

Scientific note

A scientific note on mtDNA gene order rearrangements among highly eusocial bees (Hymenoptera, Apidae)

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Animal mitochondrial DNA (mtDNA) gene order is remarkably conserved among higher taxa, however examples of gene rearrangements have been reported (Brown, 1983). We have discovered mitochondrial gene order rearrangements between two bee tribes, Apini and Meliponini within the “corbiculate Apidae” (Michener, 2000), with this being the first report of such an occurrence in highly eusocial bees. These results suggest that further knowledge of mtDNA gene order within the corbiculate Apidae may resolve the question of single or dual origin of eusocial behavior in bees.

Complete mtDNA sequences have been reported for 22 species of Arthropoda and most of the main genes show conservation of position, with few exceptions (Black and Roehrdanz, 1998). Among the mitochondrial genes, the tRNA ones are more likely to suffer transpositions (Smith et al., 1993). Such events are commonly observed between and sometimes within taxonomic orders. A complete sequence for hymenopteran mtDNA has been obtained only for *Apis mellifera* L. (Crozier and Crozier, 1993) and tRNA gene translocations are verified in comparison to other insects. In our study with *Melipona bicolor* mtDNA, preliminary data obtained through PCR showed different fragment lengths than expected compared to *A. mellifera*. Such differences were confirmed by sequencing. We found transposition involving tRNA genes in three different amplified regions (Fig. 1). After the complete genome is obtained, more differences will probably be found. The gene rearrangements observed in *M. bicolor* were also observed through PCR analysis in other 11 meliponine species, comprising three genera (unpublished data). Thus this characteristic appears to be well fixed in the Meliponini. Gene rearrangement in Hymenoptera had been described between two species of parasitic

wasps (Braconidae) involving one tRNA gene (Dowton, 1999).

The controversial question about how many times of social behavior arose in the family has been tentatively addressed by morphology (Winston and Michener, 1977; Engel, 2001) and DNA sequencing data (Schultz et al., 1999). However these studies differed in their conclusions. Further, the molecular and morphological data collected do date have failed to convincingly resolve phylogenetic relationships among corbiculate bees (Schultz et al., 1999). The molecular marker we found (tRNA translocations) is very rare among closely related organisms (Brown, 1983). As changes in mtDNA gene order are considered phylogenetically informative (Boore et al., 1995) and homoplasies are rarely described (Flook et al., 1995), this marker may be of extreme value to study the evolution of social behavior in bees. The fact that the tribes Apini and Meliponini have a different mtDNA gene order is suggestive of an independent origin of eusociality within the two groups. We are now investigating this hypothesis by analyzing the mtDNA gene order present in the two remaining tribes within the corbiculate Apidae, Bombini and Euglossini, and also in several other tribes belonging to the family.

This work reports the first description of tRNA transpositions between animal tribes and, it opens a new perspective for studying phylogeny and evolution of social behavior in Hymenoptera using a powerful molecular tool. To our understanding it is crucial to increase the available data about these rare genomic changes, especially in Hymenoptera. Comparative studies may help to understand not only the evolution of the mtDNA molecule itself, but also the evolution at the organism level.

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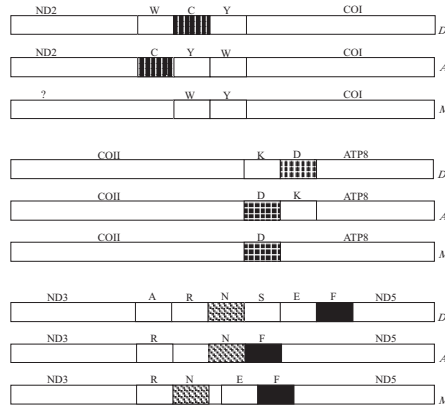


Figure 1. MtDNA regions analyzed and gene order comparisons among *Drosophila yakuba* (D), *Apis mellifera* (A) and *Melipona bicolor* (M). *Melipona bicolor* sequences were initially amplified using universal primers and others derived from *A. mellifera*; the PCR products were cloned and sequenced using an ABI automated sequencer. The sequences obtained are deposited in GenBank under the following accession numbers: AF370439; AF370440; AF370441. The mitochondrial sequences of *D. yakuba* and *A. mellifera* were obtained from GenBank. The main genes (COI: cytochrome c oxidase subunit 1; COII: cytochrome c oxidase subunit 2; ATP8: ATP synthase F0 subunit 8; ND3: NADH dehydrogenase subunit 3; ND5: NADH dehydrogenase subunit 5) showed conserved position among the three species. The hatched boxes indicate the tRNA genes presenting different positions or lacking in the studied regions; tRNA genes are indicated as following: W: Tryptophan; C: Cysteine; Y: Tyrosine; K: Lysine; D: Aspartic acid; A: Alanine; R: Arginine; N: Asparagine; S: Serine; E: Glutamic acid; F: Phenylalanine. Question mark indicates a region not yet sequenced.

Note scientifique sur le réarrangement des gènes de l'ADNmt chez des abeilles hautement eusociales.

Eine wissenschaftliche Notiz zur Umordnung von mtDNA Genen unter hoch eusozialen Bienen.

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