A Standard DNA Taxonomy for Insects?

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Identification of insect species is often problematic because of limited morphological and/or biological characters. DNA data have been used in many phylogenetic studies to help identify and revise species boundaries (Savolainen and others 2005). For many studies, percent similarity DNA compared between species was summarized and intra- and interspecific variation patterns were observed (for example, Brower and Boyce 1991). These patterns suggested that animal species, at most, exhibit 2.0 percent difference among conspecifics (Hebert and others 2004). A group of individuals that exhibited DNA difference greater than the 2.0 percent boundary would potentially represent a new species. Thus, the difference between intra- and interspecific percent DNA variation has been used as a "genetic yardstick" to recognize new species (for example, Hung and others 1999).

The establishment of a standardized percent nucleotide divergence to predict species boundaries would aid in cases where species status is suspect. However, given variation in nucleotide mutation rates and species concepts, association between a standard percent sequence divergence and species is questionable. Cognato (2006) reviewed the percent DNA sequence difference found between insect sister-species in order to assess whether a standard divergence was associated with named species. Comparisons of intra- and interspecific pairwise DNA differences were made for mitochondrial and nuclear loci spanning many insect families. Intra- and interspecific sequence divergences varied widely, 0.04 to 26.0 percent and 1.0 to 30.7 percent, respectively. The ranges of intra- and interspecific sequence divergences overlapped in 28 of 62 comparisons. This implied that a standardized percent sequence divergence would fail to correctly diagnose species for 45 percent of the cases. Common occurrence of non-monophyly among closely related species likely explains this observation. Non-monophyly and overlap of intra- and interspecific divergences were significantly associated. The reviewed studies suggested that a standard percent sequence divergence does not predict species boundaries.

However, a taxon specific genetic yardstick may better predict species boundaries. The application of a standard percent DNA difference to predict species boundaries for an important forest beetle pest, *Ips* was investigated. A phylogeny was reconstructed using nuclear and mitochondrial genes for all species including individuals collected in China whose species identity was suspect. One most parsimonious tree was recovered with a similar topology to previous studies (Cognato and Vogler 2001). Individuals of the suspect species were monophyletic and sister-clade to *Ips amitinus*. The amount and type of nucleotide difference that characterized this clade were similar to the changes observed with currently valid species. Compelling evidence of monophyly, shared nucleotide substitutions from multiple genes, and unique and diagnostic morphological characters suggested that the sister clade to *I. amitinus* is a new species and will be described.

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Mitochondrial cytochrome oxidase (c) I (COI) nucleotide data is currently the most popular gene for delimiting species boundaries. The *Ips* COI data was analyzed independently with an expanded taxon data set, including pairwise nucleotide differences between recognized sister species. The wide range of average intraspecific pairwise nucleotide difference (0.8 to 13.0 percent) was in concordance with the results found in Cognato (2006). This suggests a limited application of taxon specific standard percent nucleotide difference as a means to identify species boundaries. At most, the overall average COI nucleotide intraspecific difference (1.0 percent) provides an informal guide to identify potential clades that may warrant further systematic investigation. DNA data can help best to predict species boundaries via its inclusion in nonphenetic phylogenetic analysis and subsequent systematic expert scrutiny.

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