Article

DNA barcodes reveal cryptic genetic diversity within the blackfly subgenus *Trichodagmia* Enderlein (Diptera: Simuliidae: *Simulium*) and related taxa in the New World

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Abstract

In this paper we investigate the utility of the COI DNA barcoding region for species identification and for revealing hidden diversity within the subgenus *Trichodagmia* and related taxa in the New World. In total, 24 morphospecies within the current expanded taxonomic concept of *Trichodagmia* were analyzed. Three species in the subgenus *Aspathia* and 10 species in the subgenus *Simulium s.str.* were also included in the analysis because of their putative phylogenetic relationship with *Trichodagmia*. In the Neighbour Joining analysis tree (NJ) derived from the DNA barcodes most of the specimens grouped together according to species or species groups as recognized by other morphotaxonomic studies. The interspecific genetic divergence averaged 11.2% (range 2.8–19.5%), whereas intraspecific genetic divergence within morphologically distinct species averaged 0.5% (range 0–1.2%). Higher values of genetic divergence (3.2–3.7%) in species complexes suggest the presence of cryptic diversity. The existence of well defined groups within *S. piperi*, *S. duodenicornium*, *S. canadense* and *S. rostratum* indicate the possible presence of cryptic species within these taxa. Also, the suspected presence of a sibling species in *S. tarsatum* and *S. paynei* is supported. DNA barcodes also showed that specimens from species that were taxonomically difficult to delimit such as *S. hippovorum*, *S. rubrithorax*, *S. paynei,* and other related taxa (*S. solarii*), grouped together in the NJ analysis, confirming the validity of their species status. The recovery of partial barcodes from specimens in collections was time consuming and PCR success was low from specimens more than 10 years old. However, when a sequence was obtained, it provided good resolution for species identification. Larvae preserved in 'weak' Carnoy's solution (9:1 ethanol:acetic acid) provided full DNA barcodes. Adding legs directly to the PCR mix from recently collected and preserved adults was an inexpensive, fast methodology to obtain full barcodes. In summary, DNA barcoding combined with a sound morphotaxonomic framework provides an effective approach for the delineation of species and for the discovery of hidden diversity in the subgenus *Trichodagmia*.

Key words: Simuliidae, Blackflies, DNA Barcoding, Cryptic diversity

Introduction

Rapid and reliable identification of species is paramount in many aspects of biological research, especially for studies in systematics, ecology, evolutionary biology, conservation biology, biodiversity, and biomonitoring. Rivera & Currie (2009) stressed that correct species identification not only allows access to the literature on a taxon, but also permits the implementation of control measures for species of medical or agricultural importance. They further argued that misidentifications could lead to inadequate control measures with negative socioeconomic implications. Threats of biodiversity loss induced by anthropogenic change, and the ongoing loss of taxonomic expertise (known as the taxonomic impediment) (Hoagland 1995, House of Lords 2002, Wheeler *et al.* 2004) have