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Case Study

First record of *Megaselia scalaris* (LOEW) (DIPTERA: PHORIDAE) infesting a spinose ear tick, *Otobius megnini*, colony in Sri Lanka

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Abstract. *Megaselia scalaris* (Loew) is a cosmopolitan polyphagous small fly with the ability of exploiting variety of ecological niches. Different life history stages act as detritivore, parasite, and parasitoid of wider spectrum of plant and animal matter under natural and laboratory conditions. Here, for the first time we present the opportunistic parasitism of *M. scalaris* on *Otobius megnini*, which act as a vector of Q fever and is capable of causing paralysis, toxic conditions, otoacariasis and otitis in humans and other animals. Tick samples from the ear canals of 14 thoroughbred horses were brought to the laboratory and several days later, larvae of *M. scalaris* were found feeding on immature stages of *O. megnini*. When the development was completed pupae were found attached to adult ticks and all nymphs were found dead. This context reveals the capability of *M. scalaris* surviving on *O. megnini* and the risk of their invading ear canals of horses.

INTRODUCTION

Megaselia scalaris (Diptera: Phoridae) is a cosmopolitan fly (2 to 3 mm) with medical, forensic and veterinary importance. They are also known as scuttle flies or hump-backed flies due to their erratic movement on surfaces and morphological features of the thorax respectively (Sukontason *et al.*, 2006; Boonchu *et al.*, 2004). These flies are capable of exploiting diverse ecological niches in tropics and subtropics (Smith, 1986; Costa *et al.*, 2007). *Megaselia scalaris* has adapted to polyphagous life style, feeding and breeding in wider spectrum of plant and animal matter (Karunaweera *et al.*, 2002; Costa *et al.*, 2007; Disney, 2008). Flies are attracted to putrid odors and lay eggs on decaying organic matter. The larva (maggot) undergoes two molts leading to three larval stages. Saprophagous, sarcophagous and necrophagous feeding, as well as parasitic, behaviors of *M. scalaris* larvae are well recognized (Costa *et al.*, 2007, Koch *et al.*, 2013) as they actively infest laboratory colonies of ticks (Barr'e *et al.*, 1991), hemipterans (*Triatoma brasiliensis*) (Costa *et al.*, 2007) and mantid colonies (Koch *et al.*, 2013), cause internal and open wound myiasis (infection cause by developing fly larvae) in humans (Huntington *et al.*, 2008) and other animals (Valin *et al.*, 2013), infest frog eggs (Brown *et al.*, 2012) and are a potential threat to banana (*Musa accuminata*) cultivations (Karunaweera *et al.*, 2002). Furthermore, due to the relative small size of these flies, *M. scalaris* is found breeding in tightly sealed corpse and may play a significant role in estimating minimum post-mortem interval in forensic investigations (Sukontason *et al*., 2006).

The spinose ear tick, *Otobius megnini* (Dugès) (Acari: Ixodida: Argasidae) is an economically important soft tick that parasitizes the ear canal of domesticated animals and occasionally humans causing otoacariasis (Diyes *et al*., 2014). They can cause paralysis, irritations, toxic conditions, allergies, eardrum perforation and muscle spasms; apart from that they cause severe otitis in livestock and humans due to deep ear feeding and act as vector of Q fever (*Coxiella burnetii*) in nature (Jellison *et al*., 1948; Estrada-Peña *et al*., 2010). *Otobius megnini* has one host life cycle, which is characterized by the larvae and the variable number of nymphal instars feeding on a single host. Non-feeding adults live in borrows (Nava *et al*., 2009) and mate off the host and lay separate egg batches.

Unlike hard ticks (Acari: Ixodida: Ixodidae), chemical control of soft ticks is challenging because they survive in burrows or cracks and crevices. Moreover the use of acaricides (e.g., carbamate, organophosphate, synthetic pyrethroid, formamidine, macrocyclic lactone and pyrazole) (Lovis *et al*., 2011) associated with many problems such as ticks can acquire resistance, harm host animal and their products (e.g., milk, meat and hind) and lack of environmental safety (Graf *et al*., 2004). This increases the attention towards the alternative and more sustainable tick control methods as immunization of hosts and biological tick control (Merino *et al*., 2013).

Few organisms have been recognized as biological control agents for selected tick species. Examples include some bacteria, protozoa, fungi and arthropods (Samish *et al*., 2004) such as *Ixodiphagus* sp., *Solenopsis* sp., *Pogonomyrmex* sp., *Iridomyrmex* sp. *Aphaenogaster* sp. and *Monomorium* sp. which belongs to the order Hymenoptera, some beetles in Carabidae and dipterans such as *Megaselia scalaris* and *Megaselia rufipe* (Meigen) (Diptera: Phoridae) have been identified successfully infesting hard and soft ticks in different parts of the world (Mwangi *et al*., 1997; Samish and Alekseev, 2001; Samish *et al*., 2004). This case report presents the parasitic behavior of *M. scalaris* on *O. megnini* for the first time in Sri Lanka.

CASE REPORT

Stables in Nuwara Eliya race course (GPS: 6.962829 N, 80.769207 E) were routinely visited since 2013 to collect ticks infesting ear canals of horses for a seasonality and life cycle study. Ticks were collected from both left and right ear canals of 14 thoroughbred horses (10 males and 4 females) by using small pieces of white open wove cotton bandages. The ticks and associated contents of the ear (e.g., ear wax, dust and dirt) were transferred into a small 100 ml plastic vial. Live ticks were brought to the Parasitology Laboratory, Department of Zoology, University of Peradeniya and were kept under laboratory conditions (28ºC, 80% RH) until they molted into next stage of their development. The vials were capped with perforated lids and occasionally (once in every two days for 15 minutes) opened to facilitate ventilation and to minimize the putrid odor caused by the ear wax.

Ticks that were brought to the laboratory were identified as immature *Otobius megnini* (Figure 1). Several days after collection, we noted some were dead. Upon closer examination, dipteran larvae were observed feeding on ticks. A sample of the larvae were collected and preserved in 70% alcohol. They were subsequently identified as larvae of *M. scalaris* using standard keys (Smith, 1986; Liu and Greenberg, 1989; Disney, 2008). Remaining larvae in the vials with the dead ticks were allowed to complete development to the adult stage approximately 15 days after first being observed. Emergent adults were preserved in absolute alcohol. Unfortunately, some of the adults escaped from the plastic vials due to their extraordinary ability of escaping seemingly closed containers (Disney, 2008). Therefore, the number of adults to successfully emerge was based on empty pupual cases in the vials.

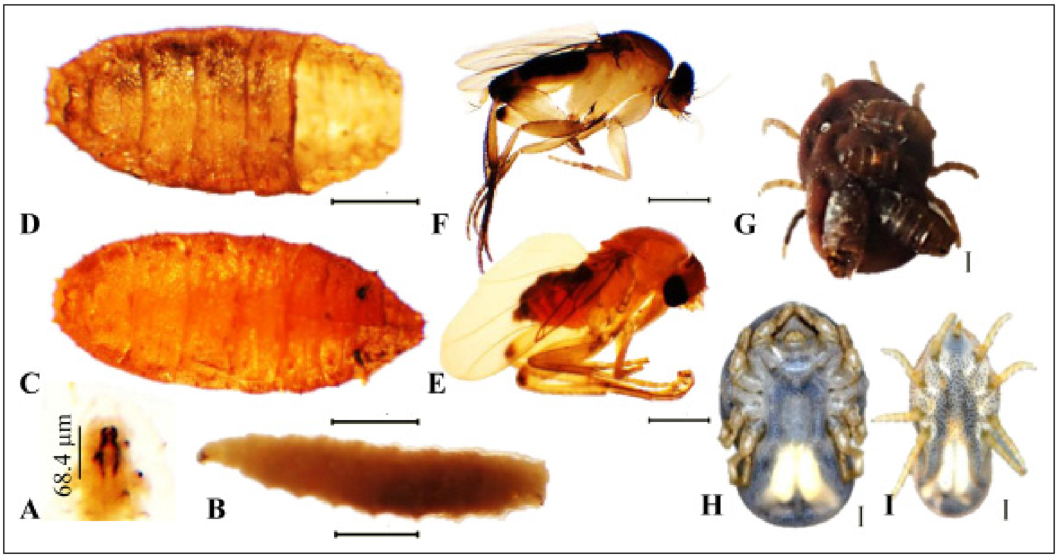

Figure 1. Different life history stages of *Magaselia scalaris* and *Otobius megnini*. (A) Cephalopharyngeal structure of second instar larva, (B) second instar larvae, (C) Pupa, (D) Open pupal case, (E) Female imago and (F) Male imago of *M.scalaris*. (G) *O. megnini* adult female with several pupa attached on dorsal side (H) Ventral side of a healthy *O. megnini* female (I) *O. megnini* nymph. Scale bars represent length of 1 mm.

Together with remains of dead tick nymphs and adults, counts (molted individuals) were considered as the initial number of ticks present. Second instar *M. scalaris* were found feeding on ticks during the first observation indicating a significant amount of time had elapsed since colonization (Figure 1). Pupae (n= 52) were found attached to the bottom walls of the container as well as to adult ticks (Figure 1). The number of imagos remained in vials after the eclosion were 11 (9 females and 2 males) and the total adults were expected as 52 (Figure 1). Fourteen of the 20 adult ticks were found dead at the time of initial *M. scalaris* infestation. The adult ticks remaining alive were lethargic and appeared to have difficulty moving. The initial number of nymphs was predicted as 41 but the tick larvae were not found even though they were observed during sampling.

DISCUSSION

According to previous observations it is less evident that the *M. scalaris* were directly attracted to *O. megnini* even though the predatory behavior of this phorid fly has previously documented in other countries (Barr'e *et al.*, 1991; Miranda *et al.*, 2011). We speculate that adult flies were attracted to the putrid odor of the ear wax of horses and laid eggs with the resulting larvae preying upon the ticks. Nevertheless, the geographic distribution and plasticity of feeding and breeding sources (Costa *et al.*, 2007) of *M. scalaris* and the ability to penetrate small apparently sealed spaces may have allowed the opportunistic-parasitoid behavior on *O. megnini*. If the adult females are attracted to the odor of ear wax of horses in Nuwara Eliya, the situation may increase the risk of *M. scalaris* infesting the ear canal of horses.

Prior to this study Garris (1983) and Barr'e *et al.* (1991) reported *M. scalaris* infesting laboratory colonies of *Amblyomma variegatum* (Acari: Ixodida: Ixodidae) whereas Andreotti *et al.* (2003) and Miranda *et al.* (2011) recognized larvae of this phorid feeding on engorged females of *R.* (*Boophilus*) *microplus* (Acari: Ixodida: Ixodidae). However, the information on *M. scalaris* infesting *O. megnini* or any other soft ticks is scant, yet other insects, such as the ants (Hymenoptera: Formicidae)

Monomorium minimum, *Pogonomyrmex barbatus var. molefaciens* have identified feeding on this particular soft tick (Samish and Alekseev, 2001). Other than Arthropods like *M. scalaris*, many authors have observed bacterial species such as *Rickettsia* sp., *Cedecealapagei* and *Proteus mirabilis* (which is pathogenic to, *Demacentor andersoni*, *Amblyomma hebraeum*, *Hyalomma marginatum*), protozoans such as *Babesia bigemina* (pathogenic to *Rhipicephalus microplus*) and fungi including *Metarhizium anisopliae* and *Beauveria bassiana* (pathogenic to *R. microplus*, *Rhipicephalus appendiculatus*) infesting some hard tick species (Kaaya *et al.*, 1996; Samish *et al.*, 1999). Despite the fact that many different groups have been documented infesting ticks under laboratory conditions, the parasitic nature of these organisms under natural conditions is still to be recognized (Andreotti *et al.*, 2003). In Sri Lanka, Karunaweera *et al.* (2002) described the capability of *M. scalaris* feeding on flesh of ripe bananas under natural conditions and the possibility of a potential health risk of intestinal myiasis.

Some researchers suggest that by keeping tick colonies under regular inspection with eliminating dead individuals from the rest of the colony and keeping less crowding colonies under less moist conditions (Costa *et al.*, 2007) could minimize the infestations of *M. scalaris*. This context reveals the possibility of surviving *M. scalaris* on laboratory colonies of *O. megnini* and the risk of invading the ear canal of horses while being a possible candidate for biological control of *O. megnini*.

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