



Morphology and DNA sequences confirm the first Neotropical record for the Holarctic sepsid species *Themira leachi* (Meigen) (Diptera: Sepsidae)

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Even for the most cosmopolitan of species, climate frequently presents effective barriers for dispersal. For example, many eurytopic and synanthropic species go extinct when introduced into a new climatic zone, and translocated ants remain in sheltered environments reminiscent of their home climate (McGlynn, 1999). Here we report the occurrence of a primarily Holarctic dipteran species, *Themira leachi* (Meigen), in Neotropical Cuba. This discovery suggests that the species may have a large disjunct distribution, as the next closest record lies almost 3,500 km to the north in Nearctic Newfoundland, Canada (Ozerov, 1998).

The genus *Themira* comprises 35 species and belongs to the relatively small clade of the cosmopolitan dung-fly family Sepsidae (Ozerov, 2005). The genus is primarily distributed in the Holarctic, with only four species bordering on other biogeographic regions (Ozerov, 1998; Pont and Meier, 2002; Meier, 2007). *Themira leachi* has been recorded throughout Northern Europe, spanning eastwards through Asiatic Russia and Mongolia. Ozerov (1998) added the species to the Nearctic fauna by reporting specimens from Northern Canada. Recently, five specimens (four males, one female) were collected from dung in Cuba (2002: Pinares de Mayari pine forest, Sierra Cristal National Park, ca. 650m ASL). The morphology of the males suggested that they are *Themira leachi*, but since this record is so far outside the known range of the species, we used detailed morphological study and DNA sequencing to confirm this preliminary identification. Line drawings were prepared for the Cuban specimens in order to compare them to drawings for European specimens. In addition, we generated high-resolution color-photographs of the habitus and important diagnostic structures for European and Cuban specimens with a Visionary Digital™ Plus Lab System, using a Canon EOS 40D with a mounted Infinite K2 Long Distance Microscope (CF4 objective at position 1 and 3). For some images, a 10X Olympus objective was used (position 3).

Detailed morphological investigations reveal that the Cuban specimens are indeed very similar to specimens from Europe and consistent with Ozerov's (1998) and Meier and Pont's (2002) diagnoses. Forelegs, sternites and hypopygia were used for comparison; Cuban and European specimens are shown in Plate 1. The fore femora and tibiae of both specimens possess similar modifications whose function and co-evolution with female wings have been discussed in the recent literature [femur: *c.f.* B & H (anterior view), C & I (posterior view); tibia: *c.f.* D & J; Ang *et. al.*, 2008; Ingram *et. al.*, 2008; Puniamoorthy *et. al.*, 2008]. The 4th sternite and hypopygium can be seen on the abdomen and are also very similar in structure and diagnostic for *T. leachi* (lateral view: *c.f.* E & K; ventral view *c.f.* F & L). Even more striking are the 2nd and 3rd sternites, which are well developed and have been modified into a raised, anteriorly open crater on the 2nd sternite and a pronounced protrusion on the 3rd sternite. These sternite modifications are unique to *T. leachi* and the only difference between the specimens is minor (a more pronounced and hook-like ventral protrusion on the 4th sternite of the European specimen). Overall, the foreleg, sternite, and hypopygium morphology are very similar between the European and Cuban specimens of *T. leachi* and suggest the presence of only one species.

Recent studies of morphologically uniform species with wide distributions have suggested that such species frequently contain 'cryptic' species that can be discovered once DNA sequence data become available (Bickford *et. al.*, 2007). We thus compared the Cuban and European specimens with regard to the mitochondrial gene *COI*. A ca. 660 bp piece of the *COI* gene was sequenced for four Cuban specimens using the DNA extraction, amplification, and sequencing protocols described in Su *et. al.* (2008). These sequences were submitted to Genbank (EU831274 – EU831277) and compared to a known sequence of *T. leachi* from Europe (Genbank: EU435823) as well as *COI* sequences for ten other *Themira* species (Su *et. al.*, 2008). Pairwise distances between the European and Cuban sequences were 0.5% to 0.8%. Whether such distances are typical for inter- or intraspecific variability can be judged when they are compared to a distri-