Molecular Characterization of Spittlebug *Mahanarva spectabilis* (Distant, 1909: Hemiptera: Cercopidae) Populations Cccurring in "Legal Amazon" in Brazil

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Molecular characterization of spittlebug Mahanarva spectabilis (Distant, 1909 :Hemiptera : Cercopidae) populations occurring in "Legal Amazon" in Brazil

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Key words: tropical forages, *brachiaria brizantha*, cerrados, rapd markers

Introduction Recent reports have indicated the occurrence of the spittlebug *Mahanarva* in pastures of *Brachiaria brizantha* cv. 'Marandú' in Cerrados and transition areas, located in the Legal Amazon" in Brazil. Paula-Moraes et al. (2006) identified this spittlebug species as *Mahanarva spectabilis* Distant (1909). Since then studies related to the aspects of the *Mahanarva* bioecology, economic impact, its geographic distribution, as well as information on the genetic inter and intra-specific variability have been done. Considering this, the correct identification of the nymphs and adults of the species occurring in different areas has extreme importance. Molecular markers have been used as a tool to support taxonomy studies and to access the genetic diversity of populations of insects and plants. So, the objective of this study was to validate a DNA exctration protocol of *M. spectabilis* stored in different conditions. It also had the objective to analyze the genetic similarity between recollected nymphs and adults of *M. spectabilis* in different sites.

Materials and methods Genomic DNA of adults and nymphs of *M. spectabilis* stored in alcohol 70% and 100% was extracted using a protocol proposed by Faleiro et al. (2003). These insects were recollected in 9 different sites located in Minas Gerais, Distrito Federal, Tocantins and Goiás states. After DNA quantification and a quality check two primers of ten nucleotides length from the Operon Technologies kit were used to amplify genomic DNA. Amplification products were separated by electrophoresis on 1.5% agarose gels and banding patterns were visualized by staining the gels in ethidium bromide solutions and viewing under UV radiation. For the analysis of genetic similarity RAPD markers of 18 specimens, a nymph and an adult, collected in 9 places were analyzed. Genetic distance was later used as a criterion for differentiation among specimens to prepare a cluster analysis.

Results Amplifications were obtained using these two primers. Eighty-five different RAPD bands were amplified. The proportion of polymorphic RAPD loci was 96.5%. The results of the genetic similarity indicate small differences among nymphs and adults recollected in the same place. This result indicates that the nymph can be used to represent a population of a specific collection point. This is very important, because when only nymphs are available, confident identification of the species is often compromised. In such occasions molecular markers become an important tool for inferences on the taxonomic identification of the nymphs. No differences were observed among specimens stored in 70% and 100% alcohol.

Conclusions Molecular characterization was achieved with RAPD molecular markers, which proved to be very informative and efficient to characterize the genetic diversity and relationship among populations of the species. RAPD molecular markers proved to be a useful tool to support taxonomy studies and the corrected identification of insects of *M. spectabilis*. The results of this work offer the basis for future studies of the genetic variability, mapping of occurrence of the *Mahanarva spectabilis* in pastures and population genetics studies.

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