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Ethiopian muscids (Diptera, Muscidae) egg-carriers of *Stylogaster* Macquart (Diptera, Conopidae)

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Abstract: We here report for the first time on the presence of three species of the conopid genus *Stylogaster* Macquart (Diptera, Conopidae) in Ethiopia, viz. *S. nitens* Brunetti, *S. westwoodi* Smith and *Stylogaster* sp.. We further screened 908 muscid flies (Diptera, Muscidae) for the presence of impaled eggs of *Stylogaster* and recorded eggs on 89 individuals (9.8%). Eggs were impaled on eight species, viz. *Limnophora translucida* Stein, *Musca lusoria* Wiedemann, *Musca splendens* Pont, *Neomyia chrysopyga* (Emden), *Pseudohelina nigritarsis* (Jaennicke), *Stomoxys omega* Newstead, *Stomoxys taeniatus* Bigot and *Stomoxys varipes* (Bezzi). The maximum number of eggs found on a single muscid was six. We illustrated the dissected eggs. *L. translucida*, *M. lusoria*, *M. splendens*, *N. chrysopyga* and *S. varipes* are reported as new muscid hosts species for *Stylogaster*.

Key words: Afrotropical region, dart-eggs, Ethiopia, new record.

INTRODUCTION

Conopidae (Insecta, Diptera), also known as thick-headed-flies, and their larvae are internal parasites mainly in Hymenoptera. Females of the genus *Stylogaster* Macquart (subfamily Stylogastrinae) have a unique oviposition behaviour during which they bend the long and thin abdomen forward to 'throw' their eggs as darts against various body parts of other insects (hence their vernacular name, dart-egg-flies), especially of other brachycerous Diptera, and Orthoptera (cockroaches and crickets). The biology of the genus was discussed by, *e.g.*, Stuckenberg (1963) and Smith (1967) while Couri and Barros (2010) summarized available data on calyptrate Diptera *Stylogaster* hosts. *Stylogaster* larvae are endoparasites in Orthoptera, especially cockroaches and crickets (Skevington et al. 2010). Even though impaled eggs of *Stylogaster* so far have been recorded from 30 species of Muscidae, Stuke (2012) suggests that Calyptratae are not normal larval hosts since no larva are found internally.

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Muscid hosts are probably egg-carriers since no conopid has been reared from a muscid species (see Stuke 2017: XXV for a discussion). In Africa, the egg-darting behavior is usually associated with the presence of army ants as, when they pass, other insects fly away and become vulnerable to the darted eggs of *Stylogaster* females which hover over the army ants. It is also believed that *Stylogaster* females are attracted to dung, darting their eggs on other Diptera attracted by the faeces (Stuckenberg 1963, Couri and Pont 2006, Couri and Barros 2010).

Smith (1980) listed 14 *Stylogaster* species for the Afrotropical Region, but recently Stuke (2012) added 21 new species to the genus and also presented new faunistic records for nine previously described species, an identification key to the Afrotropical species and a checklist with a revised distribution. Of the 35 Afrotropical *Stylogaster* species, 22 are endemic to Madagascar. More recently, Stuke (2017) provided an updated catalogue of the genus *Stylogaster* and reported a total of 125 species worldwide of which 43 are found in the Afrotropical region. Only in the Neotropical region more (73) species have been recorded.

MATERIALS AND METHODS

Muscidae were sampled in February and March 2016 with insect nets and malaise traps (black, British museum type) in two locations in the highlands of SW Ethiopia, viz. the Limmu Coffee Farm (Horizon Plantations P.L.C.) and the Girmo Forest complex. Both are in the Limmu-Kosa area of the Jimma zone, ca. 50 km N of Jimma. The Limmu Coffee Farm is a shaded coffee (*Coffea arabica* Linnaeus, 1753) plantation with a cleared understory and a low density of large canopy trees, dominantly *Acacia* spp. and *Albizia schimperiana* Oliver, 1871. The Girmo Forest complex is a semi-natural montane forest where *C. arabica* grows naturally. The forest has a dense and species rich understory and canopy layer, with *Olinia rochetiana* Jussieu, 1846, *Aningeria adolfi-friederici* (Engler) Robyns and Gilbert, 1947, *Dracaena steudneri* Engler, 1895 and *Ficus* spp. as the dominant tree species. The collected muscides were stored in 70% v/v ethanol and exported to Belgium (export permit: EBI71/5065/2016). Upon arrival, they were pinned, labelled. and stored at the Plant Conservation and Population Biology unit of the University of Leuven.

Muscidae were identified by MC using keys in Emden (1951), Zielke (1971) and Zumpt (1973). The flies were screened for the presence of *Stylogaster* eggs and if present, eggs were counted and the body part(s) to which they were attached was (were) noted. In order to allow dissection of the *Stylogaster* eggs, several pinned muscid specimens were kept in a moist chamber for 24 hours. Afterwards, eggs were removed using entomological pins and macerated in a solution of 10% potassium hydroxide for 24 hours. After identification and making color photos using a Syncroscopy Auto-Montage with a Leica MZ16 optical microscope, eggs were stored in a microtube with glycerol attached to the pin of the pinned specimens (for more information on the procedure, see Couri and Barros, 2010). Eggs and adult *Stylogaster* were identified using Smith (1967) and Stucke (2012).

RESULTS AND DISCUSSION

A total number of 908 Muscidae specimens were identified and checked for the presence of *Stylogaster* eggs. *Stylogaster* eggs were found on 89 specimens (9.8 %) of eight muscid species, viz. *Limnophora translucida* Stein, 1913; *Musca lusoria* Wiedemann, 1824; *Musca splendens* Pont, 1980; *Neomyia chrysopyga* (Emden, 1939); *Pseudohelina nigritarsis* (Jaennicke, 1867); *Stomoxys omega* Newstead, 1907; *Stomoxys taeniatus* Bigot, 1888 and *Stomoxys varipes* (Bezzi, 1907). The three *Stomoxys* species accounted for 90 % of the impaled specimens (85.4 %) with *S. omega* (85.4 %) and *S. taeniatus* (9 %) showing the

highest frequences. Frequencies of other impaled muscids were very low (< 3.5 %).

Most of the impaled muscids were females (76/89 or 85.4 %) (Table I). A similar high prevalence for females was observed by Couri and Barros (2010) and Couri et al. (2013). Several *Stylogaster* eggs were found on the ventral part of the abdomen of the muscid hosts and this suggests that eggs were not darted during the flight of the hosts fleeing the army ants. More likely, *Stylogaster* females visit dung where they dart their eggs on hosts that are attracted to dung, such as *Stomoxys* spp. This might also explain the high number of *Stomoxys* female Muscidae infested with eggs compared to males. The maximum number of eggs found on a single muscid specimen was highly variable and ranged from one to six (a female of *S. omega*) (Table II).

We identified three *Stylogaster* species, *viz*. *S. nitens* Brunetti, *S. westwoodi* Smith and *Stylogaster* sp. Both *S. nitens* and *S. westwoodi* are widely distributed in the Afrotropical Region, and previously have been recorded from Angola, Democratic Republic of Congo, Ghana, South Africa, Uganda and from Democratic Republic of Congo, Kenya, Malawi, Nigeria, South Africa, Tanzania, Zambia and Zimbabwe, respectively.

Illustrations of the dissected eggs are depicted in Figs 1f and 1g. One of the eggs is probably of *S. nitens* (Fig. 1f) because adults of this species were collected along with the muscid specimens impaled with *Stylogaster* eggs. The other eggs showed high morphological resemblance with the egg of an unknown *Stylogaster* sp. figured by Smith (1967, Figs 16 and 17 therein).

In conclusion, we here record, for the first time, the presence of three species of the conopid genus *Stylogaster*, in Ethiopia, and record the muscid species *Limnophora translucida, Musca lusoria, Musca splendens, Neomyia chrysopyga* and *Stomoxys varipes* as new hosts for *Stylogaster* species.

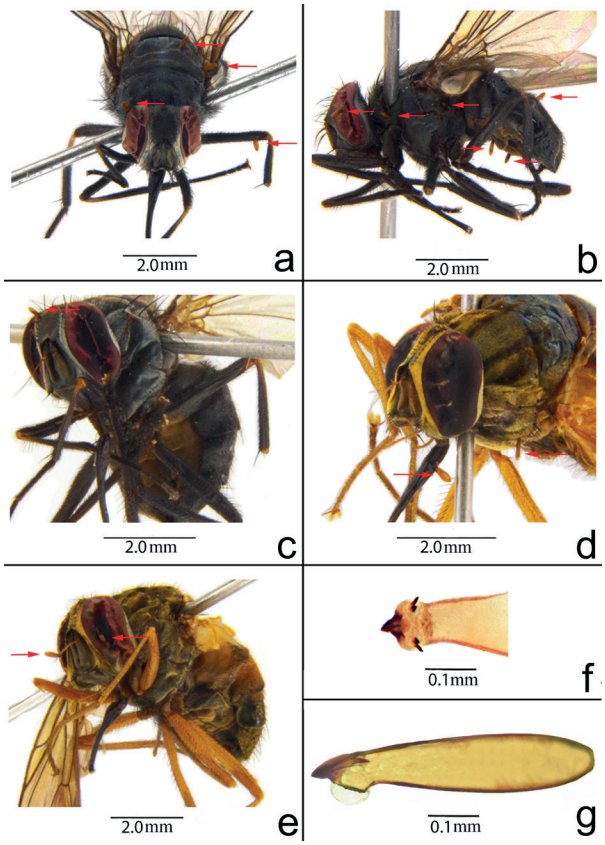

Figure 1 – **a**) Lateral view of *Stomoxys omega* Newstead (ENT000027971, adult ♂); **b**) Lateral view of *Stomoxys omega* Newstead (ENT000028668, adult ♀); **c**) Lateral view of *Stomoxys omega* Newstead (ENT000027797, adult ♀); **d**) Lateral view of *Stomoxys taeniatus* Bigot (ENT000028471, adult ♂); **e**) Lateral view of *Stomoxys taeniatus* Bigot (ENT000028756, adult ♂); **f**) Ventral view of the anterior part of the egg of *Stylogaster nitens* Brunetti; Figure **g**) Lateral view of the egg of *Stylogaster* sp.

MATERIAL EXAMINED

Muscidae

Limnophora translucida Stein - ETHIOPIA: Jimma, Girmo, 07°52'49"N, 36°51'51"E, 19.ii.2016, 1961 m, coll. L. Geeraert, Random sweepnetting, Semi-forest coffee management system, 1♂, ENT000028289.

Musca lusoria Wiedemann - ETHIOPIA: Jimma, Limmu-Kosa, 07°57'12"N, 36°52'46"E, 9.ii.2016, 1777 m; coll. L. Geeraert, Random sweepnetting, Plantation coffee management system, 1♀, ENT000027990; 1♀, ENT000027954;

TABLE I
Muscidae species as *Stylogaster* (Conopidae) hosts in Ethiopia.

Muscidae species	N° of males	N° of females	Range of no of eggs on a single host
Limnophora translucida	1	0	1
Musca lusoria	0	3	1
Musca splendens	0	1	1
Neomyia chrysopyga	0	1	1
Pseudohelina nigritarsis	3	0	1
Stomoxys omega	7	64	1–6
Stomoxys taeniatus	2	6	1–2
Stomoxys varipes	0	1	1
TOTAL	13	76	

TABLE II
Muscidae host of *Stylogaster* spp. with details on number of eggs and body part where impaled were recorded.

Muscidae species	Identification nr	Locality	Sex	Part of body impaled	Nr of eggs
Limnophora translucida Stein	ENT000028289	Girmo	♂	Tergite 5 dorsal	1
Musca lusoria Wiedemann	ENT000027990	Limmu-Kosa	♀	Back of left eye	1
Musca lusoria Wiedemann	ENT000027954	Limmu-Kosa	♀	Tergite 4 dorsal	1
Musca lusoria Wiedemann	ENT000028811	Limmu-Kosa	♀	Right anepistenum	1
Musca splendens Pont	ENT000028275	Girmo	♀	Left anepimeron	1
Neomyia chrysopyga (Emden)	ENT000027926	Limmu-Kosa	♀	Left katepisternum	1
Pseudohelina nigritarsis (Jaennicke)	ENT000028063	Girmo	♂	Tergite 4 left lateral	1
Pseudohelina nigritarsis (Jaennicke)	ENT000027757	Girmo	♂	Right eye	1
Pseudohelina nigritarsis (Jaennicke)	ENT000028393	Girmo	♂	Left eye	1
Stomoxys omega Newstead	ENT000028080	Girmo	♀	Right greater ampulla; base of right wing; right anepimerum; tergite 5 left lateral; tergite 1+2 ventral	5
Stomoxys omega Newstead	ENT000028071	Girmo	♀	Left wing insertion	1
Stomoxys omega Newstead	ENT000028052	Girmo	♀	Right anepisternum	1
Stomoxys omega Newstead	ENT000028051	Girmo	♀	Right notopleurum; left katepisterno; meron; tergite 3 ventral	4
Stomoxys omega Newstead	ENT000028050	Girmo	♀	Right notopleurum	1
Stomoxys omega Newstead	ENT000028006	Girmo	♀	Left eye	1
Stomoxys omega Newstead	ENT000028022	Girmo	♂	Right notopleurum	1
Stomoxys omega Newstead	ENT000027971	Girmo	♂	Postpronotum 1; escuto dorsal; scutum; apex of right fêmur	4
Stomoxys omega Newstead	ENT000027970	Girmo	♀	Left postpronotum; bellow the anterior spiracle; Left meron	3

TABLE II (continuation)

Muscidae species	Identification nr	Locality	Sex	Part of body impaled	Nr of eggs
Stomoxys omega Newstead	ENT000027968	Girmo	♀	Left eye	1
Stomoxys omega Newstead	ENT000027966	Girmo	♀	Right notopleurum; left anepisternum	2
Stomoxys omega Newstead	ENT000027956	Limmu-Kosa	♂	Right eye	1
Stomoxys omega Newstead	ENT000028724	Girmo	♀	Right anterior fêmur	1
Stomoxys omega Newstead	ENT000028717	Girmo	♂	Left notopleurum; left eye; tergite 3 dorsal	3
Stomoxys omega Newstead	ENT000028685	Girmo	♀	Pre-scutum, lateral left	1
Stomoxys omega Newstead	ENT000028683	Girmo	♀	Left eye; left notopleurum	2
Stomoxys omega Newstead	ENT000028682	Girmo	♀	left notopleurum	1
Stomoxys omega Newstead	ENT000028681	Girmo	♀	Left anepisternum	1
Stomoxys omega Newstead	ENT000028680	Girmo	♀	Left notopleurum; base of right wing	2
Stomoxys omega Newstead	ENT000028676	Girmo	♀	Left notopleurum; left katepisternum	2
Stomoxys omega Newstead	ENT000028675	Girmo	♀	Tergite 4 dorsal	1
Stomoxys omega Newstead	ENT000028671	Girmo	♀	Left notopleurum; left anepisternum	2
Stomoxys omega Newstead	ENT000028668	Girmo	♀	Left eye; anterior left spiracle; posterior left spiracle; tergite 3 ventral; two in tergite 4, ventral and dorsal	6
Stomoxys omega Newstead	ENT000028666	Girmo	♀	Right katepisternum	1
Stomoxys omega Newstead	ENT000028663	Girmo	♀	Left katepisternum	1
Stomoxys omega Newstead	ENT000028661	Girmo	♀	Right notopleurum	1
Stomoxys omega Newstead	ENT000028651	Girmo	♀	Right eye	1
Stomoxys omega Newstead	ENT000028649	Girmo	♀	Right eye	1
Stomoxys omega Newstead	ENT000028647	Girmo	♀	Left notopleurum; left eye	2
Stomoxys omega Newstead	ENT000028643	Girmo	♀	Right anepisternum; base of right wing	2
Stomoxys omega Newstead	ENT000028642	Girmo	♀	Right postpronotum	1
Stomoxys omega Newstead	ENT000028640	Girmo	♀	Right prescutum	1
Stomoxys omega Newstead	ENT000028637	Girmo	♀	Tergite 1+2 dorsal	1
Stomoxys omega Newstead	ENT000028633	Girmo	♀	Tergite 4 dorsal	1
Stomoxys omega Newstead	ENT000028630	Girmo	♂	Scutum; right eye	2
Stomoxys omega Newstead	ENT000028625	Girmo	♀	Left eye	1
Stomoxys omega Newstead	ENT000028620	Girmo	♀	Left postpronotum	1
Stomoxys omega Newstead	ENT000028616	Girmo	♀	Above postalar wall; base of left wing	2
Stomoxys omega Newstead	ENT000028615	Girmo	♀	Right katepisternum	1
Stomoxys omega Newstead	ENT000028606	Girmo	♀	Scutum	1
Stomoxys omega Newstead	ENT000028589	Girmo	♀	Tergite 2 dorsal	1
Stomoxys omega Newstead	ENT000028586	Girmo	♀	Left anepimerum	1
Stomoxys omega Newstead	ENT000028577	Girmo	♀	Prescutum	1
Stomoxys omega Newstead	ENT000028575	Girmo	♀	Left eye	1
Stomoxys omega Newstead	ENT000028513	Girmo	♀	Left eye; scutum	2
Stomoxys omega Newstead	ENT000028503	Girmo	♀	Tergite 1+2 right lateral	1
Stomoxys omega Newstead	ENT000028502	Girmo	♀	Tergite 1 ventral	1
Stomoxys omega Newstead	ENT000028493	Girmo	♀	Right eye	1

TABLE II (continuation)

Muscidae species	Identification nr	Locality	Sex	Part of body impaled	Nr of eggs
Stomoxys omega Newstead	ENT000028491	Girmo	♀	Right postpronotum; left katepisternum	2
Stomoxys omega Newstead	ENT000028489	Girmo	♀	Scutum	1
Stomoxys omega Newstead	ENT000028488	Girmo	♀	Right eye; left anepimerum	2
Stomoxys omega Newstead	ENT000028485	Girmo	♀	Left anepisternum	1
Stomoxys omega Newstead	ENT000028482	Girmo	♀	Left spiracle	1
Stomoxys omega Newstead	ENT000027746	Girmo	♀	Tergite 1+2 ventral; tergite 3 ventral	2
Stomoxys omega Newstead	ENT000027748	Girmo	♂	Base of left wing	1
Stomoxys omega Newstead	ENT000027763	Gima	♀	Right notopleuron	1
Stomoxys omega Newstead	ENT000027771	Girmo	♀	Left anterior spiracle; tergite 5 dorsal	2
Stomoxys omega Newstead	ENT000027773	Girmo	♀	Right eye	1
Stomoxys omega Newstead	ENT000027792	Girmo	♀	Left eye	1
Stomoxys omega Newstead	ENT000027795	Girmo	♀	Left notopleurum; tergite 1+2 ventral	2
Stomoxys omega Newstead	ENT000027797	Girmo	♀	Frons	1
Stomoxys omega Newstead	ENT000027800	Girmo	♀	Left eye; right postpronotum; left upper calypter	3
Stomoxys omega Newstead	ENT000027837	Girmo	♀	Left postpronotum; tergite 1+2 right lateral	2
Stomoxys omega Newstead	ENT000027864	Girmo	♀	Anepisternum	1
Stomoxys omega Newstead	ENT000027868	Girmo	♂	Right anepisternum	1
Stomoxys omega Newstead	ENT000027874	Girmo	♀	Prescutum; scutum	2
Stomoxys omega Newstead	ENT000027908	Girmo	♀	Postpronotum	1
Stomoxys omega Newstead	ENT000027913	Girmo	♀	Scutum; anepisternum	2
Stomoxys omega Newstead	ENT000028352	Girmo	♀	Tergite 4 dorsal	1
Stomoxys omega Newstead	ENT000028456	Girmo	♀	Right postpronotum; right notopleurum; left anepisternum; apex of anterior femus; tergite 5 dorsal	5
Stomoxys omega Newstead	ENT000028253	Girmo	♀	Right eye	1
Stomoxys taeniatus Bigot	ENT000027953	Limmu u-Kosa	♀	Left eye	1
Stomoxys taeniatus Bigot	ENT000028756	Limmu-Kosa	♂	Right eye; left eye	2
Stomoxys taeniatus Bigot	ENT000028618	Girmo	♀	Left postpronotum	1
Stomoxys taeniatus Bigot	ENT000028471	Limmu-Kosa	♂	Proboscis; right anepisternum	2
Stomoxys taeniatus Bigot	ENT000027784	Limmu-Kosa	♀	Left prescutum	1
Stomoxys taeniatus Bigot	ENT000027785	Limmu-Kosa	♀	Right eye	1
Stomoxys taeniatus Bigot	ENT000027873	Girmo	♀	Right anepisternum	1
Stomoxys taeniatus Bigot	ENT000028317	Limmu-Kosa	♀	Tergite 4 dorsal	1
Stomoxys varipes (Bezzi)	ENT000028547	Girmo	♀	Tergite 5 dorsal	1

07°55’57”N, 36°51’57”E, 4.II.2016, 1786 m, 1♀, ENT000028811.

Musca splendens Pont - ETHIOPIA: Jimma, Girmo, 07°52’49”N, 36°51’51”E, 19.ii.2016, 1961 m, coll. L. Geeraert, Random sweepnetting, Semi-forest coffee management system, 1♀, ENT000028275.

Neomyia chrysopyga (Emden) - ETHIOPIA: Jimma, Limmu-Kosa, 07°57’12”N, 36°52’46”E, 9.ii.2016, 1777 m, coll. L. Geeraert, Random

sweepnetting, Plantation coffee management system, 1♀, ENT000027926.

Pseudohelina nigritarsis (Jaennicke) - ETHIOPIA: Jimma, Girmo, 07°52'49"N, 36°51'51"E, 19.ii.2016, 1961 m, coll. L. Geeraert, Random sweepnetting, Semi-forest coffee management system, 1♂, ENT000028063; 1♂, ENT000027757; 1♂, ENT000028393.

Stomoxys omega Newstead (Figs 1a-c) - ETHIOPIA: Jimma, Girmo, 07°52'51"N, 36°51'47"E, 24.ii.2016, 1920 m, coll. L. Geeraert, Random sweep netting, Semi-forest coffee management system, 2♀, ENT000027746, ENT000027748; 4♀ ENT000027792, ENT000027795, ENT000027797, ENT000027800 ; 11:57, 2♀ ENT000027864, ENT000027874; 1♂ ENT000027868; 3♀, ENT000027970, ENT000027968, ENT000027966, 1♂, ENT000027971; 1♀, ENT000028006; 1♂, ENT000028022; 11.ii.2016, 1♀ ENT000028352; 1♀ ENT000028456; 1♀, ENT000028724, 1♂, ENT000028717; 2♀, ENT000028577, ENT000028575; 21♀, ENT000028685, ENT000028683, ENT000028682, ENT000028681, ENT000028680, ENT000028676, ENT000028675, ENT000028671, ENT000028668, ENT000028666, ENT000028663, ENT000028661, ENT000028651, ENT000028649, ENT000028647, ENT000028643, ENT000028642, ENT000028640, ENT000028637, ENT000028633, ENT000028625, 1♂, ENT000028630; 13:55, 6♀, ENT000028620, ENT000028616, ENT000028615, ENT000028606, ENT000028589; ENT000028586; 11:30, 9♀, ENT000028513, ENT000028503, ENT000028502, ENT000028493, ENT000028491, ENT000028489, ENT000028488, ENT000028485, ENT000028482; 07°52'49"N, 36°51'51"E, 19.ii.2016, 1961 m, Plot 2, Dani, 10:50, 1♀ ENT000027837; 11:30, 3♀ ENT000027763, ENT000027771, ENT000027773; 12:35, 2♀ ENT000027908, ENT000027913; Siel, 12:15, 1♀, ENT000028080; 11:30, 4♀,ENT000028071, ENT000028052, ENT000028051, ENT000028050; 10:50 1♀ ENT000028253; Limmu-Kosa, 07°57'12"N, 36°52'46"E, 9.ii.2016, 1777 m, coll. L. Geeraert, 10:06, 1♂, ENT000027956.

Stomoxys taeniatus Bigot (Figs 1d-e) - ETHIOPIA: Jimma, Limmu-Kosa, 07°57'12"N, 36°52'46"E, 9.ii.2016, 1777 m, coll. L. Geeraert, Random sweepnetting, Plantation coffee management system, 1♀, ENT000027953; 07°55'57"N, 36°51'57"E, 4.ii.2016, 1786 m,, 1♂, ENT000028756; 07°55'18"N, 36°52'14"E, 10.ii.2016, 1908 m, 1♂, ENT000028471; 1♀, ENT000028317; 05.ii.2016, 2♀, ENT000027784, ENT000027785; Girmo, 07°52'51"N, 36°51'47"E, 11.ii.2016, 1920 m, coll. L. Geeraert, 1♀, ENT000028618; 07°52'51"N, 36°51'47"E, 24.ii.2016, 1920 m, coll. L. Geeraert, 1♀, ENT000027873.

Stomoxys varipes (Bezzi) - ETHIOPIA: Jimma, Girmo, 07°52'51"N, 36°51'47"E, 11.ii.2016, 1920 m, coll. L. Geeraert, Random sweepnetting, Semi-forest coffee management system, 1♀, ENT000028547.

Conopidae

Stylogaster nitens Brunneti: ETHIOPIA: Jima, Girmo, 07°52'49''N, 36°51'51''E, 19-24.ii.2016, 1961 m, coll. L. Geeraert, Malaise trap, semi-forest coffee management system; Plot 2, 3 ♂; 24-29. ii.2016, 1 ♂.

Stylogaster westwoodi Smith - ETHIOPIA: Jimma, Limmu-Kosa, 07°57'12"N, 6°52'46"E, 03-05.II.2016, 1777 m, coll. L. Geeraert, Malaise trap; Plantation coffee management system; Plot 1, 2♂; 05-09.ii.2016, 1♂; 11-16.ii.2016, 8♂, 1♀; 23-23. ii.2016, 3♂.

Stylogaster sp.; ETHIOPIA: Jimma, Lima-Kosa, 07°57'12''N, 36°52'46'E, 11-16.iii.2016, 1777m, coll. L. Geeraert, Malaise trap, Plantation coffee management system; Plot 1, 1 ♀.

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AUTHOR CONTRIBUTIONS

MSC identified the material and wrote the manuscript; KJ and LG collected the material and wrote the manuscript; RML and APVA identified the material and prepared the photos and the table; KJ revised the manuscript; all authors approved the final version of the manuscript.

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