Preliminary phylogenetic analysis of the genus *Isophya* (Orthoptera: Phaneropteridae) based on molecular data

BEATA GRZYWACZ-GIBAŁA1, 3, DRAGAN P. CHOBANOV2 & ELŻBIETA WARCHALOWSKA-ŚLIWA1

1Department of Experimental Zoology, Institute of Systematics and Evolution of Animals, Polish Academy of Sciences, Sławkowska 17, 31-016 Kraków, Poland
2Institute of Zoology, Bulgarian Academy of Sciences, Tsar Osvoboditel 1, 1000 Sofia, Bulgaria
3Corresponding author. E-mail: grzywacz@isez.pan.krakow.pl

Abstract

A preliminary phylogenetic analysis involving 22 taxa of the genus *Isophya* was conducted using four molecular markers (sequences of mitochondrial cytochrome b (cyt b), fragments of mitochondrial cytochrome oxidase subunit II (COII) and the internal transcribed spacers I and II (ITS1 and ITS2)). Our results indicate that *Isophya* is a monophyletic group with the exception of one species, *I. bivittata*. The analysis revealed a high level of polymorphism in the mitochondrial and nuclear genes of all species. MtDNA performs better in the phylogenetic reconstruction of *Isophya* than the ITS markers. Although the results show some conflict with the systematics of the group known from morphological and bioacoustic data, the study can be the basis for future reconstructions of the phylogeny of *Isophya*.

Key words: Orthoptera, Phaneropteridae, phylogeny, mtDNA, rDNA

Introduction

The genus *Isophya* includes more than 80 species (Warchałowska-Śliwa et al. 2008) distributed in Central Europe, the Carpathian Basin, the Balkans, southern Ukraine, Asia Minor and the Caucasus region, eastwards reaching northwestern Iran and Iraq and, in a disjunct range fragment, the Altai Mountains (Bey-Bienko 1954). Most species of *Isophya* have restricted ranges (Sevgili 2003; Sevgili & Heller 2003) and thus the genus exhibits quite a high rate of endemism.

The genus contains several groups of morphological sibling species (Heller 1988; Heller 2006; Warchałowska-Śliwa et al. 2008; Chobanov 2009a, 2009b) the taxonomic treatment of which is often ambiguous due to morphological convergence between the groups and/or almost cryptic morphological differences within the groups. Further, some taxonomic problems have been elucidated by bioacoustic studies (e.g. Heller et al. 2004; Sevgili et al. 2006). Random amplified polymorphic DNA (RAPD) has been used to study genetic differentiation between some species of *Isophya*. The results have shown a high level of genetic variability (Grzywacz & Warchałowska-Śliwa 2009). However, the phylogenetic relationships between species and groups remain unclear.

Various DNA sequences of mitochondrial and/or nuclear gene regions have been applied to resolving insect phylogenies at various levels (Walton & Butlin 1997; Kim et al. 2000; Knowles & Otte 2000; Caetano-Anollés 2002; Hovmöller & Johansson 2004; Ren et al. 2004; Bugrov et al. 2006; Jost & Shaw 2006; Martinez et al. 2006; Robillard & Desutter-Grandcolas 2006; Shapiro et al. 2006; Vahtera & Muona 2006; Arnedo et al. 2008; Coeur d’acier et al. 2008; Damburg et al. 2008; Hundsdoerfer et al. 2009; Suárez et al. 2009). The aim of the present study was to contribute to the understanding of relationships within *Isophya* by carrying out the first molecular phylogenetic analysis of the genus using four DNA fragments, namely cytochrome oxidase II (COII), cytochrome b (cyt b), ribosomal internal transcribed spacer I (ITS1) and ribosomal internal transcribed spacer II (ITS2).