani, Gulf of Thailand (South China Sea), Thailand, and including Singapore.

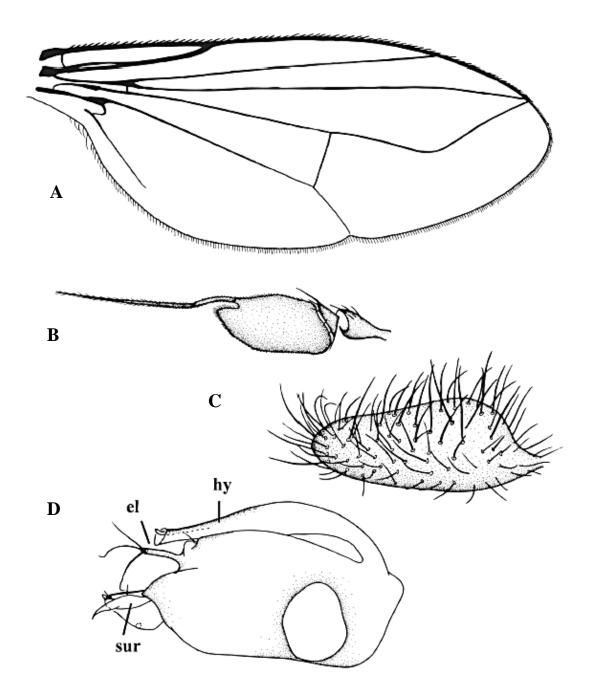


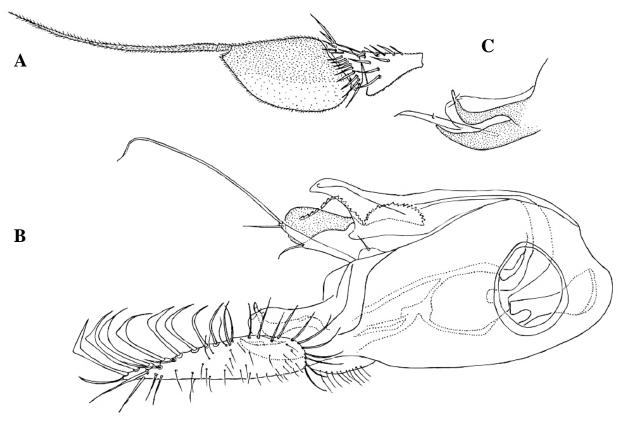
Figure 19. Paraclius adligatus Becker, male habitus: Wing (A), antenna (B), cercus (C), male hypopygium (D). Note that el- epandreal lobe; hy-hypandrium; sur-surstylus (modified from Zhang et al., 2007)

# Paraclius asiobates Zhang et al., 2007

(New record, Figure 21)

**Materials Examined**. 13; Thailand, Na Thab, Chana, Songkhla province, sweep netting, 27 September 2014, coll. A. Samoh.

**Remarks.** One of the largest marine *Paraclius* (body length 6.6 mm, wing length 4.5 mm.) in this region (southeast Asia). With dark yellow antenna, first flagellomere brown on upper half, yellowish on lower half, hind femur with row of black ventral bristles. Aedeagus with denticles. Cercus elongate triangular in lateral view, yellow, with brown



marginal clavate bristles (Zhang et al., 2007).

Figure 20. *Paraclius asiobates* Zhang et al., male habitus; antenna (A), male genitalia (B), surstyli (C) (modified from Zhang et al., 2007)

Distribution. Songkhla (Thailand), Singapore.



Figure 21. Male habitus, *Paraclius asiobates* Zhang et al., 2007.

# Paraclius digitatus Zhang et al., 2007

(New record, Figure 23)

**Materials Examined**.  $2 \Diamond 1 \heartsuit$ ; Bakan Tohthid, Langu, Satun, (6°47'29.8"N 99°48'53.5"E), sweep netting, 1 May 2015,  $1 \Diamond 1 \heartsuit$ ; Ban Bo sane, Thap Put, Phangnga, (8°27'29.7"N 98°36'17.8"E), sweep netting, 13 February 2015;  $4 \heartsuit$ ; Khlong Chi Lat, Sai Thai, Muang, krabi, (8°03'23.5"N 98°53'38.2"E), sweep netting, 21 February 2015;  $1 \Diamond 2 \heartsuit$ ; Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh.

**Remarks.** The *Paraclius digitatus* species, was a medium body size of long-legged flies (body length 4.6–4.8 mm) that was firstly reported from the Island Singapore, and very easily recognized by distinctive male genital features. Antenna dark yellow, first flagellomere brown, 2.0 times as long as wide. Mid femur with 8 ventral bristles; hind femur with row of black ventral bristles (Zhang et al., 2007).

Distribution. Satun, Krabi, Phangnga (Andaman Sea), Surat Thani (Gulf of Thailand)

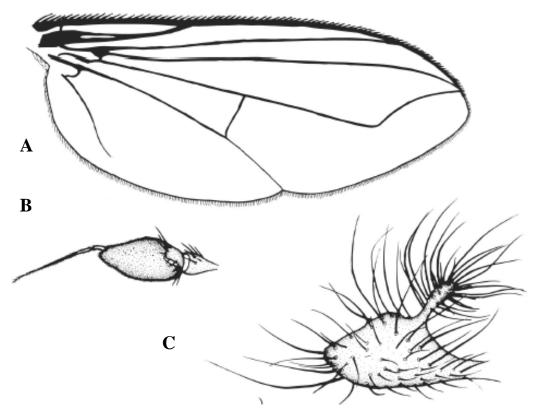


Figure 22. *Paraclius digitatus* Zhang et al., male habitus; wing (A), antenna (B), cercus (C) (modified from Zhang et al., 2007).



Figure 23. Male habitus, Paraclius digitatus Zhang et al., 2007

# Paraclius obtus Zhang et al., 2007

(New record, fig. 24)

**Materials Examined.**  $4^{\circ}$ ; Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), Malaise trap, 17 February 2015, coll. A. Samoh

**Remarks.** Male: Body length 4.7–4.8 mm, wing length 3.9–4.0 mm. Antenna dark yellow, first flagellomere with narrow dark dorsal margin, ventral margin yellowish, 2.3 times as long as wide (Zhang et al., 2007). Mid femur at its base with 3–4 black v (slightly longer than femur is wide) and with a row of 14 spinules on posterior ventral margin. It was mostly resembling to *Paraclis polychaetus*, in having the first flagellomere elongated, more than two times as long as wide, and the aedeagus with denticles, but may be separated from the latter by the cercus with the obtuse ventral apex.

**Distribution.** Chumphon (Gulf of Thailand)

#### Paraclius serratus Zhang et al., 2007

(New record, figs. 25, 26)

**Materials Examined.**  $3\overset{1}{\diamond}1^{\circ}$ ; Thailand, Ban Nua Nam, Phumriang, Chaiya, Surat Thani, (9°23'34.0"N 99°15'24.0"E), Malaise trap, 20 April 2015, coll. A. Samoh;  $1\overset{1}{\diamond}3^{\circ}$ ; Ban Khao Than, Tha Chang, Surat Thani, (9°19'43.4"N 99°12'31.6"E), 21 April 2015, coll. A. Samoh;  $1\overset{1}{\diamond}9^{\circ}$ ; Prince of Songkla University, Muang, Pattani, (6°53'04.9"N 101°14'10.1"E), sweep netting, 11 July 2014, coll. A. Samoh;  $11\overset{1}{\diamond}15^{\circ}$ ; Ban Pak Phanang Tawantok, Pak Phanang, Nakhon Si Thammarat, (8°22'30.2"N 100°10'00.4"E), sweep netting, 1 May 2015, coll. A. Samoh.

**Remarks.** Male: Body length 4.6–5.2 mm, wing length 3.9–4.2 mm. Antenna with first flagellomere 1.5 times as long as wide (Zhang et al., 2007). Mid and hind femora with row of 10 ventral bristles (shorter than femur is wide). Cercus with a round ventral margin.

Distribution. Surat Thani, Nakhon Si Thammarat, Pattani (Gulf of Thailand)

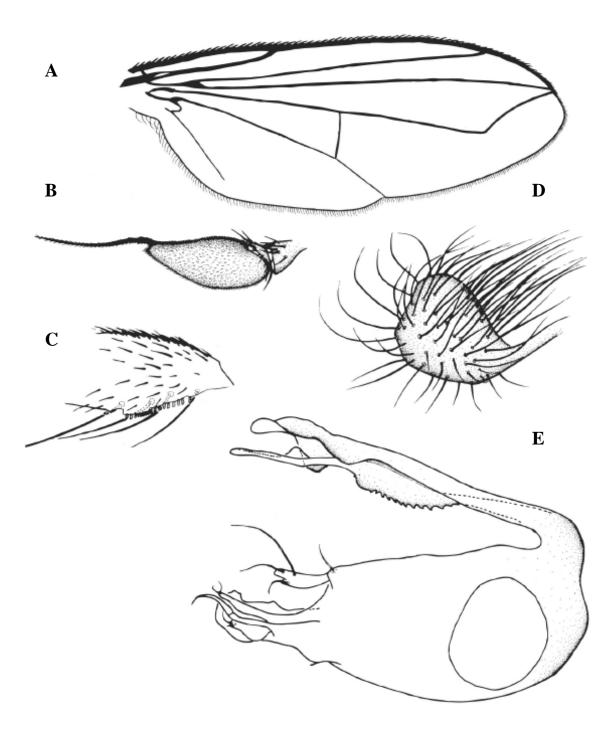


Figure 24. *Paraclius obtus* Zhang et al., male habitus; wing (A), antenna (B), apex of mid femur (C), cercus (D), male genitalia (E) (modified from Zhang et al., 2007)

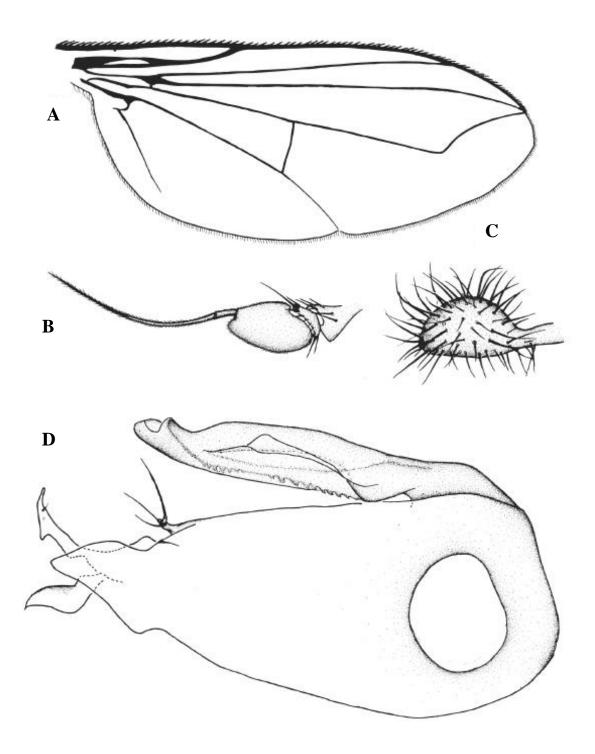


Figure 25. *Paraclius serratus*, male habitus; wing (A), antenna (B), cercus (C), male genitalia (D) (modified from Zhang et al., 2007)



Figure 26. Male habitus, Paraclius serratus Zhang et al., 2007.

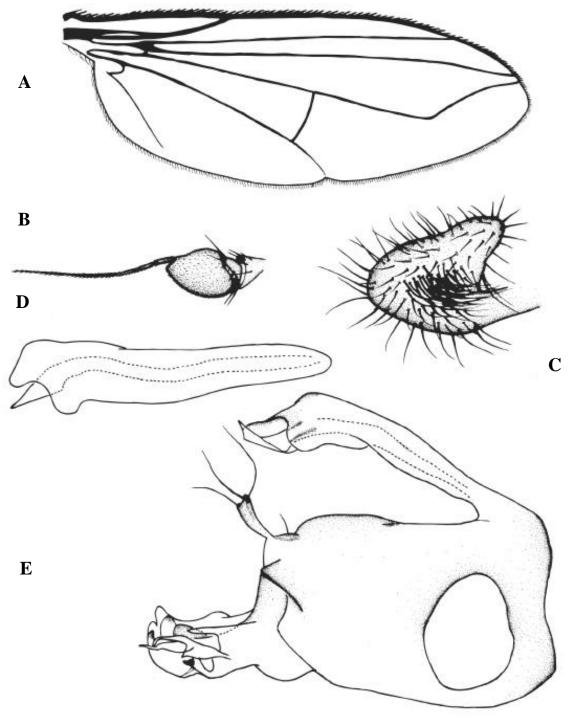


Figure 27. *Paraclius singaporensis* Zhang et al., male habitus; wing (A), antenna (B), cercus (C), hypandrium in ventral view (D), male genitalia (E) (modified fro Zhang et al., 2007).



Figure 28. Male habitus, Paraclius singaporensis Zhang et al., 2007.

#### Tachytrechus Stannius, 1831

#### (New record)

*Tachytrecus* Stannius, 1831. Erroneously treated as a nomen nudum by Foote et al. (1965), Robinson (1970b), Dyte (1975), Dyte and Smith (1980), Bickel and Dyte (1989), Negrobov (1991), and Sabrosky (1999), Type species: *Ammobates notatus* Stannius [Palaeartic], designation by Rondani, 1856 from species first include by Stannius (1831)

*Tetrecus*, error by Van Duzee (1924)

Tachyterechus, subsequent misspelling by Dyte (1975)

**Remarks and Recognition**. Globally, with 160 species were recorded from all zoogeographical regions (Grichanov, 1998), including seven species from Oriental region (Dyte, 1975), but they are mostly distributed in the Neotropical region (Brooks, 2005). Whether this genus has been also reported such a largely distribution in old world but from Thailand is unknown. Surprisingly, in this study we collected a species from two sides of peninsular Thailand.

Genus *Tachytrecus* is a part of the clade that includes *Cheiromyia*, *Paraclius*, *Stenopygium*, *Pelastonuerus*, and *Platyopsis* based on the loss of the hypandrial apodeme. For the generic concept, *Tachetrecus*, *Syntomoneurum*, *Goninoneurum* were clustered and grouped (Brooks, 2005). But Becker (1922) was placed *Syntomoneurum* into the subfamily Hydrophorinae. While Ulrich (1981) considered it to be closely related to *Tachytrecus* and transferred it to the Dolichopodinae. In addition, Brooks and Wheeler (2002) confirmed Ulrich's hypothesis of a closely related relationships (congeneric relationships) between genus *Tachytrecus* and genus *Syntomoneurum* by cladistics analysis.

Furthermore, *Tachytrechus* differ from the related genera of Dolichopodinae in the combination of characters: strong anterodorsal setae in apical half of the hind femur in addition to the true anterior subapical seta; the face is narrowed under antennae and somewhat widened towards clypeus; wing vein  $M_{1+2}$  usually has gentle curvation before the middle of distal part, running towards  $R_{4+5}$  and reaching costa far before the tip of wing; arista is short and bare; first flagellomere is usually short and suboval. Plus, with the distinctive upturned and flared postgonite of the male genitalia.

### Tachytrechus tessellatus Macquart, 1842

(New record, Figure 29)

**Materials Examined.**  $2 \cancel{3} 2 \cancel{2}$ ; Thailand, Ao Phanangtak, Muang, Chumphon Province (N 10°31'47.3', E 99°14'12.2'), sweep net, 17 February 2015, coll. A. Samoh;  $5\cancel{3} 5\cancel{2}$ ; Na Thab, Chana, Songkhla (7°01'25.8"N 100°43'05.0"E), sweep netting, coll. A. Samoh.

**Remarks.** *Tachytrechus tessellatus* is a large size Dolichopodinae and highly active in open, sunny, and wet habitats. They generally observed near shallow pools, tide pools. It can be recognized by the peculiar clypeus which mostly extends beyond the lower eye margin and is rounded below. Moreover, by the distinctive upturned and flared postgonite of male genitalia. This species has a wide range of distribution and is recorded from our region, India, to African continent (Yang et al., 2006). This study, we largely collected near tide pools at Tarutao Island, Satun province (Andaman Sea, Indian Ocean) and at shallow pools near the beach at Ban Na thab, Chana district, Songkhla province (Gulf of Thailand, South China Sea).

**Distribution**. Tarutao island, Satun (Andaman Sea), Na Thab, Chana; Ao Phanangtak, Chumphon (Gulf of Thailand).



Figure 29. Male habitus, *Tachytrechus tessellatus* Macquart, 1842.

#### Phoomyia Naglis and Grootaert, 2003

**Remarks.** *Phoomyia* or non-metallic dolichopodine fly, is usually found at beaches near front sea and easily recognised by these following combination characters: head and thorax grey, non-metallic body, more than these, one of highly distinctive character is abdomen mostly yellow or brown with grey pruinosity. In Thailand, only one species (*Phoomyia thailandensis*) was recoded from Gulf of Thailand (Naglis et al., 2013).

### Phoomyia singaporensis

#### (New record)

**Materials Examined.**  $2 \stackrel{\circ}{\circ} 2 \stackrel{\circ}{\ominus}$ ; Thailand, Talumpuk Cape, Pak Phanang, Nakhon Si Thammarat, (8°31'06.1"N 100°06'51.6"E), sweep netting, 30 April 2015, coll. A. Samoh;  $2 \stackrel{\circ}{\circ} 4 \stackrel{\circ}{\ominus}$ ; Tarutao Island, Langu, Satun, (6°44'19.2"N 99°38'45.4"E), sweep netting, 9 January 2015, coll. A. Samoh

**Remarks.** This species was collected from ghost crab burrows, bare sand at Tarutao Island, Satun province, Andaman Sea.

Distribution. Satun (Andaman Sea), Nakhon Si Thammarat (Gulf of Thailand)

#### Phoomyia talumpukensis sp. nov.

(New species)

**Materials Examined.**  $6 \cancel{3} \cancel{2}$ ; Thailand, Talumpuk Cape, Pak Phanang, Nakhon Si Thammarat, (8°31'06.1"N 100°06'51.6"E), sweep netting, 30 April 2015, coll. A. Samoh.

**Remarks.** The mature one of medium size species (body 2.7 mm, wing 2.7 mm.) of true beach-dwelling dolichopodine is very similar to *Argyrochlamys*. It seemed to be restricted to ghost crab burrows on the bare sand and the more humid at lower supralittoral zone. From our surveyed at Talumpuk cape, Pak Phanang district, Naknon Si Thammarat province, Gulf of Thailand, this species mostly observerd near ghost crab burrows and the biotope conditions were concordant with the mention of Naglis et al. in 2013.

**Distribution.** Talumpuk bay, Pak Phanang, Nakhon Si Thammarat province (Gulf of Thailand)

#### **SUBFAMILY HYDROPHORINAE Lioy, 1864**

#### Cymatopus Kertész, 1901

#### Systematic Account

#### Key to male Cymatopus and Thambemyia in Thailand

#### Type species Cymatopus tibialis, Kertész, 1901

**Remarks.** – This hydrophorine fly is medium in body size. With dark brown to black in body colour. Face broad. Clypeus broadening downwards. Palpi small and bristled. Rostrum very stout. Postoculars uniseriate above. Occiput convex, and eyes pubescent. Furthermore, thorax dusted, with a distinct flattened posterior slope (Meuffels and Grootaert, 1984). They are mostly composed with unique chaetotaxy pattern across the legs: no acrostichal, 4-5 strong dorsocentrals, and 1 humeral. While forelegs are raptorial in shaped. Femora in male apparently thickened than female and mostly ornamented with several types of bristles called male secondary sexual characters (MSSCs) (Figure 30B). Wing narrow, nearly as long as thorax and abdomen together. Mostly hyaline with dark brown vein and gradually changed to yellow towards base.

Costa shortly spinulose. Second and third longitudinal veins  $(R_{2+3})$  generally close together, sometimes parallel (Figure 30A) (Meuffels and Grootaert, 1984). Interestingly, this genus is largely live at rocky shores of the seacoasts.

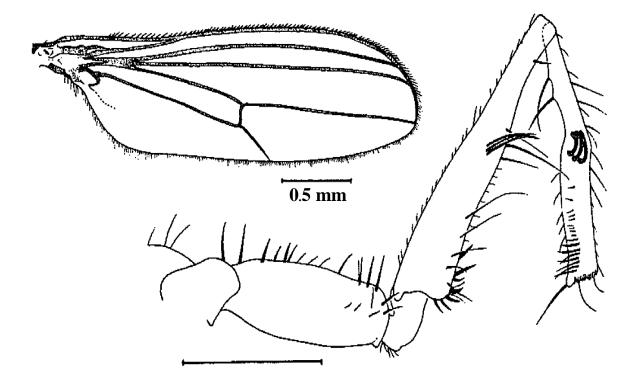


Figure 30. *Cymatopus leopoldi*, male habitus, (A) wing, (B) front legs in anterior view (modified from Meuffels and Gootaert, 1984)

#### The malayensis-group

Only one species recorded from Thailand.

### Cymatopus malayensis Parent, 1935

(Figure 31)

**Materials examined.** – 16 males, 16 females, Laem Kho Kwang, Chumphon Province, Gulf of Thailand (South China Sea), 17 February 2015, sweep netting, 10°30'48.7"N, 99°15'52.0"E; 13 male, 13 females, Ban Thong Tom Yai, Sawee, Chumphon Province,

Gulf of Thailand, 19 February 2015, sweep netting, 10°12'39.2"N 99°12'21.4"E; 8 males, 4 males, Tarutao Island, Langu, Satun Province, Andaman Sea (Indian Ocean), 9 January 2015, sweep netting, 6°44'19.2"N 99°38'45.4"E, coll. Abdulloh Samoh.

**Remarks.** – This species is discovered from both sides of peninsular Thailand. It seems particularly live at rocky shores of front sea. Previously, this species was recorded from the Malay peninsula (Evenhuis and Grootaert, 2002) along the seacoasts, Island of Borneo, and deeply distributed to the Island of Singapore and Malaysia (Pulau Hantu and Pulau Tioman). Rocky seashores with high salinity seemed to be a preferred biotope for this species.

**Distribution.** Laem Kho Kwang, Chumphon Province, Gulf of Thailand (South China Sea); Ban Thong Tom Yai, Sawee, Chumphon province, Gulf of Thailand; Tarutao Island, Langu, Satun, Andaman Sea (Indian Ocean). Moreover, this species has also been reported from Borneo, Singapore, and Malaysia.

## The *thaicus*-group

The *thaicus*-group is just based on the smaller size of the specimens and the shape of the wings. The group-name is provisionally conserved as such. It is represented by two species in Thailand: *C. thaicus* and *C. mayakunae* new species

## Cymatopus thaicus Grootaert & Meuffels, 2001

### (fig.34, 35)

**Materials examined.** – 4 males, Lidi Island, Langu, Satun Province (Andaman Sea), 6°46'56.4"N, 99°45'58.5"E, 30 July 2015, sweep netting, coll. A. Samoh; 12 male, 4 females, Tarutao Island, Langu district, Satun Province (Andaman Sea), 6°44'19.2"N 99°38'45.4"E, 9 January 2015, sweep netting, coll. A. Samoh.

**Remarks.** – The holotype of small species (body length: 2-2.1 mm; wing length: 2.25 mm.) is firstly described from Phang Nga province, Andaman sea (Grootaert and Meuffels, 2001), with yellow legs, fore tibia with a black foliaceous bristle and without spur, but a long black apical bristle. Fourth tarsal segment laterally flattened but not excavated as equal as terminal segment. Closely related to *Cymatopus calcaratus* Parent, 1935 and *C. calcaratoides* Grootaert and Meuffels, 1993, but both have a long apical spur on fore tibia.

Distribution. Satun Province (Andaman Sea).

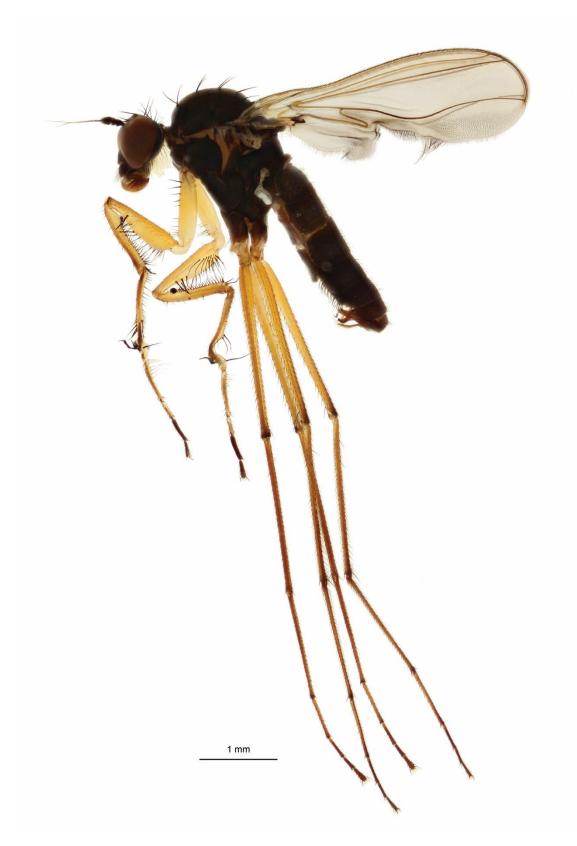


Figure 31. Male habitus, Cymatopus malayensis Parent



Figure 32. Female habitus, Cymatopus malayensis Parent.

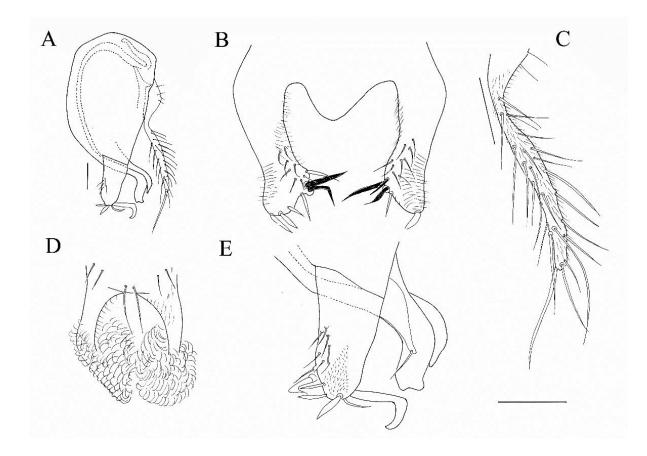


Figure 33. *Cymatopus thaicus* Grootaert & Meuffels male terminalia. A. Genital capsule in lateral view; B. surstyli dorsal view; C. Cercus lateral; D. Extension on sternite 6; E. Detail surstylus in lateral view with tip hypandrium and aedeagus. Scale 0.1 mm.



Figure 34. Male habitus, Cymatopus thaicus Grootaert & Meuffels.

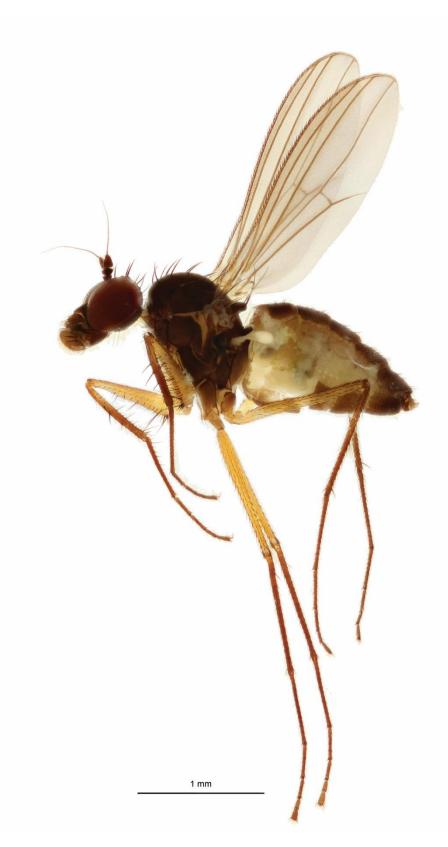


Figure 35. Female habitus, Cymatopus thaicus Grootaert & Meuffels

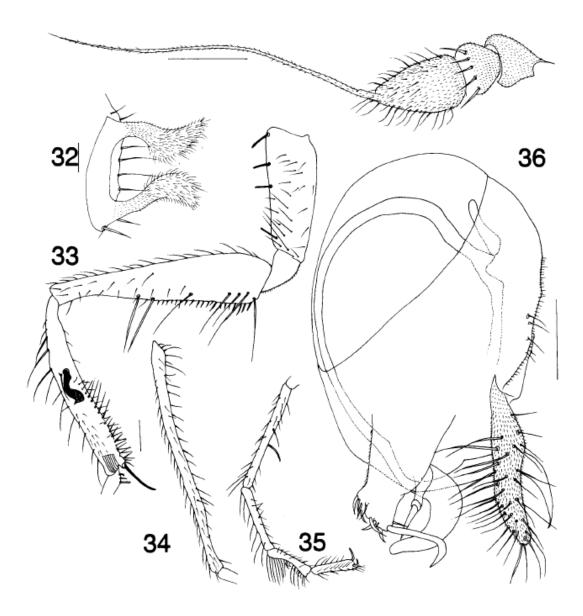


Figure 36. Male habitus, *Cymatopus thaicus*: antenna (A), fore femur and tibia anteriorly (B),

### Cymatopus mayakunae sp. nov.

(New species, pending for description, Figure 34)

**Materials examined**. - Holotype male: THAILAND, Laem Pakarang, Khao Lak, Takuapa, Phanga Nga Province (Andaman Sea), 8°44'09.9"N, 98°13'21.5"E, 10 February 2015, sweep netting, coll. A. Samoh.

Paratypes: 30 males, 10 females, same collection as holotype. 13 males, 6 females, Tarutao Island, Langu, Satun Province (Andaman Sea), 6°44'19.2"N 99°38'45.4"E, 9 January 2015, sweep netting, coll. A. Samoh.

**Diagnosis**. A small species (2.0-2.2 mm) with yellow legs. Fore tibia without black foliaceous bristle, without apical spur and apical bristle. Hind tibia with a dorsal row of bristles with dilated tips. Wing with veins  $R_1$  and  $R_{2+3}$  deformed and thickened. Posterior wing border a little deformed with longer hairs.

## Remarks.

Male

Body length 2.5 mm; wing length 2.5 mm.

Head. Frons and face black in ground-colour, greyish dusted. Clypeus protruding. Face wider than postpedicel is wide. Palpus brown with short black hairs, tips of apical bristles pale. A pair of strong ocellars, a pair of slightly shorter fronto-orbitals and a pair of minute postocellars. Postocular bristles black above, becoming whitish and hair-like below. Antenna black, pedicel darker than scape and postpedicel. Postpedicel conical, 1.5 times as long as wide. Arista nearly twice as long as scape, pedicel and postpedicel together.

Thorax black in ground-colour, greyish dusted. No acrostichals, 5 dc (anterior 4 equally long, prescutellar dc longer); a pair of long scutellars with a minute hair at outside. A minute humeral, a very long posthumeral, a short sutural, a minute notopleural, and a longer supra-alar and a long postalar. 3 pale propleurals.

Legs yellow (Figure 37) with mid and hind coxae black, apical two tarsomeres slightly brownish. Fore leg. Coxa with 2-3 short black bristles at base and some longer black apical bristles. Femur swollen in basal half with a long posteroventral bristles, near base as long as femur is wide, in apical half longer than femur is wide. The row is interrupted at the basal third and there 2 shorter bristles. Tibia as long as femur, without apical spur and without apical spine-like bristle; ventrally set with a double row of spine-like bristles as long as tibia is wide; basal fifth of tibia dorsally set with a double row of short bristles

with curved tip. Tarsomeres not flattened. Mid leg. Coxa with 2 short black exterior bristles. Femur much longer and thinner than fore femur. Tibia shorter than femur without particular bristles. Hind leg. Coxa with a short black exterior bristle. Femur a little wider than mid femur and shorter. Tibia dorsally set with a double row of short bristles with enlarged tips (Figure 37).

Wing brownish tinged with brown veins. Costa near middle darker brown and slightly bowed.  $R_{2+3}$  (Figure 37, 38) thickened and undulating near middle. Apical half of Cu pale, the hind border is a little notched there and the wing membrane is folded to the exterior and bears some longer bristles at that level. Haltere and squama white, bearing long with cilia.

Abdomen black in ground-colour, greyish dusted. Tergites with minute black on apical border. Terminalia (Figure 37): Cercus yellow with brown bristles longer than cercus is wide.

Female

Identical to male but fore legs with shorter bristles and wing with veins not deformed.

Distribution. Shores of the Andaman sea, Phang Nga province.

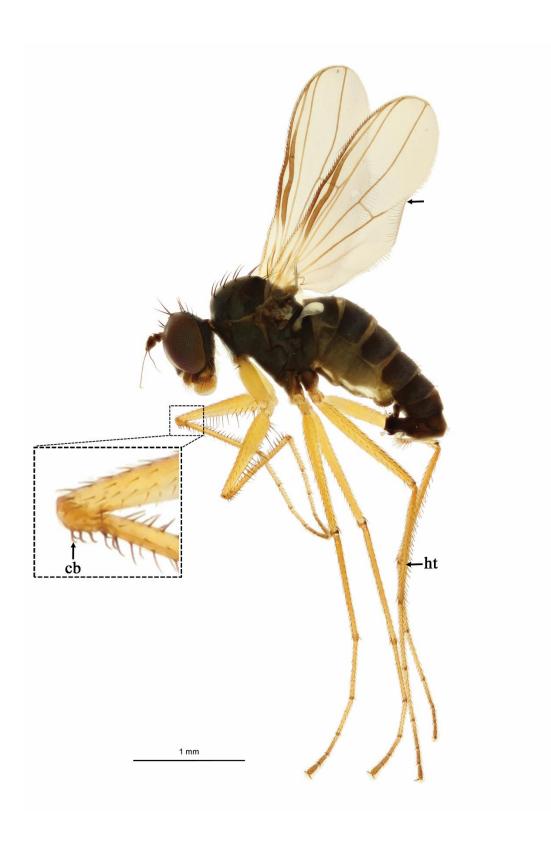


Figure 37. Male habitus, Cymatopus mayakunae sp. nov.



Figure 38. Female habitus, Cymatopus mayakunae sp. nov.

#### Nanothinophilus Grootaert & Meuffels, 1998

**Remarks**. – This genus is firstly recorded from Andaman seacoast in 1998. However, here is the first recorded from South China Sea side (Surat Thani province, Gulf of Thailand). The known species in Thailand are including *N. armatus* Grootaert and Meuffels, 1998, *N. pauperculus* Grootaert and Meuffels, 1998, *N. dolichurus* Grootaert and Meuffels, 1998, and *N. hoplites* Grootaert and Meuffels, 2001.

### Key to male Nanothinophilus from Thailand

brownish hair-like bristles...... *N. dolichurus* Grootaert & Meuffels

### Nanothinophilus hoplites Grootaert & Meuffels, 2001

### (Figure 36)

**Materials examined**.  $2\sqrt[3}69$ ; Thailand, Ban Elet, Pak Nam, Muang, Chumphon Province, (N 10°30'28.7', E 99°14'29.8'), Malaise trap, 16 February 2015, coll. A. Samoh;  $2\sqrt[3}29$ ; Phanangtak, Muang, Chumphon, (N 10°30'23.9', E 99°13'55.6'), Malaise trap, 17 February 2015;  $3\sqrt[3}49$ ; Ban Nua Nam, Phumriang, Chaiya, Surat Thani Province, (9°23'34.0"N 99°15'24.0"E), sweep netting, 20 April 2015, coll. A. Samoh;  $8\sqrt[3}29$ ; Ban Khao Than, Tha Chang, Surat Thani Province, (9°19'43.4"N 99°12'31.6"E), sweep netting, 21 April 2015, coll. A. Samoh;  $8\sqrt[3}19$ ; Laem Pho, Phumriang, Chaiya, Surat Thani Province, (9°22'33.6"N 99°16'00.3"E), sweep netting, 21 April 2015, coll. A. Samoh;  $8\sqrt[3}19$ ; Laem Pho, Phumriang, Chaiya, Surat Thani Province, (9°22'33.6"N 99°16'00.3"E), sweep netting, 21 April 2015, coll. A. Samoh;  $8\sqrt[3}19$ ; Laem Pho, Phumriang, Chaiya, Surat Thani Province, (9°22'33.6"N 99°16'00.3"E), sweep netting, 21 April 2015, coll. A. Samoh;  $8\sqrt[3}19$ ; Laem Pho, Phumriang, Chaiya, Surat Thani Province, (9°22'33.6"N 99°16'00.3"E), sweep netting, 21 April 2015, coll. A. Samoh;  $8\sqrt[3}19$ ; Laem Pho, Phumriang, Chaiya, Surat Thani Province, (9°22'33.6"N 99°16'00.3"E), sweep netting, 21 April 2015, coll. A. Samoh;  $35\sqrt[3}$ , 509, Tanjong Po (6°36'59.5"N 99°57'23.9"E), 6 May 2015, sweep netting, coll.A. Samoh.

**Remarks**. – The holotype of *Nanothinophilus hoplites* was collected from mudflat in mangroves at Ao Nang, Krabi, Andaman Sea (Indian Ocean), during low tide in 1997 by Grootaert and Meuffels (Grootaert and Meuffels, 2001). This tiny species (body length, 1.55-1.6 mm; wing length:1.3 mm) is resembled to *N. armatus*, but chaetotaxy pattern are

entirely different. Fore tibia of *N. hoplites* is contained with one row of 4-5 long, strong, and bent bristles. Fore femur ventrally with bristles which are longer than diameter of femur. Moreover, the hypopygium reaching beyond base of fourth abdominal segment (Figure. 35).

**Distribution.** Chumphon, Surat Thani (Gulf of Thailand), Satun, Phang Nga, Krabi (Andaman Sea).

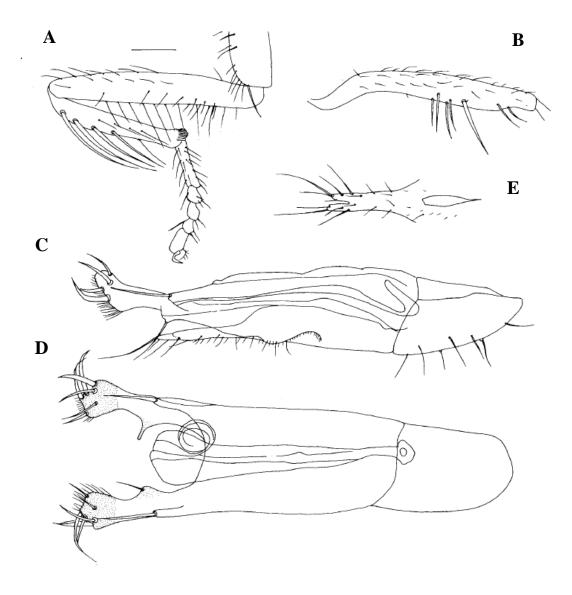


Figure 35. Male habitus, *Nanotinophilus hoplites* Grootaert and Meuffels, fore leg anteriorly (A), hind leg anteriorly (B), hypopygium laterally (C), hypopygium ventrally (D), cerci, sacle is 0.1 mm (modified from Grootaert and Meuffels, 2001).



Figure 36. Male habitus, Nanothinophilus hoplites Grootaert & Meuffels, 2001.

# Nanothinophilus pauperculus Grootaert & Meuffels, 1998

**Materials Examined.** – 58 $\stackrel{\circ}{\circ}$  67 $\stackrel{\circ}{\ominus}$ ; Thailand: Khlong Chilat, Ban Sai Thai, Muang, Krabi Province, (8°03'23.5"N 98°53'38.2"E), Malaise trap, 27 February 2015, coll. A. Samoh.

**Remarks.** – This remarkable small species was first recoded from Ranong province, Andaman Sea by Grootaert and Meuffels in 2001. This study was extended found from great mangrove in Krabi city. The body size rather smaller than *N. hoplites* (body size:1.5 mm, wing length: 1.25 mm.)

Distribution. Krabi (Andaman Sea)



Figure 37. Nanothinophilus pauperculus Grootaert & Meuffels, 1998.

### Thambemyia Oldroyd, 1956

Thambemyia Oldroyd, 1956. Type-species: T. pagdeni Oldroyd (original designation).

Subgenus Thambemyia Oldroyd, 1956.

**Remarks**. – The type species of this genus is first designated by Oldroyd in 1956 as a monotypic genus. *Thambeyia* Oldroyd, 1956, *Acymatopus* Takagi, 1965, *Conchopus* Takagi, 1965 are more closely related to each other than *Cymatopus* Kertész, 1901. There are widely distributed from Southeast Asia (Brunei, Malaysia, Thailand), Hong Kong, Taiwan, China to Japan. In Thailand, there are recently recorded from the Andaman sea coast, Pakbara, Satun.

## Thambemyia pagdeni Oldroyd, 1956

**Materials Examined**. – 23 males, 15 females, Sakom (Tepha), Songkhla Province, Gulf of Thailand (South China Sea), 28 March 2017, cliffs, 6°57'42,97"N 100°50'57.02"E; 29 March 201, pier on sandy beach 6°56'52,88"N 100°51'52.72"E.

1 male, 2 females Khao Lak, Nangtong, Phang-Nga Province, rocky beach (reg. 96050, leg. P. Grootaert).

**Remarks**. - Small metallic green with grayish white pollinosity long-legged flies such *Thambemyia pagdeni* was collected firstly by light traps in Malaysia (Masunaga, 2005). *Thambemyia* is similar in external appearance to the species of *Conchopus* (as a synonym of *Conchopus*) that do not belong to the rectus group of that genus. It is distinguished readily from the non-rectus group of *Conchopus* by the following combination of characters: presence of gena, absence of posterior notopleural bristles, white pollinosity of male mesonotum weak, metatarsus weakly modified, female postabdomen extensively setose on sixth and seventh segments, female cercus weakly sclerotized, female tenth abdominal tergum with two pairs of spine-like setae, and female paraproct reduced.

Distribution. Sakom, Songkhla (Gulf of Thailand), Tarutao Island, Satun (Andaman Sea)

## Thinophilus Wahlberg, 1844

*Thinophilus* Wahlberg, 1844: 37. Type species: *Rhaphium flavipalpe* Zetterstedt, 1843 (monotypy).

*Parathinophilus* Parent, 1932: 161. Type species: *Parathinophilus expolitus* Parent, 1932 (monotypy).

## Systematic Account

## Key to male Thinophilus from the Thai-Malay Peninsula

1) Wing with dark spot on middle of apical section of  $M_{1+2}$  (level of wing boss), on cross vein and sometimes on vein  $R_{4+5}$ , if the clouding on the veins is weak: male with a tuft of long bristles on sternite 3 and 4 .....**T.** setiventris Grootaert & Meuffels, 2001

255

- Wing without spots and sternites with at most short hairs ......2

- Fore coxa black. Fore femur weakly dilated at base. Fore tibia a little shorter than fore femur (Fig. 30) Fore tarsomere 1 about as long as fore tibia. Fore tarsomere 3 not paler than preceding tarsomeres. Fore tarsomeres 4 and 5 not widened, black.....

.....*T. spinatus* sp. nov.

5) Fore coxae darkened on basal half or completely darkened (variabilis sp. nov. usually has yellow fore coxa, but they might be brownish infuscate
- Fore coxae completely yellow (except for extreme base)11
6) Tarsomere 2 of mid leg with a shield-like dorsal black protuberance (Figs 16–17), tarsomere 3 white
- Tarsomere 2 of mid leg without dorsal protuberance
7) Mid femur at base with a cluster of distinct black ventral bristles (Fig. 16). Hind femur with ventral bristles in apical half shorter than femur is wide (Fig. 16)

8) All femora darkened, if femora yellow, fore femur with long white curly bristles at base. Tip of fore tibia and all tarsomeres 5 darkened at tip. Hypopygium short, less than one-third length of abdomen (Fig. 1) ......**T. boonrotpongi** sp. nov.

9) Fore coxa entirely black. Large robust species with distinctly bristles legs ......10

- Larger species. Fore tibia without strong brown posteroventral bristles near base

13) Fore coxa anteriorly near base with long, soft white bristles and a single black bristle; apical bristles black. Fore femur in basal half with a row of 4–5 ventral bristles about as long as femur is wide (all trochanters yellow). Fore tarsomere1 ventrally set with a row of black spinules; mid leg with apical tarsomeres 2 black (fresh water species) .....**T. nitens** Grootaert & Meuffels, 2001

## Thinophilus boonrotpongi, new species

(Fig. 38)

### Materials Examined.

### **Type material**

### Holotype

THAILAND: ♂, Sai Thai, Muang, Krabi Province, 8°03′23.5″ N, 98°53′38.2″ E, sweep netting,

A. Samoh leg., 27 February 2015 (NHM-PSU).

## Paratypes

THAILAND: 7  $\Im \Im$ , 10  $\Im \Im$ , Sai Thai, Muang, Krabi Province, 8°03'23.5" N, 98°53'38.2" E, sweep netting, A. Samoh leg., 27 February 2015; 1  $\Im$ , 7  $\Im \Im$ , Khlong Phon, Khlong Thom, Krabi province, 7°48'11.2" N, 99°10'11.9" E, sweep netting, A. Samoh leg., 13 June 2015; 1  $\Im$ , 1  $\Im$ , Ban Bakan Tohtid, Langu, Satun province, 6°47'29.8" N, 99°48'53.5" E, sweep netting, A. Samoh leg., 3 June 2015; 1  $\Im$  (with yellow femora), Ban Bakan Tohtid, Langu, Satun province, 6°47'29.8" N, 99°48'53.5" E, sweep netting, A. Samoh leg., 3  $\Im \Im$ , 2  $\Im \Im$ , 80°48'53.5" E, sweep netting, A. Samoh leg., 3  $\Im \Im$ , 2  $\Im \Im$ , 80°48'53.5" E, sweep netting, A. Samoh leg., 3 June 2015; 1  $\Im$  (with yellow femora), Ban Bakan Tohtid, Langu, Satun province, 6°47'29.8" N, 99°48'53.5" E, sweep netting, A. Samoh leg., 4 June 2015 (RBINS), 3  $\Im \Im$ , 2  $\Im \Im$ , 80°48'53.5" E, February 2015 (NHM-PSU).

## Diagnosis

A medium-sized species with black fore coxa bearing long white bristles. Apical half of fore tibia pale, almost white with black tip. Tip of all apical tarsomeres black.

## Etymology

This species is dedicated to Dr. Singtoe Boonrotpong, a promoter of my PhD thesis, in recognition of his help and support during the current project.

### Description

Male (Fig. 38A)

## Length. Body 3.5 mm; wing 2.8 mm. Diagnosis

A medium-sized species with black fore coxa bearing long white bristles. Apical half of fore tibia pale, almost white with black tip. Tip of all apical tarsomeres black.

**Head.** Frons and face with shiny dark metallic green ground colour. Face as wide as length of postpedicel. Clypeus about one third of epistoma, protruding. A pair of long divergent black ocellars. Two very short postocellars. A pair of convergent proclinate verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars.

Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna brownish at tip and above, yellowish below. Arista dorsal, twice as long as antenna, brown, bare. Basal article short. Palpus yellowish to brown with black bristly hairs. Proboscis dark brown.

**Thorax.** Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 equally long dc in one row, preceded by a short bristle and a longer prescutellar outside the row. Scutellum with 2 marginals, without lateral hairs. Two short white upper propleural bristles and 2 longer lower propleural bristles.

Legs. Brownish, but tibiae and tarsi pale. Fore coxa completely black; mid and hind coxae entirely black. All femora generally black. All tibiae with basal half brownish, becoming whitish towards tip. Fore tibia with black spot on tip ventrally. All tarsomeres whitish, but tip of terminal tarsomere black. Coxa anteriorly with long white bristles in apical half. Trochanter with long white bristles. Fore femur thickened in basal two-thirds. Ventrally at base with 2 rows of white bristles, longer than femur is wide, apical twothirds with few short black bristles; with 3 strong equally long posterior preapical bristles. Fore tibia shorter than femur, ventral bristles short; posteroventral bristles of tibia on basal third longer than following bristles. Tarsomere 1 densely set with spine-like bristles. Mid coxa: exterior bristles white and longer than coxa; anterior bristles long and white. Mid femur thinner than fore femur; with row of black ventral bristles, longer at base. Mid tibia with a long anterior dorsal at apical quarter; 2 dorsal and 2 pd; crown of apicals, ventral bristles longest. Hind coxa with short white exterior bristles. Hind femur a little thicker than mid femur; a long dorsal and anterodorsal bristle at apical third; row of black ventral bristles about as long as femur is wide. Hind tibia with 2 anterodorsal and 2 shorter dorsal bristles and a crown of long apicals. Hind tarsomere 1 long but shorter than tarsomere 2.

**Wings.** Uniformly brownish tinged, without spots. Tp straight, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.

**Abdomen.** Shining dark metallic green. Hairs and hind-marginal bristles on tergites short, black. Sternites with short white bristles.

**Terminalia** (Figs 38C–E). Phallus long strap-shaped. Cerci pale brownish with pale hairs; epandrium black. Cerci not fused (Fig. 38E).

Female (Fig. 38B)

**Length.** Body 3.6 mm, wing 3 mm. Larger than male. Similar to male except following characters. Clypeus 1/4 length of face, bulging. Fore coxa with short white bristles only; fore femur with minute bristles; mid and hind femora also with minute ventral bristles. Sternites with short white bristling.

Distribution. Southern Thailand, only known from Andaman Sea.

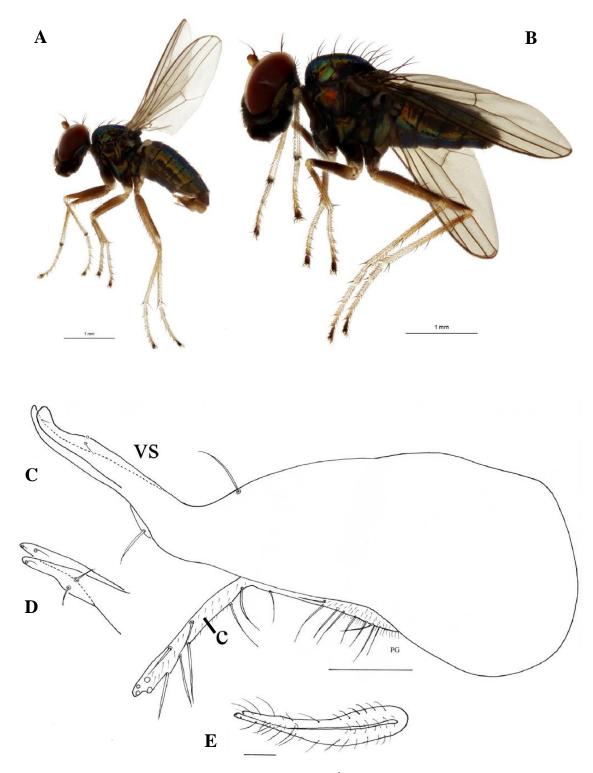


Figure 38. *Thinophilus boonrotpongi* sp. nov. (A).  $\mathcal{J}$ , habitus. (B).  $\mathcal{Q}$ , habitus, terminalia. (C). Genital capsule, lateral view. (D). Apex of surstylus, dorsal view. (E). Cerci, dorsal view.

### Remarks

*Thinophilus boonrotpongi* sp. nov. is quite unique in having the fore coxa black bearing long white bristles combined by having the apical half of the fore tibia almost white with a black apex. All apical tarsomeres are also darkened. Only T. nitens Grootaert & Meuffels, 2001 has white bristles on the fore coxa, with a single black bristle among them, but the fore coxa itself is yellow. We have seen one male specimen in the material examined, with all femora and tibiae yellow, that we attribute to *T. boonrotpongi* sp. nov. The tarsi are yellowish and not whitish (cf. Fig. 38). Other characters such, as the fore femur with long white soft bristles at the base, the general bristling of the legs and the male genitalia, also suggest that it represents *T. boonrotpongi* sp. nov. A future molecular analysis should ascertain if there is a genetic difference.

### Thinophilus langkawensis sp. nov.

(Figs. 40, 41, 41, 42)

### Diagnosis

A large species. Antenna completely yellow. Tibiae and tarsomeres completely yellowish white. Hypopygium elongate, more than half length of abdomen. Cerci in male reaching almost to thorax. Surstyli are movable and out folding surstyli with a veil like membrane.

### Etymology

The specific epithet refers to the island of Langkawi (Malaysia) where the species was found for the first time.

### **Type material**

### Holotype

THAILAND:  $\bigcirc$ , Ko Tarutao, Molae bay, Satun province, 6°40′21.0″ N, 99°38′20.9″ E, sweep netting, A. Samoh leg., 9 January 2015 (NHM-PSU).

### **Paratypes**

THAILAND: 5  $\Diamond \Diamond$ , 7  $\bigcirc \Diamond$ , ko Tarutao, Molae bay, Satun province, 6°40′21.0″ N, 99°38′20.9″ E, sweep netting, A. Samoh leg., 9 January 2015 (NHM-PSU) (1  $\Diamond$  and 1  $\bigcirc$  in RBINS).

### Description

### Male (Fig. 39)

Length. Body 6.4 mm; wing 5 mm.

**Head.** Frons and face with shiny dark metallic green ground colour. Face twice as wide as length of postpedicel. Clypeus a third of length of face. Ocellar tubercle pronounced but sunken between the eyes, not depassing eye borders (Fig. 39). A pair of long divergent black ocellars. No postocellars. A pair of convergent proclinate verticals, a little shorter than ocellars. Vertex excavated; postcranium metallic green. Two converging postverticals, stronger and longer than, and not in row with, upper postoculars. Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna yellow; pedicel and postpedicel hardly darkened dorsally. Arista dorsal, 2.5 times as long as antenna, not pubescent. Basal article short, yellowish brown; arista white, base a little browned. Palpus yellow with short white bristly hairs. Proboscis brown.

**Thorax**. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 5 equally long dc, prescuttelar a litle longer and dc row preceded by a short bristle. Scutellum with 2 marginals, and a short lateral bristle. 4 short white propleurals above and 7 longer white propleural bristles below.

**Legs.** Yellowish white including all tarsomeres. Fore coxa black on basal  $\frac{2}{3}$ , yellowish on apical third; mid and hind coxae brownish, apices pale. Fore coxa anteriorly with short white bristles. Trochanter bare. Fore femur narrower than mid femur. Ventrally almost bare, except for some minute white hairs; 2 short posterior preapical bristles. Fore tibia shorter than femur with only minute ventral bristles. Mid coxa with a long, black exterior bristle near middle, with short, white anterior bristles at tip. Mid femur wider than fore femur; ventrally with an anterior row of 3 short brown bristles and a posterior row of 5 bristles. Mid tibia as long as femur, with 3 short ad, 2 longer ad and 2 pd. Hind coxa with black exterior bristle and minute white anterior bristles. Hind femur only a little wider than mid femur; ventrally on apical  $\frac{2}{3}$  with a row of long white bristles, twice as long as femur is wide; in addition a few minute ventral bristles on basal third; 2 long black ad bristles on apical third. Hind tibia with 3 ad, 2 very long pd; a row of short black pd on basal third as long as tibia is wide; 2 somewhat recurved ventral bristles at basal third.

**Wings.** Clear, without spots. Tp straight, apical part of  $M_{3+4}$  1.5 times as long as Tp. Anal vein not reaching wing margin.

**Abdomen.** Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites with short pale hairs.

**Terminalia** (Figs 8–11). Elongate, more than half length of abdomen with surstyli reaching tip of sternite 3 but cerci almost reaching to base of thorax. Cerci pale yellowish (Fig. 39), ventrally not fused. Apex cercus with remarkable pattern of bristling (Fig. 11).

Surtylus movable connected by a veil-like membrane to the epandrium, suspended by black rod-like structures. Phallus long strap-shaped, but not coiled (Fig 10). Epandrium elongate, brown.

## Female (Fig. 40)

**Length.** Body 6.4 mm, wing 5.6 mm. Stouter than male otherwise similar except following characters. Hind femur lacking long white ventral bristles. Sternites with minute pale bristling.

Distribution. Southern Thailand and Malaysia (Andaman Sea).

# Remarks

The male of this robust species with yellow legs has very long terminalia that in rest position are partly hidden in a cavity formed by the sternites 4 to 6. When the terminalia are extended, the surstyli move and open a veil like lined cavity (Figs 8, 10). This phenomenon was not previously observed in *Thinophilus*. This large species was found on the adjacent islands of KoTarutau in Thailand and Langkawi Island in Malaysia.



Figure 39. Male habitus, Thinophilus langkawensis Samoh et al., 2017



Figure 40. Female habitus, Thinophilus langkawensis Samoh et al., 2017.

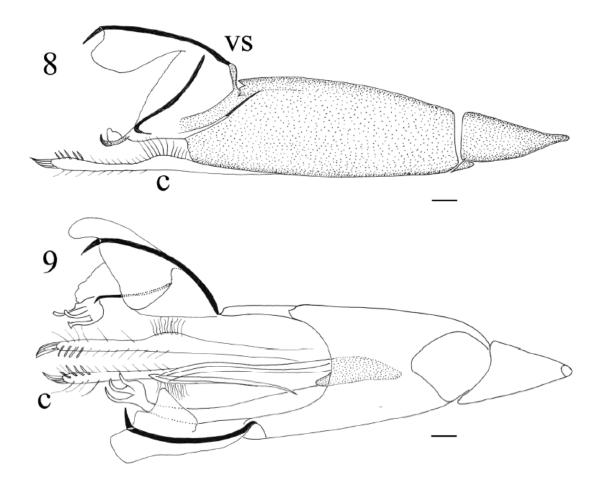


Figure 41. *Thinophilus langkawensis* sp. nov.,  $\Diamond$ , terminalia. 8. Genital capsule, lateral view. 9. Genital capsule, dorsal view.

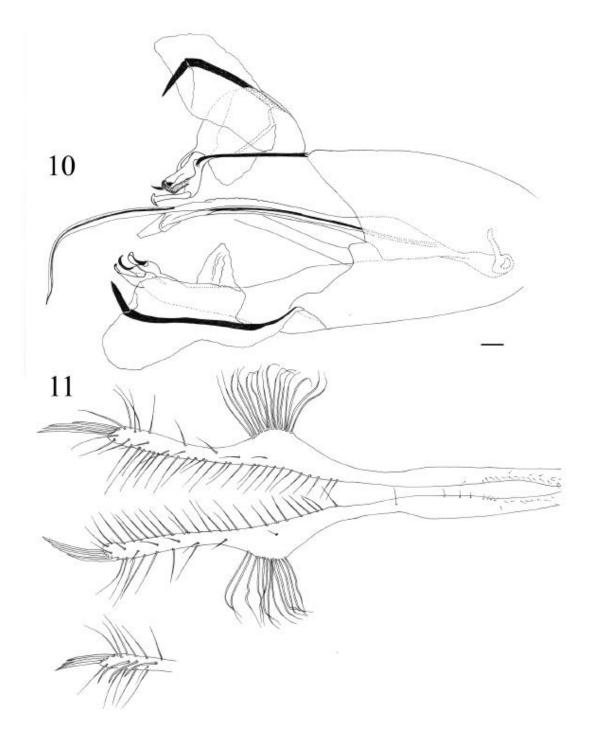


Figure 42. *Thinophilus langkawensis* sp. nov., ♂, terminalia. 10. Genital capsule and surstyli, ventral view. 11. Detail of apex of cerci, dorsal view.

## Thinophilus minutus, new species

(New species, Figure 43)

## Diagnosis

A small species with completely yellow antenna, yellow fore coxa, brown mid and hind coxa and legs further completely yellow. Fore tibia without a ventral row of spine like bristles. Only mid and hind femur with distinct black ventral bristles.

# Etymology

The specific epithet refers to the small size of the species.

# **Type material**

# Holotype

THAILAND: ♂, Ban Laem Son, Langu, Satun province, 6°56′27.9″ N, 99°42′12.4″ E, sweep netting, A. Samoh leg., 27 February 2015 (NHM-PSU).

# Paratypes

THAILAND: ♂, Ban Laem Son, Langu, Satun province, 6°56′27.9″ N, 99°42′12.4″ E, sweep netting, A. Samoh leg., 27 February 2015 (NHM-PSU); 1 ♂, Phanang Tak, Muang, Chumphon province, 10°30′23.9″ N, 99°13′55.6″ E, sweep netting, A. Samoh leg., 17 February 2015 (NHM-PSU); 1 ♂, Bang Yai, Bang Nai Si, Takuapa, Phang-Nga province, A. Samoh leg., 9 February 2015 (NHM-PSU).

# Description

Male (Fig. 43)

Length. Body 2.4 mm; wing 2 mm.

Head. Frons and face with shiny dark metallic green ground colour. A pair of long divergent black ocellars. Two very short postocellars. A pair of convergent proclinate long verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars. Postoculars uniseriate, black above, white and becoming multi-seriate below. Antenna pale brownish. Arista dorsal, 2.5 times as long as antenna, shortly pubescent. Basal article short. Palpus yellow, with short, black bristly hairs, only anteriorly. Proboscis dark brown.

Thorax. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 equally long dc in one row, preceded by a short bristle and prescutellar outside the row and hardly longer than preceding bristles. Scutellum with 2 marginals, without lateral hairs. 3 short lower pale brownish propleural bristles.

Legs. Yellow including all tarsomeres. Fore coxa yellowish white; mid and hind coxae entirely brownish. Fore coxa anteriorly with short brown bristles. Trochanter bare. Fore femur club shaped, a little thickened in basal half, apical half thin. No ventral bristles; 3 distinct posterior bristles on apical third. Fore tibia shorter than femur, no ventral bristling. First tarsomere densely set with spine-like bristles. Mid coxa with a long black exterior near middle and some long anterior bristles in basal half. Mid tibia with a short ad and pd in basal quarter and a short ad and pd near middle; a crown of short apical bristles. Hind coxa with a black exterior bristle. Hind femur wider and longer than mid femur; short ventral bristles, short, upright anterior bristles near middle. Hind tibia with 1 ad and 2 dorsal bristles, a crown of long apicals.

Wings. Yellowish brown, without spots. Tp straight, brownish seamed, apical part of M3+4 1.5 times as long as Tp. Anal vein not reaching wing margin.

Abdomen. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites with short brown hairs.

Terminalia (Figs 13–15). Phallus long strap-shaped (Fig. 13). Cercus whitish with long brown apical bristles (Figs 14–15), epandrium brown.

### Female. Unknown

**Distribution** Southern Thailand (Andaman Sea and Gulf of Thailand).

### Remarks

*Thinophilus minutus* sp. nov. is quite unique among *Thinophilus* in southern Thailand by having a few distinct bristles on the legs. Only mid and hind femora have distinctly longer ventral bristles. It is similar to *T. peninsularis* Parent, 1935, a sympatric species that also exhibits a few distinct characters on the legs. The latter species, however, has a dorsal bristle on the basal quarter of the fore tibia, lacking in *T. minutus* sp. nov. Further, it has the fore coxa darkened on basal two thirds and the apical tarsomere darkened as well. The fore coxa and even the apical tarsomere of all legs are yellow in *T. minutus* sp. nov. Finally, in *T. peninsularis* the first tarsomere of the fore leg is as long as the following tarsomeres together, while in *T. minutus* sp. nov. the first tarsomere is half as long as the following four tarsomeres together. Both species share a brownish tinged wing. In *T. minutus* sp. nov. the Tp and M are brownish seamed.

*Thinophilus minutus* sp. nov. should also be compared with *T. dongae* Grootaert et al., 2015, known from southern China. The latter species also has yellow fore coxae, no ventral bristles on fore femur, no ventral spinules or bristles on fore tibia. It has, however, the apical tarsomere of all legs black and mid and hind femora without ventral bristles. In *T. minutus* sp. nov. all tarsomeres are yellow and the mid and hind femora have short but distinct bristles. Both species are likely related forming a species-group as indicated by the similar shape of the cerci and surstyli.



Figure 43. Male habitus, *Thinophilus minutus* Samoh et al., 2017.

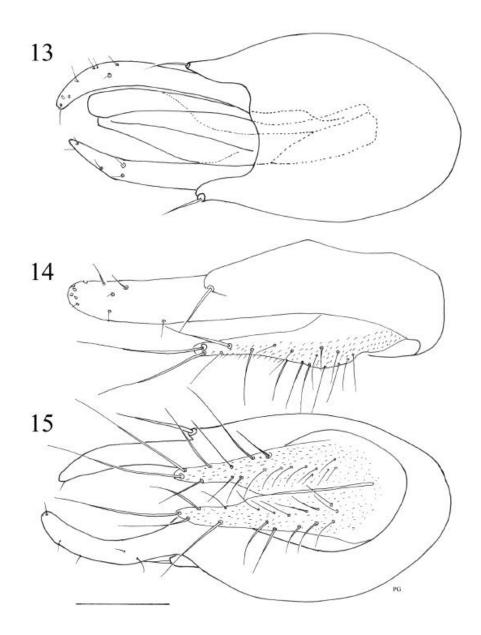


Figure 44. *Thinophilus minutus* sp. nov., ♂, terminalia. 13. Genital capsule, ventral view. 14. Genital capsule, lateral view. 15. Genital capsule, dorsal view.

# Thinophilus parmatoides, new species

(Figure XX)

# Diagnosis

A medium-sized species with a shield-like protuberance on mid tarsomere 2. Mid femur with a cluster of about 10 short spine-like ventral bristles at base.

# Etymology

The specific epithet refers to the resemblance with *T. parmatus* Grootaert & Meuffels, 2001, also described from southern Thailand.

# **Type material**

# Holotype

THAILAND: ♂, Pak Phanang Tawantok, Pak Phanang, Nakhon Sri Thammarat province, 8°24′09.4″ N, 100°11′29.9″ E, sweep netting, A. Samoh leg., 30 April 2015 (NHM-PSU).

# Paratypes

THAILAND: 7 33, 10 99, Pak Phanang Tawantok, Pak Phanang, Nakhon Sri Thammarat province, 8°24′09.4″ N, 100°11′29.9″ E, sweep netting, A. Samoh leg., 30 April 2015 (NHM-PSU; 2 33, 2 99 at RBINS).

# Description

Male (Fig. 16)

Length. Body 2.6 mm; wing 2.4 mm.

Head. Frons and face with shiny dark metallic green ground colour. Face above as wide as length of postpedicel, near middle half as wide as postpedicel. A pair of long divergent black ocellars. Two very short postocellars. A pair of minute verticals at level of ocellar tubercle. Vertex a little sunken. A pair of minute postverticals. 4 black upper postoculars, followed by a row of yellowish uniseriate lower postoculars. Antenna yellowish; only postpedicel dusky above. Arista subdorsal, 3.5 times as long as antenna, brown, with short pubescence. Basal article very short. Palpus yellowish brown, with a few fine black bristles along sides, centrally only minute bristles.

Thorax. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 short equally long dc, prescutellar twice as long as preceding dc. Scutellum with 2 long crossing marginals, and a short lateral bristle; without lateral bristle. No upper propleurals and a few very short lower propleurals.

Legs. Yellow, but fore coxa completely black, densely set with black bristles; mid and hind coxae brown. Fore and mid trochanters yellow, ventrally brown. Fore femur a little wider than mid femur, especially on basal half; ventrally near base with a few short bristles. Fore tibia longer than femur, with a ventral row of bristles over entire length longer than tibia is wide; bristles near middle longest. Mid coxa with a long, black exterior bristle, half as long as coxa is high; anterior bristles very dense, black. Mid femur with spindle shaped base; at base a cluster of about 10 black bristles (shorter than femur is wide). Mid tibia much longer than femur; without prominent bristles; ventrally in apical quarter with long hair-like bristles. Mid tarsomere 2 bearing a black shield-like dorsal extension; tarsomere 3 shorter than tarsomere 2, white (Fig. 16). Hind coxa with black exterior bristle. Hind femur a little spindle-shaped at base; ventrally in apical half with only 2 short black bristles. Hind tibia with a short ad near middle.

Wings. Brownish tinged, without spots. Tp straight, longer than apical part of  $M_{3+4}$ . Anal vein not reaching wing margin. Anal vein not reaching wing margin.

Abdomen. Shining dark metallic green. Hairs and hind-marginal bristles on tergites black. Sternites 2 and 3 with minute hairs; sternite 4 with a few longer black apical bristles.

Terminalia (Figs 18–20). Phallus long, strap-shaped. Cerci pale brownish with pale hairs; dorsally fused (Fig. 20); surstyli and epandrium a little darker than cerci.

# Female

Length. Body 2.9 mm, wing 2.6 mm. similar to male, except following characters. Mid femur without cluster of ventral bristles at base; mid tarsomere 2 without shield-like protuberance.

# Distribution

Southern Thailand (Gulf of Thailand).

# Remarks

This species is similar to *T. parmatus* in having a black shield like protuberance on tarsomere 2 of the mid leg. There are a few black bristles at the base of the fore femur, a thick tuft of black bristles at the base of the mid femur, long hair-like bristles on the tip of the mid tibia and only short ventral bristles on the hind femur. In *T. parmatus*, there is a single long bristle at the base of the fore femur, the mid femur has only 4 thin bristles at its base, the hind femur has longer bristles in the apical half. The shield on tarsomere 2 of the mid leg is rounded in *T. parmatoides* sp. nov., but elongated in *T. parmatus* (Fig. 17). The shape of the male genitalia is very similar in both species.

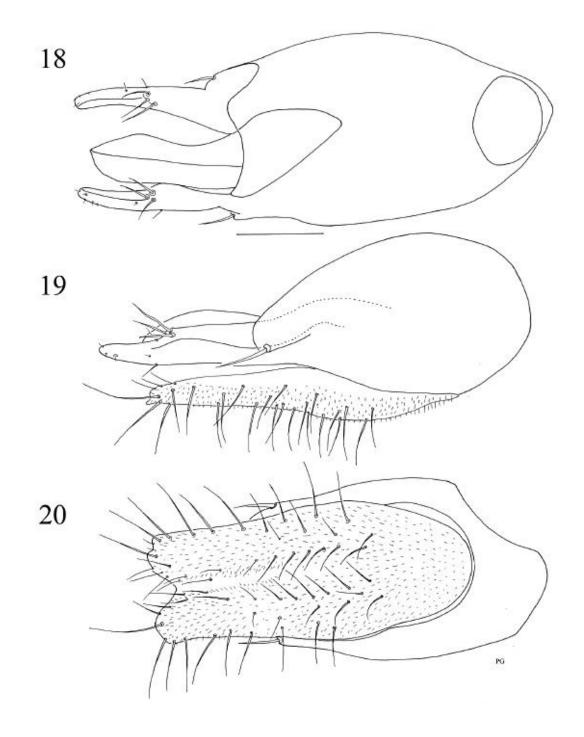


Figure 45. *Thinophilus parmatoides* sp. nov., ♂, terminalia. 18. Genital capsule, ventral view. 19. Genital capsule, lateral view. 20. Cerci, dorsal view.

# Thinophilus parvulus, new species

(New species, Figure 46)

# Diagnosis

A small species with fore tibia bearing a short and a long black posterodorsal bristle near base.

# Etymology

The species name is derived from the Latin '*parvulus*', referring to the very small size of the species.

# Type material

# Holotype

THAILAND: ♂, Muang, Pattani province, Prince of Songkhla University, Pattani campus, 6°53′04.9″ N, 101°14′10.1″ E, Malaise Trap, A. Samoh leg., 11 April 2015 (NHM-PSU).

# Description

Male (Fig. 21)

Length. Body 1.8 mm; wing 1.7 mm.

Head. Frons and face with shiny dark metallic green ground colour. Face at narrow down point wider than postpedicel. Clypeus about a quarter as long as face. A pair of long divergent black ocellars. No postocellars. A pair of convergent verticals, a little shorter than ocellars. Postcranium dark metallic green. Two converging postverticals, stronger and longer than, and not in row with upper postoculars. Postoculars uniseriate, black above and white below. Antenna brownish. Arista dorsal, 3 times as long as antenna, brown, bare. Basal article short. Palpus yellow with pale bristly hairs. Proboscis dark brown.

Thorax. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 4 dc, anterior 3 dc equally long, prescutellar twice as long. Scutellum with 2 marginals, without lateral hairs. Two very short lower white propleurals.

Legs. Yellow, tarsomeres 4 and 5 brown. Fore coxa yellowish white, mid and hind coxa entirely brown, extreme tips yellowish. Fore coxa anteriorly with short white bristles. Trochanter bare. Fore femur a little thickened in basal half. Anteroventrally with a row of whitish to pale brownish long hair-like bristles, up to three times as long as femur is wide; a little coiled at tip, and with a posteroventral row of white bristly hairs, also 3 times as long as femur wide. Fore tibia as long as femur, with 2 remarkable

posteroventral bristles in basal half. Mid coxa without exterior bristle. Mid femur thickened in basal <sup>2</sup>/<sub>3</sub>, a little thicker than fore femur; with a row of 4 brownish ventral bristles in basal third, half as long as femur as wide, anteriorly with row of 4 tiny preapicals; a stronger preapical pv. Mid tibia with a short ad and pd. Hind coxa without exterior bristle. Hind femur thickened in basal half, a little thicker than mid femur; double row of pale ventral bristle in apical half, as long as femur is wide, dorsally near base with a few erect bristle, anteriorly with 2 fine preapical bristles, posteriorly with 1 preapical bristle. Hind tibia with a row of ventral bristles, near middle as long as tibia is wide.

Wings. Without spots. Tp straight, apical part of M3+4 2 times as long as Tp. Anal vein not reaching wing margin.

Abdomen. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short and pale. Sternites with brownish, inconspicuous bristles.

Terminalia (Figs 22–24). Phallus long strap-shaped. Cerci yellowish, not fused and with long apical bristles (Figs 23–24).

## Female

Unknown

## Distribution

Southern Thailand (Gulf of Thailand).

# Remarks

*Thinophilus parvulus* sp. nov. is a very small species characterized by the yellowish white fore coxa and the 2 long posteroventral bristles near the base of the fore tibia.

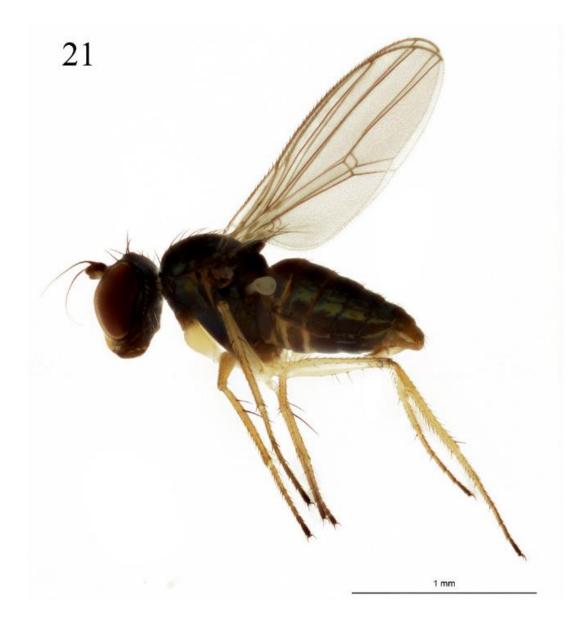


Figure 46. Male habitus, Thinophilus parvulus Samoh et al., 2017

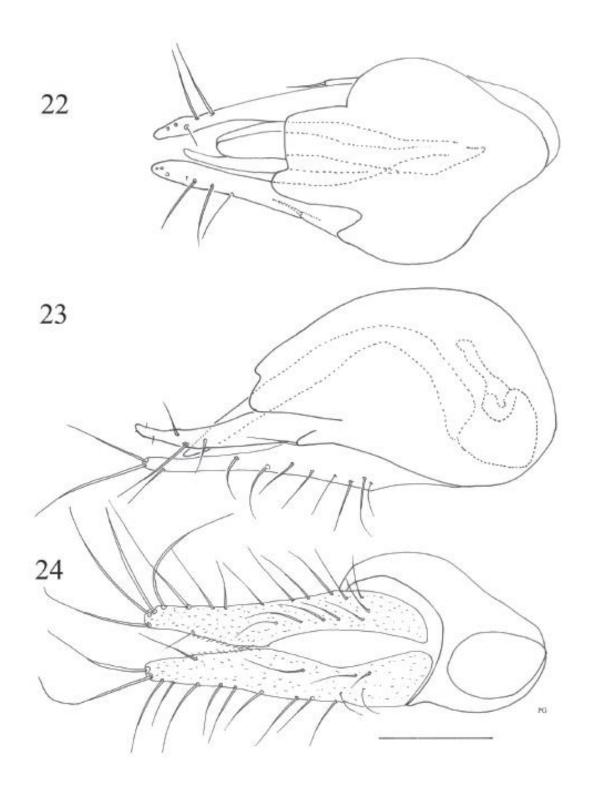


Figure 47. *Thinophilus parvulus* sp. nov., ♂, terminalia. 22. Genital capsule, ventral view. 23. Genital capsule, lateral view. 24. Genital capsule, dorsal view.

# Thinophilus spinatoides, new species

# Diagnosis

A medium-sized species with very long yellow legs. Both male and female with a set of 4–5 long, stiff brown ventral bristles on fore femur. Fore femur spindle-shaped, basal quarter much dilated. Fore tarsomere 1 very long and slender, twice as long as fore tibia. Tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 widened, black.

# Etymology

The specific epithet refers to the resembles with *T. spinatus* sp. nov., also described from southern Thailand.

## **Type material**

## Holotype

THAILAND:  $\bigcirc$ , Bakan Tohtid, Langu, Satun province, 6°47′29.8″ N, 99°48′53.5″ E, sweep netting, A. Samoh leg., 3 June 2015 (NHM-PSU).

## Paratypes

THAILAND: 2  $\bigcirc \bigcirc \bigcirc$ , 1  $\bigcirc$ , Bakan Tohtid, Langu, Satun province, 6°47′29.8″ N, 99°48′53.5″ E, sweep netting, A. Samoh leg., 3 June 2015 (NHM-PSU).

### Description

Male (Fig. 25)

Length. Body 4.5 mm; wing 3.8 mm.

Head. Frons and face with shiny dark metallic green ground colour. A pair of long divergent black ocellars. No postocellars. A pair of tiny proclinate verticals at level of front ocellars. Postcranium dark metallic green. Postverticals not differentiated from the upper post oculars. Upper postoculars uniseriate, short, black; with a few yellow lower postoculars. Antenna yellowish. Arista dorsal, 2.5–3 times as long as antenna, brown, not pubescent. Basal article short, brown; rest of arista paler. Palpus yellow, with few black bristly hairs. Proboscis dark brown.

Thorax. Thorax and scutellum shiny dark metallic green, with coppery and purple reflections. No dull black spots. Bristles on thorax black. Acr lacking; 7 rather short dc, gradually growing longer toward scutellum, ending in a very long prescutellar. Scutellum with 2 long marginal with a tiny hair at outside. 2 short black propleural bristles.

Legs. Yellow, only apical tarsomeres 2 of all legs black. Fore coxa with only basal quarter darkened; mid and hind coxae black, tip yellow. Coxa anteriorly with a short bristle near base and a long bristle at basal third. Trochanter with short white bristles.

Fore femur club shaped, very thickened in basal quarter; apical <sup>3</sup>/<sub>4</sub> very thin. Ventrally with 4 long black bristles; longest bristle nearly twice as long as femur as wide; others shorter. Fore tibia much longer than femur, without ventral bristling. Fore tarsomere 1 very long and slender, twice as long as fore tibia. Tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 widened, black. Mid coxa with a short black exterior bristle above middle; anterior bristles short, black. Mid femur ventrally without bristles; no preapical av. Mid tibia longer than mid femur, with a crown of short of apical bristles and 2 minute ad. Mid tarsomere 1 almost twice as long as following tarsomeres. Hind coxa without exterior bristle. Hind femur without ventral bristles; no preapical anterodorsal bristles. Hind tibia with 2 short ad and crown of apical bristles. Hind tarsomere 1 a little longer than tarsomere 2.

Wings. Uniformly yellowish tinged, without spots. Tp straight, apical part of M3+4 1.5 times as long as Tp. Anal vein not reaching wing margin. Anal vein not reaching wing margin.

Abdomen. Shining dark metallic green. Hairs and hind-marginal bristles on tergites short, black. Sternites without bristles; except sternite 4 with tuft of short black bristles.

Terminalia (Figs 27–29). Phallus long strap-shaped. Cerci pale brownish with pale hairs, dorsally fused (Fig. 29).

Female (Fig. 26)

Length. Body 4.5 mm, wing 4.2 mm. Similar to male except following characters. Fore femur basally not so strongly swollen like in male and with 5 strong black ventral bristles up to 3 times as long as femur as wide. Tarsomere 1 of fore and mid legs more than twice as long as following tarsomeres together. Sternites 3, 4, and 5 with pale bristles.

**Distribution** Southern Thailand (Andaman Sea).

# Remarks

*Thinophilus spinatoides* sp. nov. is particular in that it has the fore femur with basal quarter much spindle-shaped dilated. It is less dilated in *T. spinatus* sp. nov. Fore tibia much longer than fore femur; shorter in *T. spinatus* sp. nov. Fore tibia slender and without ad in male, present in female; fore tibia stouter and with 2 long ad in *T. spinatus*. Fore tarsomere 3 contrastingly yellowish white, tarsomeres 4 and 5 much widened black. Fore tarsomere 3 has the same pale yellowish colour as tarsomeres 1 and 2. Tarsomeres 4 and 5 black, not widened. Only base of fore coxa brown; basal 2/3 of fore coxa brown in *T. spinatus* sp. nov. Lower postocular bristles yellow; black in *T. spinatus* sp. nov. Anal vein distinct in basal 2/3; anal vein not distinct at all in *T. spinatus* sp. nov.

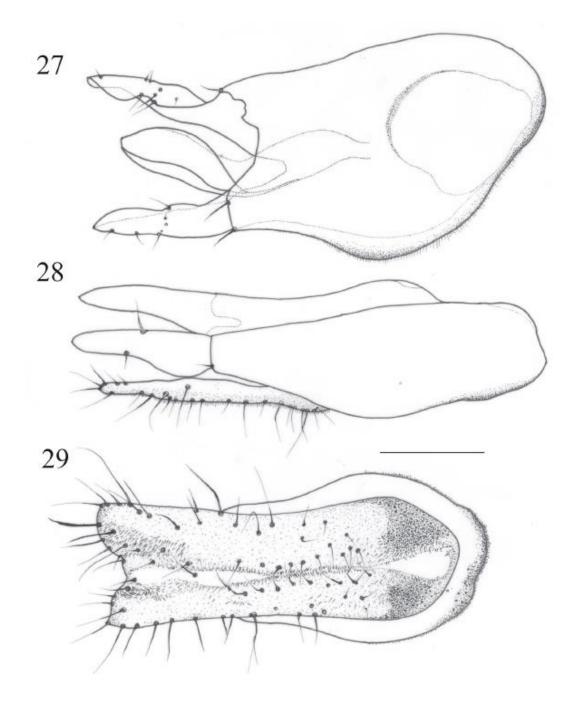


Figure 48. *Thinophilus spinatoides* sp. nov., ♂, terminalia. 27. Genital capsule, ventral view. 28. Genital capsule, lateral view. 29. Genital capsule, dorsal view.

### Thinophilus spinatus sp. nov.

# Materials Examined.

## Holotype

THAILAND: Male, Bang Phat, Muang, Phang Nga Province (8°21'48.8"N, 98°34'38.8"E), Samoh, A., Malaise trap, 13 February 2015.

# Paratypes

THAILAND: 1 male, 1 female, Bang Phat, Muang, Phang Nga Province (8°21'48.8"N, 98°34'38.8"E), Malaise trap, 13 February 2015.

Remarks.- There are occurred in southern Thailand and Singapore (Unpublished record of a female). The femora are spindle-shaped and the fore femur in the male as well as in the female bear long, brown stiff bristles as in *T. spinatoides* sp. nov. The main difference is that the fore femur in the *T. spinatoides* sp. nov. males are much more inflated than in *T. spinatus* sp. nov. For further differences see under remarks in *T. spinatoides* sp. nov.

# Thinophilus variabilis sp. nov.

## Materials Examined.

Holotype

THAILAND: Male, Laem Pho, Hat Yai, Songkhla province (7°09'15.9"N, 100°28'03.6"E) Samoh, A., Sweep netting, 27 June 2015.

# Paratypes

THAILAND: 6 males, 10 females, Ban Nua Nam, Phumriang, Chaiya, Surat Thani (9°23'34.0"N, 99°15'24.0"E), Samoh, A., Sweep netting, 18 April 2015. 2 males, 4 females, Ban Nua Nam, Phumriang, Chaiya, Surat Thani (9°23'34.0"N, 99°15'24.0"E), Samoh, A., Sweep netting, 20 April 2015. 5 males, 18 females, Ban Dato, Yaring, Pattani (6°55'17.1"N, 101°19'50.7"E) Samoh, A., Sweep netting, 12 April 2015.

*Remarks.* -*Thinophilus variabilis* sp. nov., is a small species, that differs from *Thinophilus minutus* sp. nov. in having distinct ventral bristles on all femora. Coxae and femora can vary in colour from yellow to brown and even dark brown. Such a variation in colour is fairly unusual in *Thinophilus* and might be due to preservation of the specimens in the denaturised ethanol. The species seems to be widespread in peninsular Thailand (Andaman Sea and Gulf of Thailand).

## Thinophilus apicatus

**Material Examined.** 3 males, 7 females, Khlong Yang, Koh Lanta, Krabi Province (7°47'41.3"N 99°05'23.2"E), Samoh, A., sweep netting, 13 June 2015; 2 males, Khlong Phon, Khlong Thom, Krabi Province (7°48'11.2"N 99°10'11.9"E, Samoh, A., Malaise trap, 26 February 2015; 19 males, 13 females, Ban Elet, Pak Nam, Muang, Chumphon Province (N 10°30'28.7', E 099°14'29.8'), coll. Samoh, A., sweep netting, 18 February 2015.

**Remarks.** This species have been collected from Singapore mangroves and had also been assumed to be a new species. *T. apicatus*, composes a typical morphological features. For instance, Fore coxa anteriorly with two very long bristles. Legs yellow, but all tibiae narrowly, and conspicuously darkened at tips (less on hind tibia).

**Distribution.** Krabi, Phang Nga (Andaman Sea), Chumphon, Nakhon Si Thammarat, Surat Thani (Gulf of Thailand).

## Thinophilus chaetulosus

**Material Examined.** 2 males, 3 females, Ban Phanang Tak, Muang, Chumphon Province (N 10°30'23.9', E 99°13'55.6'), coll. Samoh, A., sweep netting, 17 February 2015; 1 male, Ban Elet, Pak Nam, Muang, Chumphon Province (N 10°30'28.7', E 099°14'29.8'), coll. Samoh, A., sweep netting, 18 February 2015; 7 males, 11 female, Ban Bang Kong Khong, Pak Phanang, Nakhon Si Thammarat (8°24'09.4"N 100°11'29.9"E) coll. Samoh, A., sweep netting, 1 May 2015; 7 males, 11 females, Prince of Songkla University, Pattani Campus, Muang, Pattani (6°53'04.9"N 101°14'10.1"E) coll. Samoh, A., sweep netting, 4 November 2014.

**Remarks.** It contains a unique morphological characters in having fore leg with apical tarsomere black. Ventral bristles of fore tibia over entire length, as long as tibia is wide. Besides, they are larger species and most resemblance to *T. pallitarsis*.

**Distribution.** Chumphon, Nakhon Si Thammarat, Pattani, Surat Thani, Songkhla (Gulf of Thailand)

### Thinophilus melanomerus

**Material Examined.** 5 males, 2 females, Tarutao Island, Langu district, Satun Province (6°44'19.2"N 99°38'45.4"E) coll. Samoh, A., sweep netting, 9 January 2015; 1 male, 1 females, Ban Bo Sane, Thap Pud, Phang Nga Province (8°27'29.7"N 98°36'17.8"E), coll. Samoh, A., sweep netting, 13 February 2015.

**Remarks.** *T. melanomerus* is easily recognised by all femora black on basal two thirds, but with contrastingly yellow tip. Hind trochanter brown; rest of legs yellow. Fore trochanter with up to six long black ventral bristles with a curve tip.

Distribution. Satun and Phang Nga (Andaman Sea)

### Thinophilus parmatus

**Material Examined.** 1 male, 1 female; Bang Yai, Bang Nai Si, Takuapa, Phang Nga Province (8°54'27.5"N 98°23'59.6"E), coll. Samoh, A., sweep netting, 9 February 2015; 1

male, Ban Khlong Yang, Koh Lanta, Krabi Province (7°47'41.3"N 99°05'23.2"E), coll. Samoh, A., sweep netting, 13 June 2015.

**Remarks.** Rather small species with yellow palpi, yellow antennae and all coxae dark. Male: second segment of mid tarsus dorsally enlarged into a dark brown lobe; third segment less widened, contrasting pale. 6-7 dc growing longer backwards, all relatively short. Cerci yellow; aedeagus without extension.

Distribution. Phang Nga and Krabi (Andaman Sea)

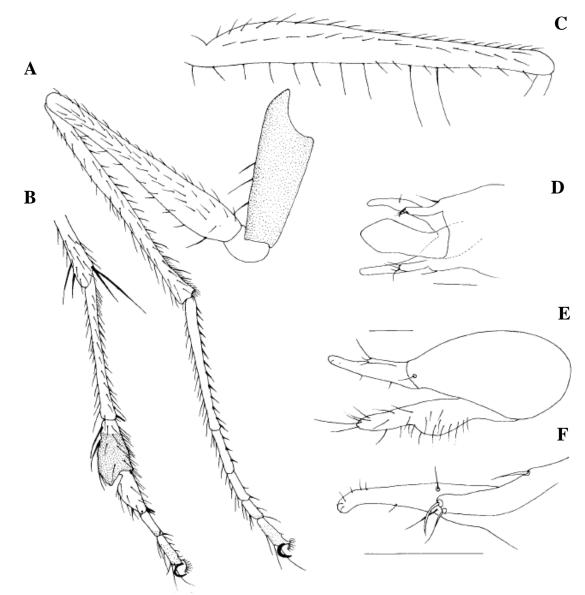


Figure 49. Male habitus, *Thinophilus parmatus* Grootaert and Meuffels, fore leg posteriorly (A); mid tarsus (B); hind femur (C); Aedeagus and surstyli ventrally (D); hypopygium leterally (E); surstyli (F); scale 0.1 mm (modified from Grootaert and Meuffels, 2001).

#### Thinophilus simplex

**Material Examined.** 732; Thailand, Ban Elet, Muang, Chumphon (N 10°30'28.7', E 99°14'29.8'), sweep netting, 16 February 2015, coll. A. Samoh.

**Remarks.** This species was firstly collected from Singapore mangroves by Grootaert and await for description (unpublished data). The distinctive morphological features is evident. *Thinophilus simplex*, in having femora without distinct bristling. Small species with small yellow strap-shaped cerci, at most 1/5 length of venter.

Distribution. Satun, Phannga, Krabi, Chumphon (Gulf of Thailand).

### Thinophilus superbus

**Material Examined.**  $9 \stackrel{\circ}{_{\sim}} 3 \stackrel{\circ}{_{\sim}}$ ; Thailand, Ban Tutarum, Langu, Satun (6°55'10.1"N 99°43'59.0"E), sweep netting, 4 May 2015, coll. A. Samoh.  $1 \stackrel{\circ}{_{\sim}} 5 \stackrel{\circ}{_{\sim}}$ ; Na Thab, Chana, Songkhla (7°01'25.8"N 100°43'05.0"E), sweep netting, coll. A. Samoh.

**Remarks.** This species was firstly collected from Pulau Semakau in April, 3, 2012 by Grootaert (unpublished data). Hind tibia with a very long thin dorsal preapical that is nearly as long as tibia is long. More secondary sexual characters on all legs.

**Distribution.** Tanyong Po, Muang, Satun; Ban Tutarum, Langu, Satun, Krabi, Phang Nga, and Songkhla (Chana, Gulf of Thailand).

#### Thinophilus yeoi

**Material Examined.**  $7\overset{\circ}{\circ}6^{\circ}$ ; Thailand, Phumriang, Chaiya, Surat Thani (9°19'43.4"N 99°12'31.6"E), sweep netting, 22 April 2015, coll. A. Samoh.

**Remarks.** *Thinophilus yeoi*, contains yellow femora. Fore femur with a few bristles near base, half as long as femur is deep and composes a row of long posteroventral bristle near tip. Large species with cerci large, blackish brown more than half the length of abdomen. Besides, this species quit obviously found Surat Thani bay, Gulf of Thailand.

Distribution. Chumphon and Surat Thani



Figure 50. Male habitus, Thinophilus yeoi (pending for description by Grootaert).

#### SUBFAMILY NEURIGONINAE

#### Neurigona Hollis, 1964

**Remark and Recognition.** The *Neurigoninae* represent with 225 species and 16 genera from all zoogeographical region (accounts for 3% of the known Dolichopodidae) (Yang et al., 2006). Unfortunately, in Thailand, this genus is very few recorded. In the context of species recognition, the delimitation of the *Neurigona* can be easily recognized by these combination characters: face with dense pruinosity, proepisternum with setae; posterior slope of mesonotum flattened; legs elongate and bare of major setae; male abdominal segments 4 and/or 5 sometimes with ventral modifications; segment 7 bare, forming a peduncle; and hypopygium usually globular (Bickel, 1998; 2009).

### Neurigona pectinata Becker, 1922

**Materials Examined.**  $\bigcirc$ ; Thailand, Na Thab, Chana, Songkhla (7°01'25.8"N 100°43'05.0"E), sweep netting, 27 September 2015, coll. A. Samoh.

**Remarks.** This species was recorded from an altitude of 500 m in the northeast Thailand (Loei province). And this studied, we collected from mid and back mangroves which were mostly affected by fishermen activities (shrimp and fish farms, and villager's residences). It is a medium size of dolichopodid fly that is always found along vertical of mangrove tree trunks. The body length was 3.2-3.5 mm, wing length 3.2-3.4 mm. With anteroventral comb of fine erect hairs on mid tibia and tarsus were distinct morphological features to delimit species. From our surveyed, a single species was identified from mid and back mangrove in Na Thap subdistrict, Chana district, Songkhla province (Gulf of Thailand). However, we expected that this species could be dispersed and colonized to the Andaman sea too.

**Distribution.** Songkhla (Gulf of Thailand)

#### SUBFAMILY PARATHALASSIINAE

#### Microphorella Becker, 1909

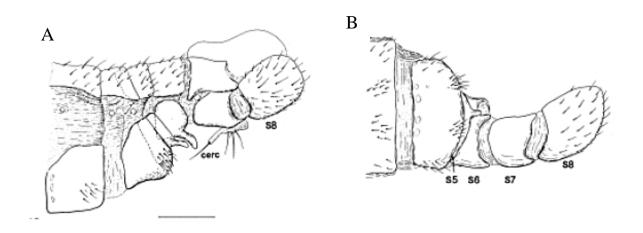
#### Microphorella malaysiana Shamshev & Grootaert, 2004

(Figure 52)

**Materials Examined.**  $6 \stackrel{\circ}{\circ} 2 \stackrel{\circ}{\ominus}$ ; Thailand, Laem Kho Kwang, Muang, Chumphon (N 10°30'48.7', E 99°15'52.0'), sweep netting, 17 February 2015, coll. A. Samoh.  $23 \stackrel{\circ}{\circ} 15 \stackrel{\circ}{\ominus}$ ; Phumriang, Chaiya, Surat Thani (9°19'43.4"N 99°12'31.6"E), sweep netting, 22 April 2015, coll. A. Samoh.

**Remarks.** A small species (1.5-1.8 mm long) with pale yellow palpi in male, brown in female; 6 dorsocentral bristles; fore tarsi thickened, wholly brown; abdominal sternites 5 and 6 of male with short median posteromarginal processes of subequal size (Figure 51); right surstylus leaf-like.

**Distribution.** There are rarely distributed in Thailand (Phumriang, Chai Ya, Surat Thani, Chumphon, and also from Satun), but greatly occurred in Malaysia, Singapore, and Indonesia.



Figures 51. *Microphorella malaysiana*, new species, male. A – postabdomen, left lateral view, B – same, ventral view. Scale = 0.1 mm. (Shamshev and Grootaert, 2004)



Figure 52. Male habitus, Microphorella malaysiana Shamshev & Grootaert, 2004

#### SUBFAMILY RHAPHIINAE

### Ngirhaphium Evenhuis & Grootaert, 2002

*Ngirhaphium* Evenhuis & Grootaert, 2002: 310. Type species by original designation: *Ngirhaphium murphyi* Evenhuis and Grootaert, 2002.

**Remarks and Diagnosis.** Medium to large sized species (4.5–8 mm) with a metallic green or blue ground colour. Antenna very long in males, a little shorter in females. Arista apical, basal article long. Rostrum in male small with well-developed labellae. Large rostrum in female Vertex excavated (cf. Sciapodinae). Mid and hind coxae without exterior bristle. Femora with inconspicuous bristling. All tibiae with strong bristles. Fore leg in male with tarsomere 4 bearing an asymmetrical, apical dorsal forked protuberance (absent in females); terminal segment with a pair of normal claws and a thickened claw-like structure beneath the posterior claw. Females with the claws as usual, but the terminal segment bears a long dorsal protuberance. Mid and hind legs with tarsomeres 1–4 with an apical comb of spinules ventrally. Wing with tip of  $M_{1+2}$  sharply bent upwards just before reaching the wing border and ending near tip of  $R_{4+5}$ .

#### Key to species of male Ngirhaphium Evenhuis and Grootaert, 2002

- **3**) Dorsal surstylus elongate, digitiform with truncate apex (Figs 9, 12) slightly shorter than cercus. Cerci with narrow apex, set with 2 apical setae (Fig. 12).....*N. murphyi* Evenhuis & Grootaert
- Dorsal surstylus with very wide apex (Fig. 7) much shorter than cercus. Cerci with expanded apex set with a bunch of yellow setae (Fig. 10).....*N. sivasothii* Grootaert & Puniamoorthy

#### Ngirhaphium caeruleum Grootaert & Puniamoorthy, 2014

### (New record)

**Materials Examined.** THAILAND:  $2 \Diamond, 4 \heartsuit$ , Chumphon province, Ban Elet Muang (N 10°30'28.7', E 99°14'29.8'), 16.ii.2015 (leg. A. Samoh) (PSU)

Remarks. A larger species with mesonotum and tergites shining metallic blue. Apical aristal segment thin (filiform) and longer than basal aristal segment. Male with dorsal and ventral surstylus at right side fused; separated at left side. Cerci long, but shorter than surstyli so that they are concealed between the surstyli. In addition, the name caeruleum (adjective) means blue in Latin and it refers to the dark blue metallic ground-colour of thorax and abdomen.

Distribution. Chumphon, Surat Thani.

## Ngirhaphium chutamasae Samoh et al., 2015

## (Figure 53)

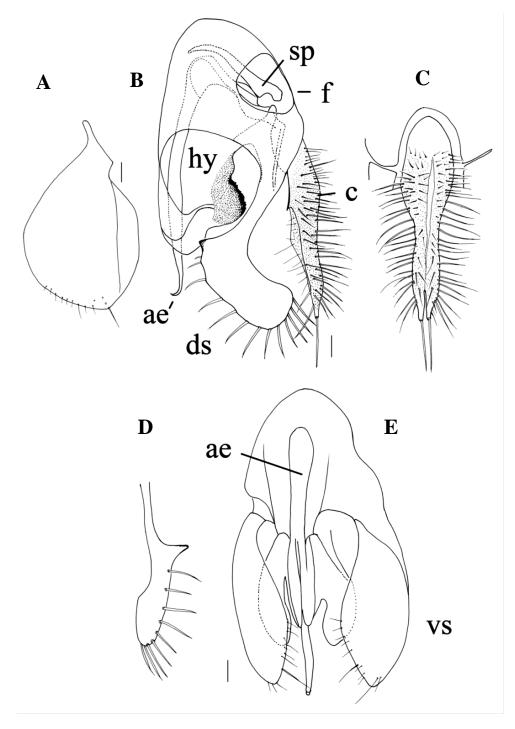
**Materials Examined.** HOLOTYPE  $\Diamond$ , labelled: "THAILAND: Satun prov., Tammalang (6°32'21.05"N, 100°04'9.42"E), 3.x.2014 (reg. 34030, leg. P. Grootaert)" (PSU); PARATYPE: 1  $\Diamond$ , Tammalang (6°32'21.05"N, 100°04'9.42"E), 6.viii.2014 (leg. A. Samoh) (RBINS).

**Remarks.** A large species differing from the other *Ngirhaphium* species mainly in the structure of the male genitalia. Cercus in lateral view slightly shorter than dorsal surstylus. Cercus brown, tip pointed bearing a single yellow bristle. Dorsal surstylus brown, bordered with short, stout yellow bristles. Outer branch of apical fork on the fore tarsomere 4 slightly longer than inner branch.  $M_{1+2}$  with a short stub on apical bend. Moreover, the species is dedicated to Associate Professor Dr. Chutamas Satasook, director of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkla University, Hat Yai as a token for her dynamic support of our research.

Distribution. Tammalang, Muang, Satun (Andaman Sea)



Figure 53. *Ngirhaphium chutamasae* sp. nov., male habitus; inset: apical tarsomeres on fore leg, showing the large fork-like extensions on tarsomere 4 and the additional claw-like structure on tarsomere 5 (photo: J. Brecko). Scale = 1 mm. (Samoh et al., 2015)



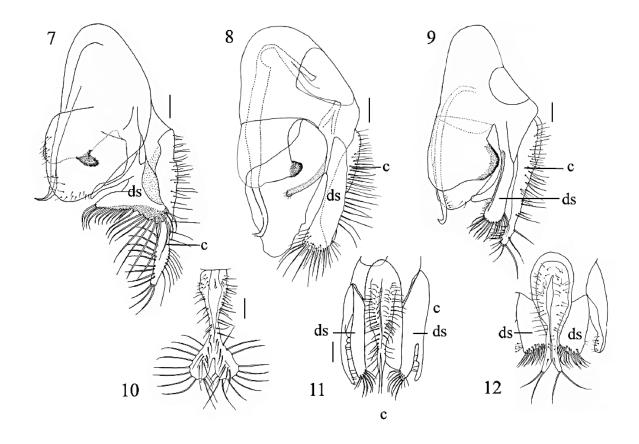
Figures 54. *Ngirhaphium chutamasae* sp. nov., holotype male genitalia: A. Left ventral surstylus; B. Lateral view of genital capsule with left ventral surstylus removed; C. Cerci dorsally; D. Dorsal surstylus in ventral view; E. Ventral view of genital capsule. Abbreviations: ae: aedeagus; c: cercus; ds: dorsal surstylus; f: foramen; hy: hypandrium lobe; sp: sperm pump; vs: ventral surstylus. Scale = 0.1 mm (Samoh et al., 2015)

### Ngirhaphium murphyi Evenhuis & Grootaert, 2002

*Ngirhaphium murphyi* Evenhuis & Grootaert, 2002: 310. Type locality: SINGAPORE: Kranji mangrove. *N. murphyi*: Grootaert & Puniamoorthy, 2014: 147 (figs 1–3, 4, 5, 18).

**Materials Examined.** - THAILAND: 1  $\Diamond$ , 4  $\bigcirc$ , Satun province, Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 11.viii.2014 (leg. A. Samoh) (PSU)

**Remarks.** - A large species (5.7–7.3 mm), generally with clear wings. Mesonotum and tergites metallic green. Apical aristal article quite thick, nearly half as long as basal aristal article. Male with cerci longer than surstyli and thus the tips are visible outside the surstyli (Figs 55C, 55F).



Figures 55. Lateral view genital capsule: A. *Ngirhaphium sivasothii*; B. N. caeruleum; C. *N. murphyi*; dorsal view cerci: D. *N. sivasothii*; E. *N. caeruleum*; F. *N. murphyi*. Abbreviations: c: cercus, ds: dorsal surstylus (modified after Grootaert and Puniamoorthy, 2014). Scale = 0.1 mm.

### Ngirhaphium sivasothii Grootaert & Puniamoorthy, 2014

*Ngirhaphium sivasothii* Grootaert & Puniamoorthy, 2014: 150 (figs 55A, 55D, 56). Type locality: SINGAPORE: Semakau Island.

*Materials examined.* - THAILAND: Satun province:  $1 \ 3, 2 \ 9$ , Tammalang (6°32'21.05" N, 100°04'9.42" E);  $4 \ 3 \ 7 \ 9$ , 6.viii.2014 (leg. A. Samoh);  $1 \ 3, 2 \ 9$ , 3.x.2014 (reg. 34030, leg. P. Grootaert & A. Samoh);  $7 \ 3, 20 \ 9$ , Tarutao Island, Talo Wao bay (6°36'58.7"N 99°40'43.1"E), 12.viii.2014 (leg. A. Samoh);  $2 \ 3, 1 \ 9$ , Tanjong Po (6°36'57.43" N, 99°57'25.66" E), 3.x.2014 (leg. A. Samoh) (PSU).

*Remarks.* - A medium-sized species (4.5–5.5 mm), generally with dark infuscate wing and with longitudinal veins and Tp (posterior cross vein) brownish seamed. Mesonotum and tergites metallic green. Apical aristal article shorter, but nearly as long as basal article. Male with dorsal surstylus half as long as cerci, with a rectangular bend, set with very long bristles (Fig. 55A). Cercus much longer than dorsal surstylus, tip wide, rounded, set with many long yellow bristles (Fig.55D). Outer branch of apical fork on the fore tarsomere 4 slightly shorter than inner branch.

Distribution. Satun, Phannga, Karbi.

# Ngirhaphium meieri sp. nov.

(New species, pending for description)

**Materials Examined.** - HOLOTYPE ♂, labelled: THAILAND: Phang Nga province, Takuapa, Bang Yai, (8°54'27.5"N, 98°23'59.6"E), sweep netting, 9 February 2015.

**Remarks.** - A large species differing from the other *Ngirhaphium* species mainly in the structure of the male genitalia. Cercus in lateral view slightly shorter than dorsal surstylus. Cercus brown, tip pointed bearing a single yellow bristle. Dorsal surstylus brown, bordered with short, stout yellow bristles. Outer branch of apical fork on the fore tarsomere 4 slightly longer than inner branch.  $M_{1+2}$  with a short stub on apical bend.

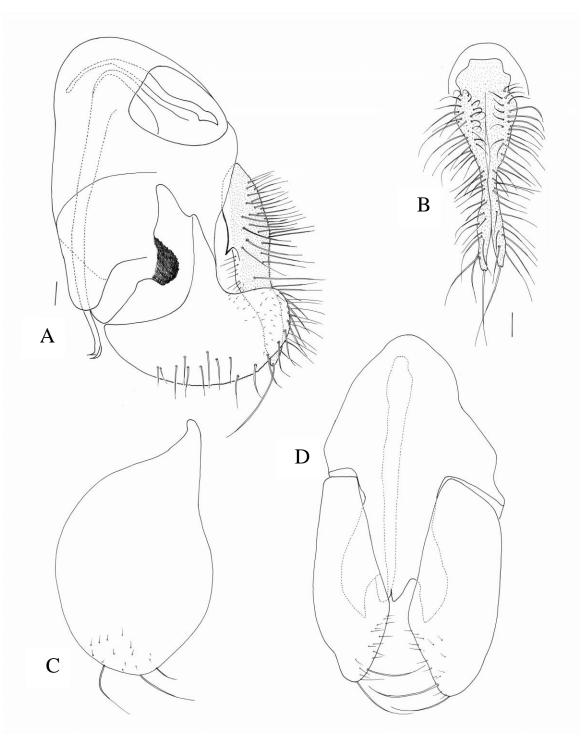
Distribution. Takuapa, Phang Nga.



Figure 56. Male habitus, Ngirhaphium sivasothii Grootaert & Puniamoorthy.



Figure 57. *Ngirhaphium meieri* sp. nov., male habitus; inset: apical tarsomeres on fore leg, showing the large fork-like extensions on tarsomere 4 and the additional claw-like structure on tarsomere 5. Scale = 1 mm.



Figures 58. *Ngirhaphium meieri* sp. nov., holotype male genitalia: A. Lateral view of genital capsule with left ventral surstylus removed; B. Cerci dorsally; C. Left ventral surstylus; D. Ventral view of genital capsule. Abbreviations: ae: aedeagus; c: cercus; ds: dorsal surstylus; f: foramen; hy: hypandrium; sp: sperm pump; vs: ventral surstylus. Scale = 0.1 mm.

### SUBFAMILY SCIAPODINAE

**Remarks.** It is known as one of the most primitive subfamilies of long-legged flies (Negrobov, 1986) based on the following combination characters: well develop or present of wing vein  $M_2$ , crossvein bm-cu incomplete (figure.59 A), male abdominal segment 7<sup>th</sup> external and setose, and hypopygium with a unique "dorsal process" (Bickel, 1994) (Figure 59 B). In Thailand, there are composed with three knowns (published species) and three unknown species. The following are those species that have been studied and registered in the Princess Maha Chakri Sirindhon Natural History Museum (MNHM) of the Prince of Songkla University.

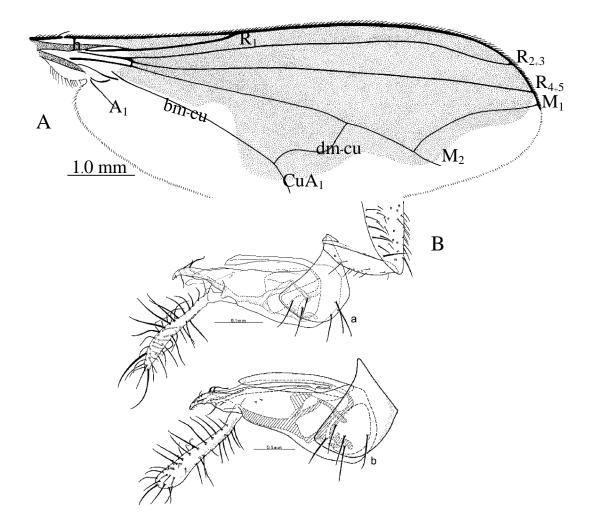


Figure 59. Male habitus, *Krakatauia luctosa*'s wing venation and hypopygium, left lateral: a, *Amblypsilopus sounwari*. b, *A. navatadoi* (modified from Bickel, 2008; 2009)

#### Amblypsilopus Bigot, 1889

Amblypsilopus Bigot, 1889: 24.

**Type species**. *Psilopus psittacius* Loew, 1861 (as *psitacinus* Fabricus), by original designation.

**Remark and Recognition.** The genus *Amblypsilopus*, previously, most members belong this genus were described in *Sciapus*, as currently classified, is a polyphyletic fly assemblage (previously described in *Sciapus*) which included distinctive external morphological features, with dorsal arista and pale hair on the lower calypter. Lots of genera are regarded as junior synonyms of this genus based on modified male wings (MSSCs, Male Secondary Sexual Characters) and represent small derived group, namely, *Australiola, Labeneura, Leptorhetum*, and *Sciopolina* (Bickel, 1994). Despite, recently study, one species was collected from marine habitats in Thailand, *Amblypsilopus abruptum*.

#### Amblypsilopus abrubtum Walker, 1859

**Materials Examined.**  $43^{\circ}3^{\circ}$ ; Khao Than, Tha Chang, Surat Thani, (9°23'34.0"N 99°15'24.0"E), sweep netting, 21 April 2015, coll. A. Samoh.

**Remarks.** – This species was firstly recognised by Dyte (1975) as a species which is distributed in Thailand.

Distribution. Chumphon, Surat Thani, Nakhon Si Thammarat, Songkhla

#### Chrysosoma Guerin-Meneville, 1831

**Remarks.** – This genus is recorded as one of the most diverse long-legged flies from the Oreintal realm and many genera seemed to be restricted to Indo-Pacific region (Bickel, 1994). With more than 158 species were recorded from Old world (Dyte, 1975).

#### Chrysosoma luecopogon

**Materials Examined.** 232; Thailand, Ban Elet, Paknam, Muang, Chumphon (N 10°30'28.7', E 99°14'29.8'), sweep netting, 16 February 2015, coll. A. Samoh.

**Remarks.** This species is containing distinctive morphological features and fairly constant in morphology. Some intraspecific variation is evident in body length (Bickel, 1994), cercus mostly consisting of a bunch of pubescence hairs (Figure 60), Fore leg chaetotaxy is somewhat variable. In addition, *Chrysosoma* generally observed from vegetation zone in mangrove forests than others.

**Distribution.** This species is a widespread distributed species in paleotropical realms includes the eastern Africa coast, India, Sri Lanka, southeast Asia, Madacascar, Taiwan, Papua New Guinea, New Calidonia, and Australia (Bickel, 1994). In Thailand, we mainly collected from Chumphon province.

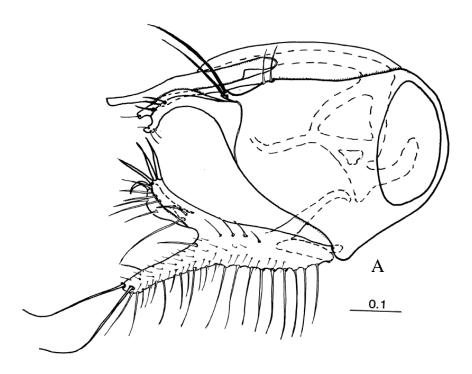


Figure 60. *Chrysosoma leucopogon*,: A – male hypopygium, left lateral (modified from Bickel, 1994).

### SUBFAMILY SYMPYCNINAE

#### Chaetogonopteron de' Meijere, 1913

#### Chaetogonopteron chaeturum

**Remarks.** Recorded in Grootaert and Meuffels (1999), this species was firstly described from this country. Recently, extended recording from Singapore mangroves and Malaysia peninsula (Evenhuis and Grootaert, 2002).

Distribution. Satun, Phang Nga, Chumphon (Gulf of Thailand).

### Chaetogonopteron vexillum

**Remarks.** The *C. vexillum* was originally described from Australia by Bickel (2013). This study was the first record for Thailand.

Distribution. Nakhon Si Thammarat, Surat Thani, Satun (Andaman Sea).

### Sympycnus Loew 1857

**Remarks.** The genus *Sympycnus* is a worldwide distributed long-legged fly, with 273 described species (Yang et al., 2006) from all zoogeographical regions. It is easily separated from other Sympycninae by the following combination characters: small to medium in body sizes; antenna scape bare; first flagellomere almost triangular in shape; mesonotum without black or brown lateral spot, metepimeron without hair; Male surstylus usually projected, generally dorsal surstylus and ventral surstylus fused with each other, basally also fused with epandrium (Yang et al., 2011).

### Sympycnus sp. A

**Remarks.** This species has not previously been recorded from this country. This recorded species was recently collected from mangroves used Malaise traps and probably represent new species.

Distribution. Chumphon, Surat Thani.

#### Teuchophorus Loew, 1857

**Remarks.** – The *Teuchophorus* is known as a small fly (body length 1.5-4 mm). It is closer to the Sympycnus-Chaetogonopteron complex. There are composed with many combinations of taxonomic characters to assess species. Firstly, chaetotaxy on the mesonotum is quite stable (with uniseriate of acrostical bristles, rarely absent). Moreover, the first tarsomere of the hind leg is shortened. Wing vein  $M_{1+2}$  is turned up immediately after the connection with the cross vein tp and lack of wing boss like in Sympicnus-Chatogonopteron complex (Grootaert 2006). In Thailand, several species were mainly described from several provinces in southern Thailand by Meuffels and Grootaert (2003), for example Teuchophorus krabienesis (firstly collected from Su San Hoi, Krabi province) (Meuffels and Grootaert, 2004), T. ornatulus, T. stenostigma (Trang province), T. singaporensis (Phangnga province), T. pauper (there are widely distributed in several provinces in peninsular Thailand, namely; Sa Nangmanora and Thap Put, Phangnga province, Ban Duson and 21 km north of Langu, Satun province, Ban Khlong Kua, Songkhla province). From our surveyed, muddy, low sun exposed or shady area in mangroves, sandy patches, small creek in the forests were seemed to be a preferable homeland for these flies.

### Teuchophorus krabiensis Meuffels and Grootaert, 2003

**Materials Examined.**  $3\$   $3\$   $5\$  ; Thailand, Takuapa, Phang Nga (6°47'29.8"N 99°48'53.5"E), sweep netting, 9 February 2015, coll. A. Samoh.  $13\$   $34\$ ; Khao than, Surat Thani, (9°23'34.0"N 99°15'24.0"E), sweep netting, 21 April 2015, coll. A. Samoh.  $3\$   $3\$ ; Phumriang, Chaiya, Surat Thani (9°19'43.4"N 99°12'31.6"E), sweep netting, 22 April 2015, coll. A. Samoh.

**Remarks.** This species is identical to *T. simplicissimus* (Figure 61). Small species (1.65-1.95 mm), without stigma. Eyes rather widely separated. Antenna with basal segments brown; third segment yellowish brown, about 1.25 times as long as deep, with a rather acute apex. Legs and coxae yellow. (Male) Mid femur with an irregular row of partly hair-like av, longest on basal half. Hind femur anteroventrally near apex with a few longer hairs. Mid tibia: two ad, one pd, without ventrals in male (in female one ventral). Hind tibia with five dorsals (Figure 61). Hypopygium small (Meuffels and Grootaert, 2004).

Distribution. Chumphon, Surat Thani, Pattani, Phang Nga (Andaman Sea).

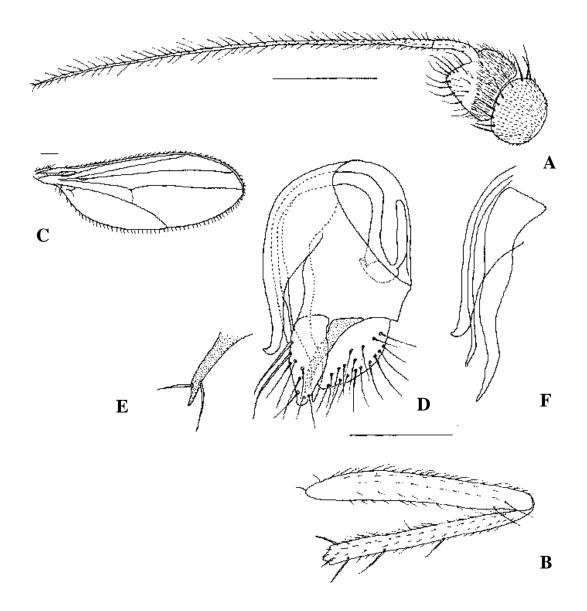


Figure 61. *Teuchophorus simplicissimus*, male habitus. (A) Antenna. (B) Hind leg: femur and tibia. (C) Wing. (D) Hypopygium (side view). (E) Tip of dorsalsurstylus. (F) Hypandrium, aedeagus and ventral appendage. Scale: 0.1 mm (Meuffels and Grootaert, 2004).

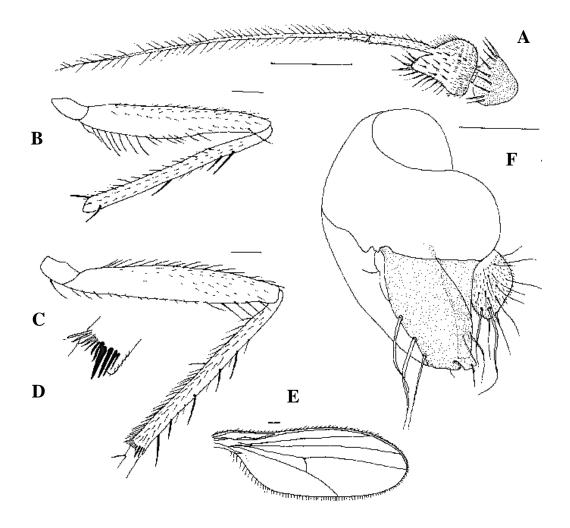


Figure 62. *Teuchophorus krabiensis*, male. Antenna (A). Mid leg: femurand tibia (B). Hind leg: femur and tibia (C). Detail of tip of hind tibia (D). Wing (E). Hypopygium (side view) (F). Scale: 0.1 mm (Meuffels and Grootaert, 2004).

## UNPLACED SUBFAMILY (INCERTAE SEDIS)

### Phacaspis Meuffels & Grootaert, 1990

**Remarks.** There are very minute metallic green species (the body size 1.1 - 1.5 mm). Meuffels and Grootaert (1988) mentioned that it was closely related to the genus *Kowmungia* Bickel, 1987 (there were found in forest along Australian coasts) which contains a stalk hypopygium. The best place to observe *Phacaspis* is waterfront and muddy mangrove near the sea and sometimes they are quickly fly in the border of large running creeks through a mangrove. Two species were recorded from Thaiand by Grootaert and Meuffels (2001): *Phacaspis mitis* Grootaert & Meuffels and *Phacaspis petiolata* Grootaert & Meuffels. From our studied, we found a species from two sides of peninsular Thailand.

### Key to the males of *Phacaspis* in Thailand

- 3 dc; third antennal segment triangular, with acute tip; fore femur with at least 4 pv bristles longer than femur is wide (Thailand)..... *P. mitis* Grootaert & Meuffels
- 4 dc; third antennal segment trapezoidal with rounded tip; fore femur with a row of pv which are shorter than femur is wide..... *P. petiolata* Grootaert & Meuffels

# *Phacaspis mitis* Grootaert and Meuffels, 2001 (Figure 63, 64)

**Materials examined.**  $7\35\$ ; Thailand, Phumriang, Chaiya, Surat Thani (6°47'29.8"N 99°48'53.5"E), sweep netting, 22 April 2015, coll. A. Samoh.  $7\38\$ ; Lidi island, Langu, Satun (6°50'30.4"N 99°46'32.9"E), sweep netting, 30 July 2015, coll. A. Samoh.  $6\32\$ ; Khong phon, Khlong Thom, Krabi (7°48'11.2"N 99°10'11.9"E), sweep netting, 30 July 2015, coll. A. Samoh.

**Remarks.** A tiny *Phacaspis mitis*, resembling to *P. petiolata*, in having these combination characters; fore femur ventrally with a row of very long, thin, straight setae, but lack of at mid and hind femora, three pairs of equally long dorsocentrally bristles at thorax, hypopygium very long (reaching beyond base of third abdominal segment).

**Distribution.** Chumphon, Surat Thani, (Songkhla Gulf of Thailand); Satun, Krabi, and Phang Nga (Andaman)

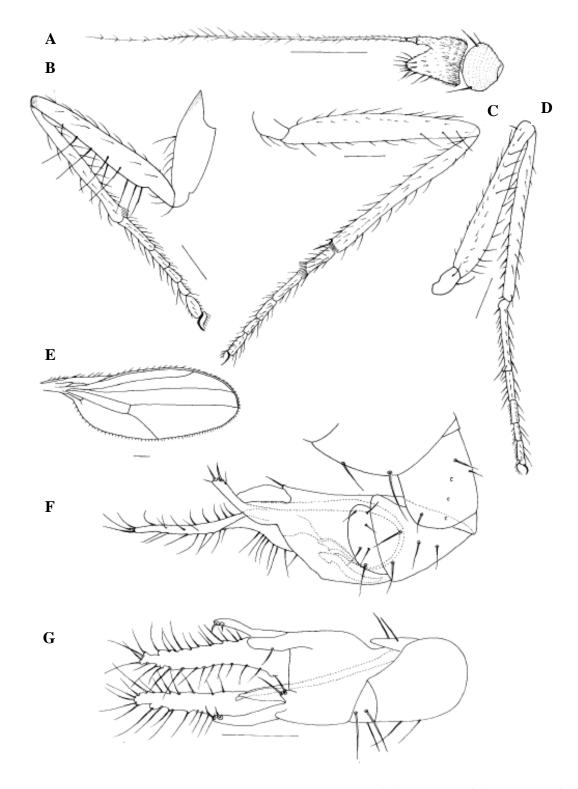


Figure 63. *Phacaspis mitis* Grootaert and Meuffels, 2001. (A) Antenna; (B) Fore leg; (C) mid leg; (D) hind leg; (E) Wing; (F) hypopygium laterally view; (G) hypopygium dorsally view. Scale = 0.1 mm.



Figure 64. Male habitus of *Phacaspis mitis* Grootaert and Meuffels, 2001

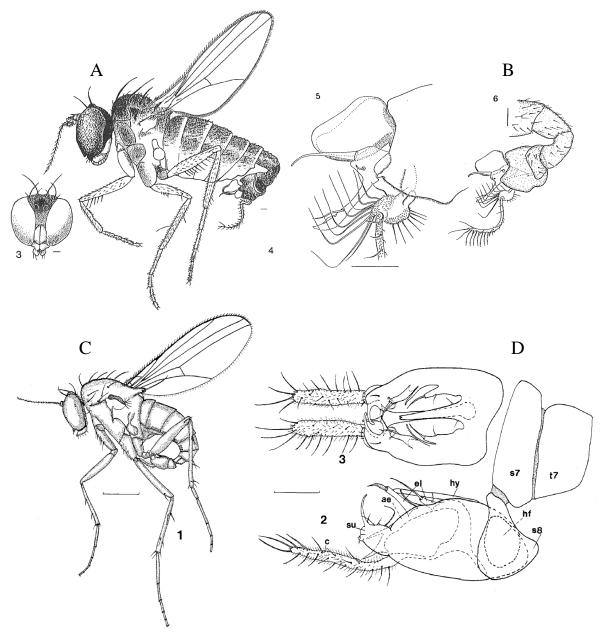


Figure 65. *Phacaspis ornata* Meuffels, 1998, (A) male habitus, (B) male genitalia (adapted from Meuffels, 1998); *Kowmungia nigrifemorata* Becker, 1987, (C), male habitus, (D) male genitalia (modified from Bickel, 1987).

#### Ornamenta gen nov.

#### (New genus)

### Ornamenta siamese sp. nov.

(Figure 66)

**Material Examined.**  $3\overset{\circ}{,}5\overset{\circ}{,}$ ; Thailand, Bakan Toh Thid, Langu, Satun (6°47'29.8"N 99°48'53.5"E), sweep netting, 4 May 2015, coll. A. Samoh.  $5\overset{\circ}{,}5\overset{\circ}{,}$ ; Thailand, Taturum, Kam Phaeng, Langu, Satun (6°55'10.1"N 99°43'59.0"E), sweep netting, 4 May 2015, coll. A. Samoh.

**Remarks.** This species will be announced for a new species and a new genus. In term of general external morphology is most resembled to *Phacaspis mitis*.

**Distribution.** Langu and Muang, Satun (Andaman Sea)

# Terpsimyia semicincta

**Material Examined.**  $3\stackrel{\wedge}{_{2}}2\stackrel{\circ}{_{1}}$ ; Thailand, Dato, Yaring, Pattani, (6°55'17.1"N 101°19'50.7"E), sweep netting, 14 April 2015, coll. A. Samoh;  $5\stackrel{\wedge}{_{3}}5\stackrel{\circ}{_{1}}$ ; Thailand, Prince of Songkla University, Pattani (6°50'30.4"N 99°46'32.9"E), sweep netting, 13 April 2015, coll. A. Samoh.

**Remarks.** This species has been reported from Gulf of Thailand by Grootaert and Meuffels (2001). This study is also recorded from Gulf of Thailand Sea, lots of T. *semicincta* were sampled from Pattani mangroves. Large mudflats seem to be a major habitat for this long-legged fly.

Distribution. Pattani, Songkhla (Gulf of Thailand)



Figure 66. Male habitus, Ornamenta siamese sp. nov.



Figure 67. Male Habitus, Terpsimyia semicincta Becker, 1922.

		Distribution Area		Habitat
Taxa	Status	Andaman Sea	Gulf of Thailand	Habitat Preference
Diaphorinae				
Asyndetus Loew, 1869				
1) Asyndetus aciliatus	-	+	-	SB
2) Asyndetus thaicus	-	+	-	SB
3) Asyndetus sp.	New species	+	-	SB
Chrysotus Meigen, 1824				I
4) Chrysotus dot	New record	+	+	М
Diaphorus Meigen, 1824	1		1	1
5) <i>Diaphorus</i> sp.	New species	+	+	М
Dolichopodinae				I
Argyrochlamys Lamb, 1922				
6) Argyrochlamys impudicus	New record	+	+	RB,SB
Hercostomus Loew, 1857				I
7) Hercostomus brevicornis	New record	+	+	М
8) Hercostomus brevidigitalis	New record	+	-	М
9) Hercostomus lanceolatus	New record	+	+	М
10) Hercostomus obtusus	New record	+	-	М
11) Hercostomus plumatus	New record	+	+	М
12) Hercostomus propermeieri	New species	+	-	М
Lichtwardtia Enderlin, 1921				
13) Lichtwardtia ziczac	New record	-	+	М
Paraclius Loew, 1864				
14) Paraclius adligatus	New record	+	+	М
15) Paraclius asiobates	New record	-	+	М
16) Paraclius digitatus	New record	+	+	М
17) Paraclius obtus	New record	-	+	М
18) Paraclius serratus	New record	+	+	М
19) Paraclius singaporensis	New record	-	+	М
20) Paraclius sp. nov.	New species	-	+	М
Tachytrechus Haliday, 1851		1	1	1
21) Tachytrechus tessellatus	New record	+	+	SB

Table 1. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach.

Table 1. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach (cont.).

	Status	Distribution Area		Habitat
Taxa		Andaman	Gulf of	Preference
		Sea	Thailand	
Phoomyia Naglis and Grootaert, 200	3		L	
22) Phoomyia singaporensis	New record	+	+	RB,SB
23) <i>Phoomyia</i> sp. nov.	New species	-	+	RB,SB
Hydrophorinae				
Cymatopus Kertész, 1901				
24) Cymatopus malayensis	-	+	+	RS
25) Cymatopus thaicus	-	+	-	RS,SB
26) Cymatopus mayakunae sp. nov.	New species	+	-	RS
Nanothinophilus Grootaert and Meuf	fels, 1998			
27) Nanothinophilus hoplites	-	+	-	M.SB
28) Nanothinophilus pauperculus	-	+	-	M,SB
Thambemyia Oldroyd, 1956				
29) Thambemyia pagdeni	-	+	+	RB,SB
Thinophilus Wahlberg, 1844				
30) Thinophilus apicatus	New record	+	+	М
31) Thinophilus boonrotpongi	New species	+	+	М
32) Thinophilus chaetulosus	New record	-	+	М
33) Thinophilus langkawensis	New species	+	-	M,SB
34) Thinophilus melanomerus	New record	+	-	М
35) Thinophilus minutus	New species	+	+	М
36) Thinophilus parmatoides	New species	-	+	М
37) Thinophilus parmatus	-	+	-	М
38) Thinophilus parvulus	New species	-	+	М
39) Thinophilus sp. nov	New species	+	-	М
40) Thinophilus simplex	New record	+	+	М
41) Thinophilus spinatoides	New species	+	-	М
42) Thinophilus spinatus	New species	+	-	М
43) Thinophilus superbus	New record	+	-	М
44) Thinophilus variabilis	New species	+	+	М
45) Thinophilus yeoi	New record	-	+	М

Table 1. Species composition, habitat preference, distribution area and status of the marine long-legged flies in Thai Peninsula. M: mangrove; RB: rocky beach; RS: Rocky shore; and SB: Sandy beach (cont.).

Microphorella Becker, 190946) Microphorella malaysiana-++RB,SBRhaphiinaeNgirhaphium Evenhuis and Grootaert, 200247) Ngirhahium caeruleumNew record-+M48) Ngirhahium caeruleumNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-MSciapodinaeAmblypsilopus Bigot, 185952) Amblypsilopus abruptum+MSympycninae+MChrysosoma leucopogon+MSympycninaeChaetogonopteron de' Meijere, 1914State of Sympycnus Loew 185756) Sympycnus Sp.New record-+MSympycnus Loew, 185757) Teuchophorus Loew, 1857-+-M	Tarra	Status	Distribution Area		Habitat
ParathalassiinaeMicrophorella Becker, 190946) Microphorella malaysiana-++RB,SBRhaphiinaeNgirhaphium Evenhuis and Grootaert, 200247) Ngirhahium caeruleumNew record-+M48) Ngirhahium caeruleumNew record-+M49) Ngirhahium chutamasaeNew species+-M50) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-MStappolinaeChaetogonopteron de' Meijere, 1914SympyeninaeChaetogonopteron de' Meijere, 191454) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympyenus Loew 185756) Sympyenus sp.New record-+M57) Teuchophorus Loew, 1857-+-M	Taxa	Status	Andaman	Gulf of	Preference
Microphorella Becker, 190946) Microphorella malaysiana-++RB,SBRhaphiinaeNgirhaphium Evenhuis and Grootaert, 200247) Ngirhahium caeruleumNew record-+M48) Ngirhahium caeruleumNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-MSciapodinaeAmblypsilopus Bigot, 185952) Amblypsilopus abruptum+MSympycninae+MChrysosoma leucopogon+MSympycninaeChaetogonopteron de' Meijere, 1914State of Sympycnus Loew 185756) Sympycnus Sp.New record-+MSympycnus Loew, 185757) Teuchophorus Loew, 1857-+-M			Sea	Thailand	
46) Microphorella malaysiana-++RB,SBRhaphinaeNgirhaphium Evenhuis and Grootaert, 200247) Ngirhahium caeruleumNew record-+M48) Ngirhahium caeruleumNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M50) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-MSciapodinaeAmblypsilopus Bigot, 185952) Amblypsilopus abruptum+MChrysosoma Guerin-Meneville, 183153) Chrysosoma leucopogon+MSympycninaeChaetogonopteron de' Meijere, 191454) Chaetogonopteron vexillum-++MSympycnus Loew 185756) Sympycnus sp.New record-+MTeuchophorus Loew, 185757) Teuchophorus krabiensis-+-M	Parathalassiinae				
Rhaphiinae         Ngirhaphium Evenhuis and Grootaert, 2002         47) Ngirhahium caeruleum       New record       -       +       M         48) Ngirhahium chutamasae       New species       +       -       M         49) Ngirhahium chutamasae       New species       +       -       M         50) Ngirhahium meieri sp. nov.       New species       +       -       M         50) Ngirhahium murphyi       New record       +       -       M         50) Ngirhahium sivasothii       New record       +       -       M         51) Ngirhahium sivasothii       New record       +       -       M         Sciapodinae       -       -       +       M         Chrysosoma Guerin-Meneville, 1831       -       -       +       M         Sympycniae       -       -       +       M       M	Microphorella Becker, 1909				
Ngirhaphium Evenhuis and Grootaert, 200247) Ngirhahium caeruleumNew record-+M48) Ngirhahium chutamasaeNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M50) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-M52) Amblypsilopus Bigot, 1859+M52) Amblypsilopus abruptum+M53) Chrysosoma leucopogon+M53) Chrysosoma leucopogon+M54) Chaetogonopteron de' Meijere, 1914-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 1857-+M57) Teuchophorus Loew, 1857-+-M	46) Microphorella malaysiana	-	+	+	RB,SB
47) Ngirhahium caeruleumNew record-+M48) Ngirhahium chutamasaeNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-M52) Amblypsilopus Bigot, 1859+M52) Amblypsilopus abruptum+MChrysosoma Guerin-Meneville, 1831+M53) Chrysosoma leucopogon+M54) Chaetogonopteron de' Meijere, 1914-++M55) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 1857-+MM57) Teuchophorus Loew, 1857-+-M	Rhaphiinae				
48) Ngirhahium chutamasaeNew species+-M49) Ngirhahium meieri sp. nov.New species+-M50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-M52) Amblypsilopus Bigot, 1859+M52) Amblypsilopus abruptum+MChrysosoma Guerin-Meneville, 1831+M53) Chrysosoma leucopogon+M54) Chaetogonopteron de' Meijere, 1914-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 1857-+M57) Teuchophorus Loew, 1857-+-M	Ngirhaphium Evenhuis and Grootaert	, 2002			
Age of the second se	47) Ngirhahium caeruleum	New record	-	+	М
50) Ngirhahium murphyiNew record+-M51) Ngirhahium sivasothiiNew record+-M51) Ngirhahium sivasothiiNew record+-MSciapodinaeAmblypsilopus Bigot, 185952) Amblypsilopus abruptum+MChrysosoma Guerin-Meneville, 1831+M53) Chrysosoma leucopogon+MSympycninae+MChaetogonopteron de' Meijere, 1914-++M55) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++MSympycnus Loew 1857-+M56) Sympycnus Loew, 1857-+M57) Teuchophorus krabiensis-+-M	48) Ngirhahium chutamasae	New species	+	-	М
51) Ngirhahium sivasothiiNew record+-MSciapodinaeAmblypsilopus Bigot, 185952) Amblypsilopus abruptum+MChrysosoma Guerin-Meneville, 183153) Chrysosoma leucopogon+MSympycninaeChaetogonopteron de' Meijere, 191454) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 185756) Sympycnus sp.New record-+M57) Teuchophorus Loew, 1857-+-M	49) Ngirhahium meieri sp. nov.	New species	+	-	М
Sciapodinae         Amblypsilopus Bigot, 1859         52) Amblypsilopus abruptum       -       -       +       M         52) Amblypsilopus abruptum       -       -       +       M         Chrysosoma Guerin-Meneville, 1831       -       -       +       M         53) Chrysosoma leucopogon       -       -       +       M         Sympycninae       -       -       +       M         Chaetogonopteron de' Meijere, 1914       -       +       +       M         54) Chaetogonopteron chaeturum       -       +       +       M         55) Chaetogonopteron vexillum       -       +       +       M         Sympycnus Loew 1857       -       +       M         56) Sympycnus sp.       New record       -       +       M         Teuchophorus Loew, 1857       -       +       -       M	50) Ngirhahium murphyi	New record	+	-	М
Amblypsilopus Bigot, 185952) Amblypsilopus abruptum+M53) Chrysosoma Guerin-Meneville, 183153) Chrysosoma leucopogon+M53) Chrysosoma leucopogon+MSympycninaeChaetogonopteron de' Meijere, 191454) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 185756) Sympycnus sp.New record-+M57) Teuchophorus Loew, 1857-+-M	51) Ngirhahium sivasothii	New record	+	-	М
52) Amblypsilopus abruptum       -       +       M         65) Amblypsilopus abruptum       -       +       M         63) Chrysosoma Guerin-Meneville, 1831       -       +       M         63) Chrysosoma leucopogon       -       -       +       M         63) Chrysosoma leucopogon       -       -       +       M         65) Chaetogonopteron de' Meijere, 1914       -       +       +       M         65) Chaetogonopteron chaeturum       -       +       +       M         65) Chaetogonopteron vexillum       -       +       +       M         65) Chaetogonopteron vexillum       -       +       +       M         56) Sympycnus Loew 1857       -       +       M         57) Teuchophorus Loew, 1857       -       +       -       M	Sciapodinae		L	I	•
Chrysosoma Guerin-Meneville, 1831         53) Chrysosoma leucopogon       -       +       M         53) Chrysosoma leucopogon       -       -       +       M         Sympycninae       -       -       +       M         Chaetogonopteron de' Meijere, 1914       -       +       +       M         54) Chaetogonopteron chaeturum       -       +       +       M         55) Chaetogonopteron vexillum       -       +       +       M         55) Chaetogonopteron vexillum       -       +       +       M         Sympycnus Loew 1857       -       +       M         56) Sympycnus sp.       New record       -       +       M         Teuchophorus Loew, 1857       -       +       -       M	Amblypsilopus Bigot, 1859				
53) Chrysosoma leucopogon-+M53) Chrysosoma leucopogon-+HSympycninaeChaetogonopteron de' Meijere, 191454) Chaetogonopteron chaeturum-++55) Chaetogonopteron vexillum-++55) Chaetogonopteron vexillum-++55) Chaetogonopteron vexillum-++56) Sympycnus Loew 1857New record-+56) Sympycnus sp.New record-+760) Sympycnus Loew, 1857-+M57) Teuchophorus krabiensis-+-M	52) Amblypsilopus abruptum	-	-	+	М
Sympycninae         Chaetogonopteron de' Meijere, 1914         54) Chaetogonopteron chaeturum       -       +       +       M         55) Chaetogonopteron vexillum       -       +       +       M         55) Chaetogonopteron vexillum       -       +       +       M         56) Sympycnus Loew 1857       -       +       M         56) Sympycnus sp.       New record       -       +       M         Teuchophorus Loew, 1857       -       +       -       M         57) Teuchophorus krabiensis       -       +       -       M					
Chaetogonopteron de' Meijere, 191454) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++M56) Sympycnus Loew 1857-+M56) Sympycnus Loew, 1857-+M57) Teuchophorus krabiensis-+-	53) Chrysosoma leucopogon	-	-	+	М
54) Chaetogonopteron chaeturum-++M55) Chaetogonopteron vexillum-++M55) Chaetogonopteron vexillum-++MSympycnus Loew 185756) Sympycnus sp.New record-+MTeuchophorus Loew, 1857-+M57) Teuchophorus krabiensis-+-M					
55) Chaetogonopteron vexillum-++MSympycnus Loew 185756) Sympycnus sp.New record-+MTeuchophorus Loew, 185757) Teuchophorus krabiensis-+-M	Chaetogonopteron de' Meijere, 1914				
55) Chaetogonopteron vexillum-++MSympycnus Loew 185756) Sympycnus sp.New record-+MTeuchophorus Loew, 185757) Teuchophorus krabiensis-+-M	54) Chaetogonopteron chaeturum	-	+	+	М
Sympycnus Loew 185756) Sympycnus sp.New record-+MTeuchophorus Loew, 1857-+-M57) Teuchophorus krabiensis-+-M		-	+	+	М
Teuchophorus Loew, 1857         57) Teuchophorus krabiensis					
57) Teuchophorus krabiensis - + - M	56) Sympycnus sp.	New record	-	+	М
57) Teuchophorus krabiensis - + - M	Teuchophorus Loew, 1857		I		1
	<u>`</u>	-	+	_	М
Incertae Sedis (Unplaced Group)					
	Ornamenta gen. nov.				
	58) Ornamenta siamese sp. nov.	New species	+	-	М
Phacaspis Meuffels and Grootaert, 1990					
	59) Phacaspis mitis	-	+	+	М
	Terpsimyia Becker, 1922	1	1	1	<u>'</u>
	60) Terpsimyia semicincta	-	-	+	М
	7 subfamilies, 1 unplaced group				

Over 3,800 specimens of marine long-legged flies in Thai Peninsula were investigated and identified based on male genital and non-genital morphological features scrutiny. It was found that the west coast of Thai Peninsula (Andaman Sea, Indian Ocean) contained higher number of species (23 morphospecies) of marine long-legged flies than the Gulf of Thailand Sea (South China Sea, Pacific Ocean) (15 morphospecies). In other words, marine long-legged flies in Thai Peninsula were greater in number of species in the Andaman seacoasts than Gulf of Thailand seacoasts, especially in mangrove habitat (Table 9). This study confirms previously surveyed report of marine long-legged flies in Thailand by Grootaert and Meuffels (2001). These authors identified thirteen morphospecies from Andaman Sea side, whereas only six species were observed from Gulf of Thailand. This could be hypothesized that the larger size and complexity of mangrove in Andaman Sea might support a greater number or variety of marine longlegged fly species than the Gulf of Thailand. According to the report of Department Of Marine And Coastal Resources (DMCR) in 2009 (in Thai version), it was clearly shown that Andaman seacoasts (1,104,892.87 RAI) contained larger mangrove size than Gulf of Thailand Sea (182,934.01 RAI). Moreover, the result also notably supports the theory of island biogeography of organisms proposed by McAthur and Wilson (1967), that the larger island may support more number of organismal species on the island.

The richest genera of marine long-legged flies in Thailand elucidated were Thinophilus Wahlberg (16 species) and Paraclius Loew (7 species), but the distribution pattern and number of specimen are confusing and uninteresting. Because several of the species from both genera were low in number of individual or specimens. For example, Thinophilus parvulus sp. nov., only a pair of them that could be captured from Pattani mangrove only, and one more problematic species Thinophilus spinatus sp. nov. which was mainly swept from Ban Bakan Toh Thid, Langu district, Satun province also provided little number of individual, when Paraclius adligatus, was solely sampled from Ban Khao Than mangrove, Tha Chang district, Surat Thani province. Unluckily, only five female specimens of this species could be trapped by Malaise trap and hand collection. However, one of the most striking results in the context of distribution pattern of the present study showed that Hercostomus lanceolatus belonging to Hercostomus Loew, 1857 was the largest range of distribution. It was widely distributed in several mangroves from the two sides of Thai Peninsula (Table 9). For example, in the Gulf of Thailand Sea, Hercostomus occurred abundantly in several mangroves in Pattani province such as replanted mangrove very near sea front at the Prince of Songkla University, Pattani campus; a clumped mangrove at Ban Dato, Yaring district, Pattani province; one back mangrove in Ban Nathab, Chana district, Songkhla province; and also largely dwelled in disturbed mangroves at Ban Khao Than, Tha Chang district, Surat Thani province; and abundantly observed from Ban Phanangtak, Muang district, Chumphon province. For the Andaman Sea, this species was obviously observed from Tammalang mangrove, Muang district, Satun province; and also sampled from Ban Bang

Nai Si and Ban Bang Dong mangroves, Takuapa district, Phang Nga province (Figure 7, circled with red colour). Whereas the genus Ngirhaphium Evenhuis and Grootaert remarkably occurred in different way and provides an interesting data in term of distribution pattern, with majority of species in this genus mostly restricted to particular mangroves. For instance, N.chutamasae sp.nov. is only occurred in Tammalang mangrove, while N.caeruleum occurred in mudflats of Surat Thani and Chumphon bays, and a new species N.meieri sp.nov. (pending for description), was restricted to a back mangrove with unique environmental condition at Ban Bang Dong, Takuapa district, Phang Nga province (Figure 8). Due to these facts, it could be concluded that those mentioned species are notably interesting in the context of species identification, distribution, and understanding of the genetic diversity and variation. Further results also elucidated that most species of marine long-legged flies occurring in Thailand are largely congruent to the Singaporean species than the other countries in Southeast Asia (SEA), with more than 37 species conforming to type (Evenhuis and Grootaert, 2002; http://evolution.science.nus.edu.sg/MIP.html), especially species the that were specifically collected from the Gulf of Thailand Sea. This could be simplifying this natural event by the locality of geography. If Singapore country and the border of the sea water were taken into account, it could be said that both the Gulf of Thailand and Singapore lies in the same side of the South China Sea (Figure 9), across Malay peninsula and without any natural barrier to limit the dispersal ability of this flies from place to place. This is the reason why most species are similar between the two countries.

However, due to lack of many available information or no survey from other neighbouring countries such as Malaysia, Brunei, Indonesia, Myanmar, Cambodia, Philippines, and Vietnam (Figure 9) which were also recognised as countries that are endowed with marine habitats, it may provide poor and not precise data of this group of flies in the context of species distribution. Nonetheless, this survey is an evidence to support the marine zoogeographical distribution in this region

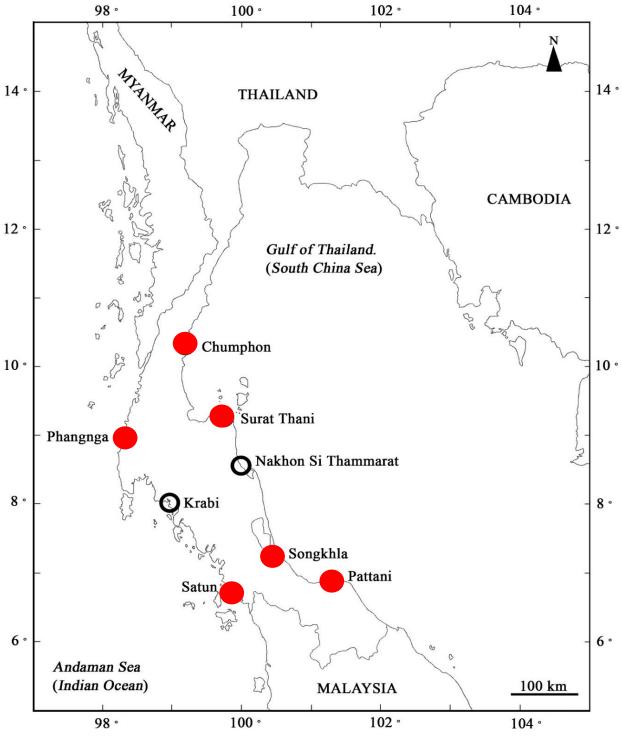
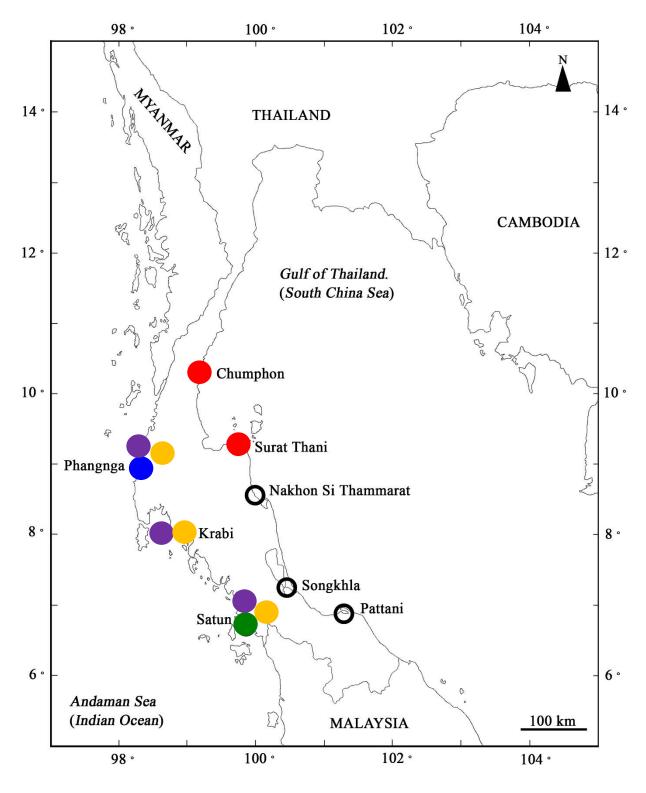
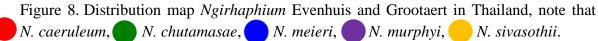


Figure 7. Distribution map of *Hercostomus lanceolatus* in Thailand.





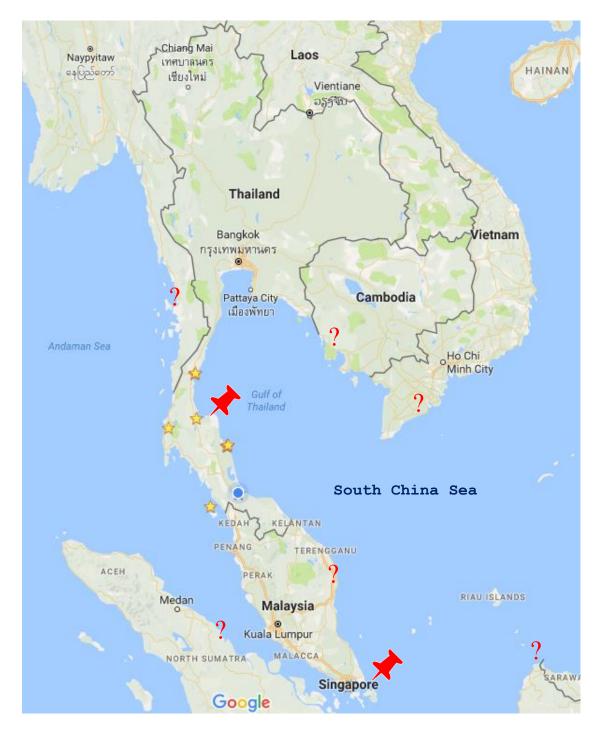


Figure 9. Map of Thailand and Singapore, red pins indicate the countries.

In various available publications (Miall, 1934, Cregan, 1941; Dytes, 1959; Pollet, 2000, 2001; Grootaert and Meuffels, 2004; Brooks, 2005; Ulrich, 2005; Grootaert, 2006) on natural history of long-legged flies in the world, it was reported that both larvae and adult of long-legged flies abundantly occurred in moist and humid habitats such as in saltmarshes, seashores, lakes, streams, canals, mangroves, rocky shores, humid rocky and sandy beaches, tide pools, waterfalls, freshwater seepages, damn soil, humid forests, swamps (Dytes, 1959; Pollet, 2001; Brooks, 2005; Ulrich, 2005, Grootaert, 2006), and also occurred in drier habitats such as agricultural fields, grasslands, and urban gardens (Books, 2005). The result of habitat preferences of the current study clearly indicated that mangrove habitat composed of the highest number of species and was assumed to be major marine habitats that could support a large number of species. In addition, this finding has led to conclusion that the most preferred habitat by marine longlegged flies in Thailand is mangrove. This could be explained by the very moist environment in containing more complexity of microhabitat than other marine habitats such as rocky and sandy beaches, rocky shores, or even tide pools. If take all those marine habitats (mangroves, rocky shores, sand and rocky beaches, tide pools) into account, and found that mangrove largely goes along with basic information on natural history of long-legged flies that many of the species largely prefer moist environments, and of course mangrove show merely fitted to the definition of high humid and moist atmospheres comparing to other marine habitats such as rocky shores, sandy-rocky beaches, tide pools. Moreover, it also contains various kinds of microhabitats and assumed that it allowed and supported species of marine long-legged flies into this habitat. Similarly, previous study by Grootaert and Meuffels (2001) have suggested that each marine species has their own favored habitats. For example, *Terpsimvia semicincta*, virtually lives in mangrove mudflats with high exposure to sun-light in the Gulf of Thailand sea side which is generally influenced by sea level of the day Grootaert and Meuffels (2001).

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## **Manuscript II**

New data on the marine genera *Cymatopus* Kertesz and *Thambemyia* Oldroyd (Insecta, Diptera, Dolichopodidae) from rocky shores in southern Thailand with the description of a new sp

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#### Abstract

Four species of the genus *Cymatopus* Kertèsz and one species of *Thambemyia* Oldroy, 1956 are found in southern Thailand. A key is given for all 5 species and new data of their distribution are provided. *Cymatopus mayakunae* is described as new for science. COI barcodes seem to be good indicators for the species delimitation but do not support a phylogeny.

#### Introduction

The present study was in the scope of a survey of the marine dolichopodid flies occurring in peninsular Thailand (Samoh *et al.*, 2015; 2017). During this study, a number of *Cymatopus* and *Thambemya* specimens were occasionally collected on rocky shores and sandy beaches. Both genera *Cymatopus* and *Thambemyia* are in fact true marine genera of which the adults are found foraging and displaying on rocks in the intertidal zone. The larvae and the pupae live inside the crusts of debris and algae on these rocks (Grootaert & Meuffels, 1993).

The genus *Cymatopus* is represented in Thailand by three species groups. The *longipilus*-group is characterised in the male by simple unmodified fore legs, but with modified hind legs bearing long bristles and hairs. The other two species groups have the fore legs modified and ornamented, but the hind legs are simple. The *malayensis*-group is composed of larger species with the hind border of the wing notched in the male and with fields of enlarged microtrichia on the wing membrane. The *thaicus*-group is composed of smaller species with the hind border of the wing not or a little folded and without fields of enlarged microtrichia on the wing membrane.

The genus *Thambemyia* is characterized by the long mouthparts that resemble an elephant snout. They are generally found in the splash zone on cliffs. Occasionally they are also found on the vertical concrete pillars of a jetty.

In the present paper, we provide new distribution data of *Cymatopus* and *Thambemyia* with a key and illustrations of the habitus (except for *C. longipilus*). A new species of *Cymatopus* is described from the coast of the Andaman Sea. NGS barcodes of 312 base-pairs provide good species delimitation the observed species.

#### Material and methods

The specimens were hand-collected or with a sweep-net during a survey of both coasts of peninsular Thailand. All specimens were preserved in 70% ethanol in a refrigerator preventing DNA degradation. Type material is deposited in the collections of the Princess Maha Chakri Sirindhon Natural History Museum of the Prince of Songkla University (NHM-PSU), Hat Yai, Songkhla, Thailand. The locality of the holotype is considered as the type locality. In addition, a few voucher specimens are also preserved in the collections of the Royal Belgian Institute of Natural Sciences (RBINS), Brussels, Belgium.

A focus stacking technique (see Brecko *et al.*, 2014) was used to photograph all specimens. The high-resolution pictures were stacked using Zerene Stacker software. The scale on the photos is 1 mm.

The NGS barcoding using COI sequencing was done according to Meier *et al.* (2015). The evolutionary history was inferred using the Neighbor-Joining method (Saitou & Nei, 1987). The optimal tree with the sum of branch length = 0.50267780 is shown. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches (Felsenstein, 1985). The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (Tamura *et al.*, 2004) and are in the units of the number of base substitutions per site. The analysis involved 53 nucleotide sequences. Codon positions included were 1st+2nd+3rd+Noncoding. All positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar *et al.*, 2016).

#### **Taxonomic account**

Key to male Cymatopus and Thambemyia in Thailand

1.	Proboscis much shorter than height of an eye (Fig. 1) Cymatopus	2
-	Proboscis much longer than height of an eye (Figs 11, 12)	
•••		ł

## The malayensis-group

Only one species in Thailand.

#### Cymatopus malayensis Parent, 1935

Figs 1 – 2

Parent, 1935a: 208 (figs 26-31).

*Material examined.* – 16 males, 16 females, Laem Kho Kwang, Chumphon Province, Gulf of Thailand (South China Sea), 17 February 2015, sweep netting, 10°30'48.7"N, 99°15'52.0"E; 13 male, 13 females, Ban Thong Tom Yai, Sawee, Chumphon Province, Gulf of Thailand, 19 February 2015, sweep netting, 10°12'39.2"N 99°12'21.4"E; 8 males, 4 males, Tarutao Island, Langu, Satun Province, Andaman Sea (Indian Ocean), 9 January 2015, sweep netting, 6°44'19.2"N 99°38'45.4"E, coll. Abdulloh Samoh.

*Diagnosis.* - A large species with modified fore leg. Fore tibia anteriorly with a black foliaceous bristle and metatarsus (tarsomere 1) elongated with a distorted tip bearing strong twisted bristles. Fore tarsomeres 4 and 5 flattened and brown.

*Bionomics.* - The adult flies are found in the splash zone of rocky shores.

*Distribution.* – Shores of the Andaman Sea and the Gulf of Thailand (Grootaert & Meuffels, 2001). Thailand, Malaysia, Singapore and Borneo.

### The thaicus-group

The *thaicus*-group is just based on the smaller size of the specimens and the shape of the wings. However, this grouping based on morphological characters is genetically not supported as can be seen on a neighbour joining tree based on the COI gene (Fig. 10).

The group-name is provisionally conserved as such. It is represented by two species in Thailand: *C. thaicus* and *C. mayakunae* new species

## Cymatopus mayakunae new species

## Figs 3 – 5

# Material examined.

**Holotype male**: THAILAND, Laem Pakarang, Khao Lak, Takuapa, Phanga Nga Province (Andaman Sea), 8°44'09.9" N, 98°13'21.5" E, 10 February 2015, sweep netting, coll. A. Samoh.

**Paratypes**: 30 males, 10 females, same collection as holotype. 13 males, 6 females, Tarutao Island, Langu, Satun Province (Andaman Sea), 6°44'19.2" N 99°38'45.4" E, 9 January 2015, sweep netting, coll. A. Samoh.

*Derivatio nominis*. The species is dedicated to Dr. Jaruwan Mayakun, a person who takes the first author to sample *Cymatopus* flies in Langu mangroves, Satun Province (Andaman Sea).

**Diagnosis.** A small species (2-2.5 mm) with yellow legs. Fore tibia without black foliaceous bristle and without apical spur and apical bristle. Fore tibia dorsally near base with short bent bristles. Hind tibia with a dorsal row of bristles with dilated tips. Wing with veins  $R_1$  and  $R_{2+3}$  deformed and thickened. Posterior wing border a little deformed with longer hairs.

Male (Fig. 3)

Body length 2.5 mm; wing length 2.5 mm.

Head. Frons and face black in ground-colour, greyish dusted. Clypeus protruding. Face wider than postpedicel is wide. Palpus brown with short black hairs, tips of apical bristles pale. A pair of strong ocellars, a pair of slightly shorter fronto-orbitals and a pair of minute postocellars. Postocular bristles black above, becoming whitish and hair-like below. Antenna black, pedicel darker than scape and postpedicel. Postpedicel conical, 1.5 times as long as wide. Arista nearly twice as long as scape, pedicel and postpedicel together.

Thorax black in ground-colour, greyish dusted. No acrostichals, 5 dc (anterior 4 equally long, prescutellar dc longer); a pair of long scutellars with a minute hair at ouside. A minute humeral, a very long posthumeral, a short sutural, a minute notopleural, and a longer supra-alar and a long postalar. 3 pale propleurals.

Legs yellow (Fig. 3) with mid and hind coxae black, apical two tarsomeres slightly brownish. Fore leg. Coxa with 2-3 short black bristles at base and some longer black apical bristles. Femur swollen in basal half with a long posteroventral bristles, near base as long as femur is wide, in apical half longer than femur is wide. The row is interrupted at the basal third and there 2 shorter bristles. Tibia as long as femur, without apical spur and without apical spine-like bristle; ventrally set with a double row of spine-like bristles as long as tibia is wide; basal fifth of tibia dorsally set with a double row of short bristles with curved tip. Tarsomeres not flattened. Mid leg. Coxa with 2 short black exterior bristles. Femur much longer and thinner than fore femur. Tibia shorter than femur without particular bristles. Hind leg. Coxa with a short black exterior bristle. Femur a little wider than mid femur and shorter. Tibia dorsally set with a double row of short bristles with enlarged tips (Fig.3).

Wing brownish tinged with brown veins. Costa near middle darker brown and slightly bowed. Costa and  $R_{2+3}$  (Fig. 3) thickened and undulating near middle. Apical half of Cu pale, the hind border is a little notched there and the wing membrane is folded to the exterior and bears some longer bristles at that level. Haltere and squama white, bearing long white cilia.

Abdomen black in ground-colour, greyish dusted. Tergites with minute black on apical border. Terminalia (Fig. 5). Cercus yellow with brown bristles longer than cercus is wide.

Female (Fig. 4)

Body length: 2.0 mm, wing length: 2.0 mm

Identical to male but fore legs with shorter bristles and wing with veins not deformed. Fore tibia and hind tibia with normal bristling.

Bionomics. - The adult flies are found in the splash zone of rocky shores.

Distribution. – Shores of the Andaman Sea.

*Remarks.* – The new species is unique in the genus having the fore tibia dorsally near base set with short bent bristles. The hind tibia has a dorsal row of bristles with dilated tips. Veins  $R_1$  and  $R_{2+3}$  are deformed and thickened. In addition, the posterior wing border is a little deformed bearing longer hairs.

### Cymatopus thaicus Grootaert & Meuffels, 2001

#### **Figs 6 – 8**

Grootaert & Meuffels, 2001: 351 (figs 31-36).

*Material examined.* – 4 males, Lidi Island, Langu, Satun Province (Andaman Sea), 6°46'56.4"N, 99°45'58.5"E, 30 July 2015, sweep netting, coll. A. Samoh; 12 male, 4 females, Tarutao Island, Langu, Satun Province (Andaman Sea), 6°44'19.2"N 99°38'45.4"E, 9 January 2015, sweep netting, coll. A. Samoh.

*Diagnosis.* - A small species (2-2.2 mm) with yellow legs. Fore tibia with a black foliaceous bristle and without an apical spur, but with a long black apical bristle. Tarsomere 4 laterally flattened but not excavated and as long as flattened tarsomere 5. Posterior wing border normal, set with equally long hairs. Veins not deformed.

Distribution. - Thailand, Malaysia and Singapore.

#### The *longipilus*-group

Only one species in Thailand.

#### Cymatopus longipilus Parent, 1935

Fig. 9

Parent, 1935b: 61 (figs 5-7).

*Material examined.* – No new material was found during the present study.

*Diagnosis.* - A very small (1.75 mm) dark species with dark brown legs. Tip of hind tibia in male enlarged and bearing very long black bristles, continuing on the hind metatarsus.

*Bionomics.* - The adult flies are found in the splash zone of rocky shores. Often, they are found on vertical oyster beds.

*Distribution.* - Shores of the Andaman Sea and the Gulf of Thailand (Grootaert & Meuffels, 2001), Christmas Islands (type locality).

*Thambemyia* Oldroyd, 1956: 210. Type-species: *T. pagdeni* Oldroyd (original designation).

*Conchopus* Takagi, 1965: 49. Type-species: *C. rectus* Takagi (original designation). Synonymized by Meuffels & Grootaert (1984), but see Masunaga *et al.* (2005) and Masunaga & Saigusa (2010).

Subgenus Thambemyia Oldroyd, 1956

Meuffels & Grootaert (1984) established the synonymy of *Conchopus* with *Thambemyia*, a decision endorsed in recent catalogues (Bickel & Dyte 1989; Pollet *et al.* 2004; Yang *et al.* 2006). Nevertheless, Masunaga *et al.* (2005) and Masunaga & Saigusa (2010) reported on their unpublished phylogeny, in which they found that *Conchopus* in the sense of Takagi (1965) and *Thambemyia* are not sister groups. Rather, there are three distinct clades: (1) the *rectus*-group of *Conchopus*, comprising the genus concept in a narrower sense, (2) the species of *Conchopus* exclusive of the *rectus*-group, and (3) *Thambemyia*. The two later are sister groups and a new genus name would be established for the clade (2).

In this scenario, the issue of establishing such a new genus is actually a decision about the generic limits of *Thambemyia*, since the species of *Conchopus* exclusive the *rectus*-group could be well accommodated into an expanded concept of *Thambemyia*.

## Thambemyia pagdeni Oldroyd, 1956

# Figs 11 - 12

Oldroyd, 1956: 211. Type locality: Malaysia: Penang.

*Material examined.* – 23 males, 15 females, Sakom (Tepha), Songkhla Province, Gulf of Thailand (South China Sea), 28 March 2017, cliffs, 6°57'42,97"N 100°50'57.02"E; 29 March 201, pier on sandy beach 6°56'52,88"N 100°51'52.72"E.

1 male, 2 females Khao Lak, Nangtong, Phang Nga Province, rocky beach (reg. 96050, leg. P. Grootaert).

*Diagnosis.* - Medium-sized (4-4.5 mm) black species with long yellowish brown to black legs. Mainly characterised by the long trump-like mouthparts. The fore legs (Fig. 11) have tarsomere 1 with a ventral notch, tarsomere 2 with a basal protuberance and tarsomere 5 enlarged and flattened. The postpedicel is elongated triangular (2.5 times as long as wide) with a long apical arista. Female with simple fore tarsus (Fig. 12).

*Bionomics.* - The adult flies are found in the splash zone of vertical walls such as cliffs and pillars of jetties.

*Distribution.* – Thailand, Malaysia and Indonesia.

# Acknowledgements

This research was supported by the Higher Education Research Promotion and National Research University (NRU) Project of Thailand (No. SCI 540531 M), and Prince of Songkla University Graduate School Research funding. We also thank an Oversea Thesis Research Funding from the Faculty of Science (Year 2015), Prince of Songkla University for financial support and opportunity to conduct our research at the Royal Belgian Institute of Natural Sciences (RBINS), Brussels, Belgium. The authors thank Dr. Singtoe Boonrotpong and Entomology Research Unit (ERU) members for their highly appreciated guidance and generous help in the field. We also acknowledge the great help of Prof. Rudolf Meier, Dr Wendy Wang and Mr Darren Yeo for the sequencing of the material at the National University of Singapore.

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## Captions

Fig. 1. *Cymatopus malayensis* Parent habitus male. t1: twisted fore tarsomere 1; fb: leaflike bristle on fore tibia; mt: fields of microtrichia on wing.

Fig. 2. Cymatopus malayensis Parent habitus female.

Fig. 3. *Cymatopus mayakunae* new species male habitus. cb: curved bristles on fore tibia; ht: hind tibia with dorsal row of bristles with swollen tip; arrow indicates fold in hind margin of wing set with long bristles.

Fig. 4. Cymatopus mayakunae new species female habitus.

Fig. 5. Cymatopus mayakunae sp. nov. male terminalia.

Fig. 6. Cymatopus thaicus Grootaert & Meuffels habitus male.

Fig. 7. Cymatopus thaicus Grootaert & Meuffels habitus female.

Fig. 8. *Cymatopus thaicus* Grootaert & Meuffels male terminalia. A. Genital capsule in lateral view; B. surstyli dorsal view; C. Cercus lateral; D. Extension on sternite 6; E. Detail surstylus in lateral view with tip hypandrium and aedeagus. Scale 0.1 mm.

Fig. 9. *Cymatopus longipilus* Parent (Rayong prov.). Mid leg and swollen hind leg bearing long bristles.

Fig. 10. Evolutionary relationships of taxa of *Cymatopus*.

Fig. 11. Thambemyia pagdeni Oldroyd male habitus

Fig. 12. Thambemyia pagdeni Oldroyd female habitus.

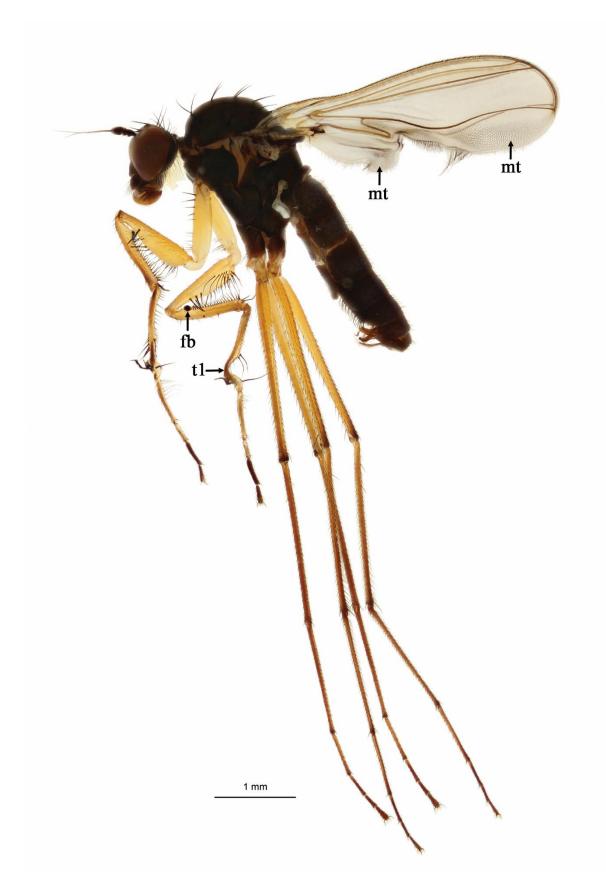


Fig. 1. Cymatopus malayensis, male habitus



Fig. 2. Cymatopus malayensis, female habitus

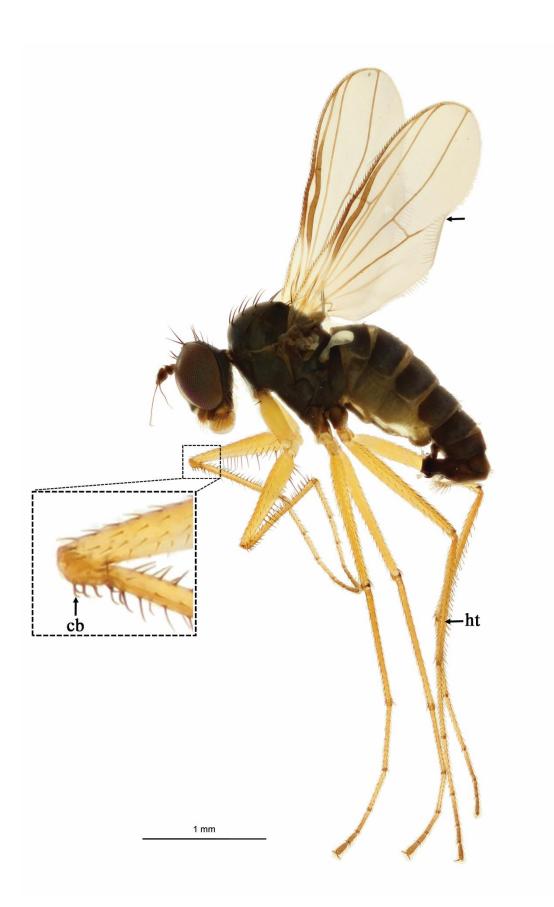


Fig. 3. Cymatopus mayakunae sp. nov., male habitus



Fig. 4. Cymatopus mayakunae sp. nov., female habitus

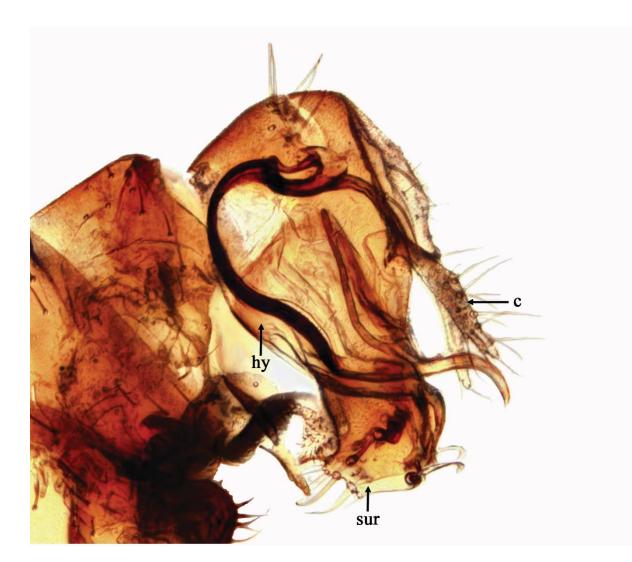


Fig. 5. Cymatopus mayakunae sp. nov. male terminalia.

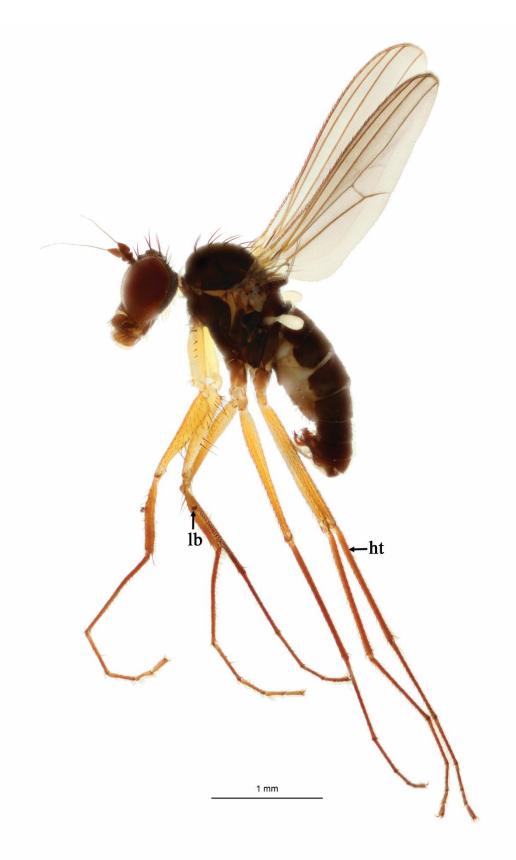


Fig. 6. Cymatopus thaicus Grootaert & Meuffels habitus male



Fig. 7. Cymatopus thaicus Grootaert & Meuffels habitus female.

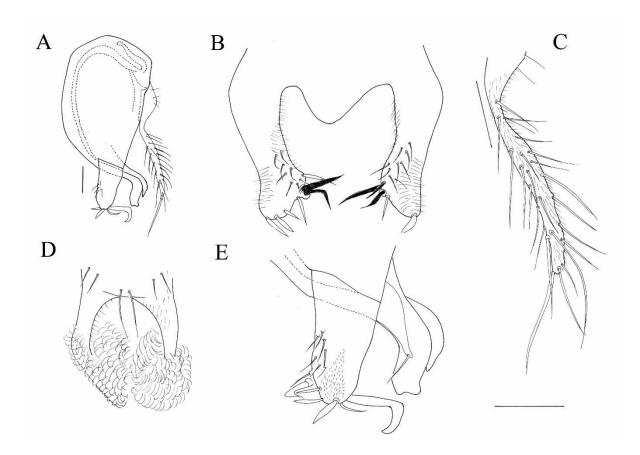


Fig. 8. *Cymatopus thaicus* Grootaert & Meuffels male terminalia. A. Genital capsule in lateral view; B. surstyli dorsal view; C. Cercus lateral; D. Extension on sternite 6; E. Detail surstylus in lateral view with tip hypandrium and aedeagus. Scale 0.1 mm.



Fig. 9. *Cymatopus longipilus* Parent (Rayong prov.). Mid leg and swollen hind leg bearing long bristles.

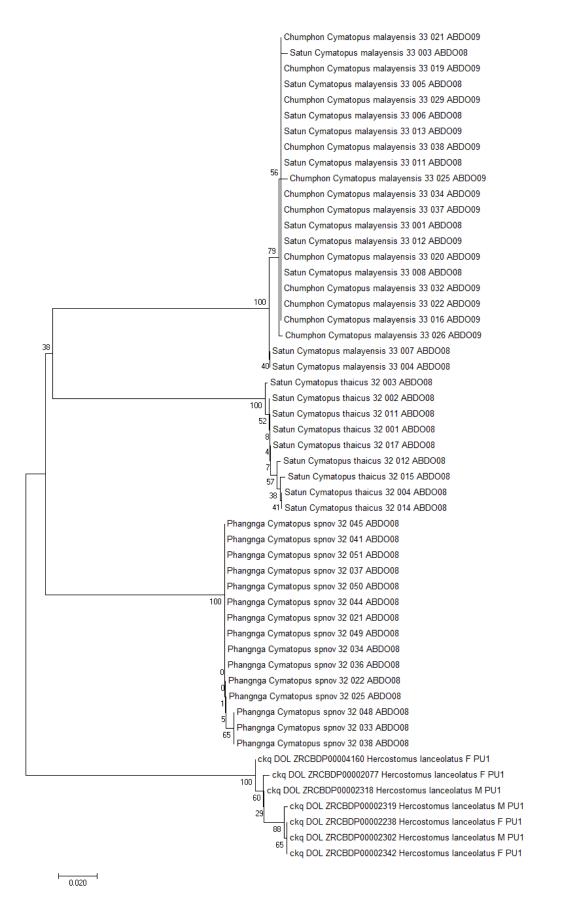


Fig. 10. Evolutionary relationships of taxa of Cymatopus.

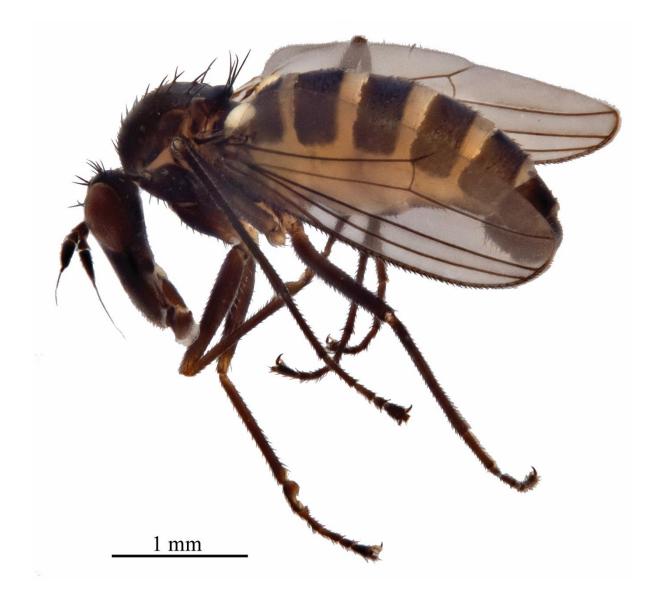


Fig. 11. Thambemyia pagdeni Oldroyd male habitus



Fig. 12. Thambemyia pagdeni Oldroyd female habitus.

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- Research Grant of the Graduate School, Prince of Songkla University.
- Oversea Research Grant of the Faculty of Science, Prince of Songkla University.

# List of Publication and Proceeding

- Samoh, A., Boonrotpong, S. and Grootaert, P. 2015. *Ngirhaphium* Evenhuis and Grootaert from southern Thailand (Diptera: Dolichopodidae) with a description of a new species. *Zootaxa*, 3946 (1): 125-132.
- Samoh, A., Satasook, C. and Grootaert, P. 2017. Eight new species of marine dolichopodid flies of *Thinophilus* Wahlberg, 1844 (Diptera: Dolichopodidae) from peninsular Thailand. *European Journal of Taxonomy*, 329: 1-40.