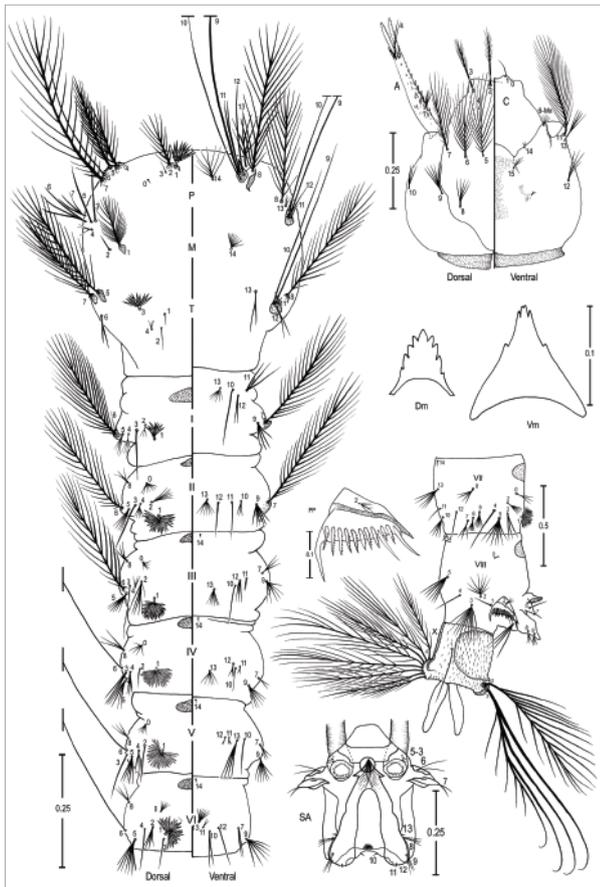


### SYSTEMATICS OF *ANOPHELES (NYSSORHYNCHUS)* (DIPTERA: CULICIDAE)

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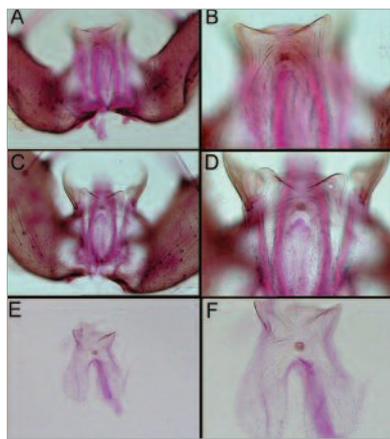
Fourth-instar larva of *Anopheles oswaldoi*. A: antenna; C: cranium; Dm: dorsomentum; M: mesothorax; P: prothorax; PP: pecten plate; SA: spiracular apparatus; T: metathorax; Vm: ventromentum; I–VIII: abdominal segments; X, anal lobe. Scales in mm. Motoki et al. 2007

The proposal aims to continue systematic studies on mosquitoes (Diptera: Culicidae), focusing on the genus *Anopheles*. The main subjects will be the Neotropical subgenera, especially the subgenus *Nyssorhynchus* and *Anopheles darlingi*. Although being the most important Neotropical vector of human *Plasmodium*, the involvement of *An. darlingi* in the dynamics of transmission of malaria in Central America and southern Brazil seems less important than in the Amazon region. In addition, its vector importance is not well established in some localities of southern Brazil. We propose to investigate the possibility that the populations of the Amazon region and southeastern Brazil, eastern of Mata Atlantica, have distinct gene pools.

The main objectives are: 1) genetic characterization of populations of *An. darlingi* of northern and southeastern Brazil; 2) estimation of the phylogenetic position of *An. darlingi* within the subgenus *Nyssorhynchus*; 3) contribution to the identification subgenus *Nyssorhynchus* species, since some species are involved in the transmission of human *Plasmodium*. Additionally, some members of the subgenus *Nyssorhynchus* are able to adapt to human environments. Within this context, we intend to carry out systematic studies on *An. darlingi* and species of the *Nyssorhynchus*, employing both morphological and molecular tools. Fragments of the mitochondrial genes cytochrome c oxidase subunit I (*Cox1*), the ATP synthase F0 subunit 6 (*Atp6*) and the NADH dehydrogenase subunit 6 (*Nd6*), the nuclear elongation factor 1-alpha (*Ef1-alpha*), the nuclear *white* gene, and the second internal transcribed spacer region (ITS2) of the ribosomal DNA will be employed to evaluate genetic structure of *An. darlingi* populations, to estimate its phylogenetic placement within *Nyssorhynchus*, and to examine the existence of species complexes.

## SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

Results of genetic analyses of Brazilian *Anopheles darlingi* *Cox1* mitochondrial gene (from the states of Amapá, Pará, São Paulo, Acre, Paraná, Rio de Janeiro, Espírito Santo, Minas Gerais and Mato Grosso), showed that individuals from the localities situated eastern of Serra do Mar, in the Mata Atlântica, share haplotypes that are distinct from those from the Amazon region and Central America. Individuals from Rio de Janeiro are more closely related to individuals from Amapá, rather than those



Photographs depicting morphological characteristics of the male genitalia of *Anopheles benarrochi* s.l.. Form 1, A: ventral claspette; B: detail of ventral claspette (AC15-109); Form 2, C: ventral claspette; D: detail of ventral claspette (AC18-115); E: dissected ventral claspette; F: detail of dissected ventral claspette. Sallum et al. 2008

from São Paulo and Minas Gerais. Considering the high genetic distance between individuals from Espírito Santo and Rio de Janeiro and those from other Brazilian localities, we have hypothesized that the population from eastern Mata Atlântica may have either a substantially different behavior or distinct vector competence and capacity to transmit human *Plasmodium*.

*Nyssorhynchus* is one of the most studied subgenus of Neotropical *Anopheles*. However, there are several questions regarding the taxonomy of this group. The combination of morphological and gene sequence

data obtained from several species of the genus *Anopheles*, either suggest or corroborate the presence of several species complexes and undescribed rate within *Nyssorhynchus*. Moreover, it is important to consider the possibility that junior synonyms of species may be valid.

*Anopheles oswaldoi* s. l. is a sibling species complex in South America. In the project, we redescribed and performed the molecular characterization of *An. oswaldoi*, using samples from the type locality, and also designated the lectotype.

*Anopheles strodei* includes five species in the synonymy, *An. ramosi*, *An. arthuri*, *An. artigasi* and *An. albertoi*, described in Brazil, and *An. lloydi*, in Panamá. Morphological and molecular evidences from the *Cox1* and *ITS2*, corroborate the fact that there are at least four species under the name *An. strodei* in Brazil and, thus, its junior synonym may be a valid species.

Regarding *An. lutzii*, morphological and molecular evidences of the *ITS2* rDNA support the presence of at least three species that can be misidentified as *An. lutzii* when using characteristics of the adult female.

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