

Prevalence of larvae of the bot fly *Cuterebra simulans* (Diptera, Oestridae) on *Gracilinanus microtarsus* (Didelphimorphia, Didelphidae) in southeastern Cerrado from Brazil

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ABSTRACT. Prevalence of larvae of the bot fly *Cuterebra simulans* (Diptera, Oestridae) on *Gracilinanus microtarsus* (Didelphimorphia, Didelphidae) in southeastern Cerrado from Brazil. The prevalence of *Cuterebra simulans* bot fly parasitism on marsupial *Gracilinanus microtarsus* was compared between sexes and seasons of the year. No significant difference was observed between females and males but prevalence was higher in warm-wet season than cool-dry season. This pattern agree with observations of others studies with oestrids flies in southern Neotropical areas, suggesting the occurrence of latitudinal change in the peak of bot flies reproductive activity related to seasons of the year along these systems. This is the first record of *C. simulans* larvae parasitism on *G. microtarsus* and its occurrence in southern areas of the Neotropical region.

KEYWORDS. Bot fly; myiasis; opossum; parasitism.

RESUMO. Prevalência de parasitismo de larvas de *Cuterebra simulans* (Diptera, Oestridae) em *Gracilinanus microtarsus* (Didelphimorphia, Didelphidae) no Cerrado do sudeste do Brasil. A prevalência de parasitismo das larvas da mosca *Cuterebra simulans* no marsupial *Gracilinanus microtarsus* foi comparada entre os sexos e estações do ano. Machos e fêmeas não apresentaram diferenças significativas, mas a prevalência de parasitismo foi mais elevada na estação quente-úmida que na estação fria-seca. Este padrão está de acordo com as observações de outros estudos com moscas da família Oestridae realizados nas áreas ao sul da região Neotropical, sugerindo a ocorrência de mudanças latitudinais nos picos de atividades reprodutivas destas moscas em relação às estações do ano ao longo destes sistemas. Este também é o primeiro registro da ocorrência de *C. simulans* em áreas mais ao sul da região Neotropical e do parasitismo de suas larvas em *G. microtarsus*.

PALAVRAS-CHAVE. Mífase; marsupial; parasitismo; mosca do berne.

Bot flies of the genus *Cuterebra* are restricted to the New World and are distributed throughout most of the temperate and tropical areas. Their larvae are obligatory parasites of mammals, producing myiasis in dermic and subdermic tissues of these animals (Catts 1982; Pape 2001). There is almost 70 species of *Cuterebra* and these flies are one of the most highly studied groups of insect parasites of New World wild mammals (Slansky 2007). However, the knowledge available on the ecology and natural history of most species comes from studies carried out in temperate regions (e.g. Wolf & Batzli 2001; Alcock & Kemp 2004; Jaffe *et al.* 2005). In the Neotropical region, the studies concerning the prevalence and dynamics of bot fly larval parasitism in wild mammals are scarce and have been carried out mainly with rodents. These studies have showed higher prevalence of bot fly larval parasitism in warm-wet season of the year, although few longitudinal studies

demonstrate that such flies may be multivoltine (Bergallo *et al.* 2000; Adler *et al.* 2003).

Marsupials are also natural hosts of bot fly larvae and are distributed widely in New World tropics with most species found in South America, principally represented by Didelphimorphia order, or opossums, with approximately 93 species (Eisenberg & Redford 1999). Nevertheless, there are only a few studies and anecdotal records on the prevalence and dynamics of bot fly larval parasitism in marsupials (Fonseca 1941; Guimarães 1971; Bossi & Bergallo 1992; Pujol-Luz *et al.* 2004). The gracile mouse opossum *Gracilinanus microtarsus* (Wagner, 1842) is a small (20-45 g), solitary, arboreal, nocturnal, and insectivorous marsupial commonly found in the Atlantic rainforest and forested physiognomies interspersed in savannas in the Cerrado biome. This species shows sexual size dimorphism with adult males being larger than adult

females, and has a markedly seasonal pattern of reproduction occurring in the warm-wet season when males competing for access to females (Costa *et al.* 2003; Martins *et al.* 2006).

Males and females may differ in prevalence of parasites due to fundamental biological differences between sexes. For example, in reproduction period males may be engaged in sexual competition for mating opportunities, which is under control of hormones that negatively affect the immune system and increase the vulnerability to parasites (Møller *et al.* 1999). On the other hand, females may increase their home range areas to find more food due to increase energetic requirements associated with pregnancy and lactation that may increase their probabilities of being infested for parasites (Krasnov *et al.* 2005) or they may restricted their activity areas near to nests that may increase the chance of the mobile parasites found them.

Since the studies in the Neotropical have showed high seasonality in the prevalence of parasitism by *Cuterebra* larvae and that biological differences between hosts sexes may affect parasitism, the aim of our study was to investigate the differential prevalence of the bot flies larvae parasitism between sexes and seasons of the year on *G. microtarsus*.

This study was carried out at the Reserva Biológica de Mogi Guaçu (22° 15' / 22° 18' S, 47° 08' / 47° 13' W), located in the city of Mogi Guaçu, São Paulo state, southeastern Brazil. Vegetation consists of Cerrado, which is a tropical savannah formation comprising different vegetation physiognomies that differ in the density and composition of plants of the woody layer and the ground layer, forming a continuum from open and dry grassland to dense forest (Oliveira-Filho & Ratter 2002; Silva & Bates 2002). The climate of the region has two well-defined seasons: a warm-wet season from October to March when the temperature varies of 16.8°C to 36.7°C and mean rainfall is 187.7 mm, and a cool-dry season from April to September when the temperature varies of 5.2°C to 31.3°C and mean rainfall is 46.2 mm.

The individuals of the *G. microtarsus* were trapped monthly from September 2005 to August 2006, over 10 consecutive nights each month and marked with a numbered ear tag and their sex was recorded. We set an 11 x 11 trapping grid with 121 trapping-stations located 15 m from each other. We visually examined all the individuals to record the occurrence or absence of the bot flies larvae parasitism and the number of the larvae in each host. Some parasitized individuals were kept at the laboratory to allow natural emergence of the bot flies larvae. After emergence the pupae were maintained buried in moist sand in gauzed-covered glass cups at ambient temperature until adult flies emerged (pupariation lasted 30 to 32 days). Three adult bot flies emerged and they were identified as *Cuterebra simulans* (Austen, 1933). The prevalence of bot flies larvae parasitism of the *C. simulans* was calculated in agreement to Bush *et al.* (1997) and the prevalence between sex and seasons were compared with binomial test for unequal proportions at the level 0.05 of the significance (Zar 1998).

We captured 78 individuals of the *G. microtarsus* (38 females and 40 males). The prevalence of bot flies larvae



Fig. 1. Map of *Cuterebra simulans* distribution in South America. Grey areas represent well-known distributions (Peru; A – Paramaribo, Suriname; B – Belém, Brazil) according to Guimarães & Papavero (1999). Black area represents the new record of occurrence (C – Mogi Guaçu, Brazil).

parasitism in females was 0.32 and 0.28 in males but the difference between sexes was not significant ($Z = 0.395$; $p = 0.348$). The number of the bot flies larvae in each host varied of 1 to 5. Some individuals showed repeated infestations by larvae of the *C. simulans* in different months of the same season and in different season after the emergence of the bot flies larvae previously recorded, but these events were considered as independent events to estimation of the prevalence in each season. Thus, the prevalence in each season was estimated by the number of times that one individual became infested for parasites. In all study, ninety-four events of *G. microtarsus* examination (62 in warm-wet season and 32 in cool-dry season) occurred. The prevalence of bot flies larvae parasitism in warm-wet season was 0.34 and 0.06 in cool-dry season and the difference between season was significant ($Z = 2.952$; $p = 0.0016$).

Some studies carried out in Neotropical systems have showed a sex-biased larvae parasitism of the oestrids bot flies in rodents with males more infested than females (Bergallo *et al.* 2000; Manrique-Saide *et al.* 2000), females more infested than males (Brigada *et al.* 1992) or no sex-biased bot flies larvae

infestation (Adler *et al.* 2003) and these patterns always are discussed in terms of the biological sexual differences of the hosts. Our results did not show differences in the parasitism prevalence of the *C. simulans* larvae between males and females of the *G. microtarsus*, although males and females showed differences in movement patterns in study area where males showed larger home ranges than females (Fernandes 2007) and distinct signals of the high level stress in the reproduction season (*e.g.* fur fall and high level of aggressiveness) being theoretically more susceptible to bot flies larvae parasitism. Therefore, other factors related to the biology of the oestrids bot flies may explain our results. First, female bot flies oviposit on habitat substrates close to the host higher activity areas (*e.g.*, near nests, burrows or tracks) (Catts 1982) and this oviposition behavior may provide equal chances for parasitism of both sexes. Second, flies of the Oestrid family undergo long periods of association with their natural hosts and have evolved a complexity of the mechanisms by which larvae down regulate host immune system (Otranto 2001; Stevens *et al.* 2006; Slansky 2007). Thus, although males were more susceptible to parasites than females because they were under control of hormones that negatively affect the immune system, *C. simulans* larvae may have ability to infest equally males and females of the *G. microtarsus*.

The neotropical oestrids bot flies seem to be multivoltine, but the season of the peak of reproductive activity varies widely between the regions. In the northern Neotropical, the highest bot flies larvae parasitism prevalence occurs in dry season (Milton 1996; Manrique-Saide *et al.* 2000; Adler *et al.* 2003) whereas in southern areas the highest bot flies larvae parasitism prevalence occur in wet seasons (Brigada *et al.* 1992). In Cerrado of the Central Brazil, the peak of prevalence of bot flies larvae parasitism appears to be not related to seasonal changes (Vieira 1993) whereas in Atlantic rainforest the highest bot flies larvae parasitism prevalence occurs on warm-wet season (Bossi and Bergallo 1992; Bergallo *et al.* 2000; Bossi *et al.* 2002). Although our study has been carried out in Cerrado, the prevalence was higher in the warm-wet season, coinciding with others areas of the southeastern of Brazil and with others southern Neotropical areas. Thus, seem to occur a latitudinal change in the peak of bot flies reproductive activity related to the seasons along these systems but more studies are necessary to reinforce or to refute this hypothesis.

The biology of the *C. simulans* is unknown. The only knowledge about this fly species was restricted to its description, type-localities and respective original hosts. The records of its distribution was restricted to Peru, Suriname, and Pará state in Brazil (Fig. 1) and its larvae were recorded as parasites of the opossums *Caluromys philander* (Linnaeus, 1758) and *Didelphis marsupialis* (Linnaeus, 1758) (Guimarães and Papavero 1999). This present study suggests an expansion of *C. simulans* distribution for areas in the south of Brazil and Neotropical region and shows the first record of *G. microtarsus* as host of oestrid fly larvae.

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