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Identification and study of phylogenetic relationships of the main banana weevil pests (Coleoptera: Curculionidae) in Sri Lanka using mitochondrial DNA sequences

Mitochondrial DNA (mt DNA) sequences are used widely as an informative guide to study the relationships among species especially those with close morphologies. The weevils *Cosmopolites sordidus* and *Odoiporus longicollis* are the two main pests of banana in Sri Lanka and are found infecting the same plant. Morphologically, the adults appear similar. In the present study, Polymerase Chain Reaction (PCR) based mtDNA sequence analysis is used to identify the banana weevils and study their phylogenetic relationships. A total of 80 weevils were collected from the banana rhizomes and pseudostems from plantations at Ambepussa. Morphological studies showed four groups of weevils with average total body lengths of 18.6 ±1.439 mm (Group 1), 16.3 ±1.070 mm (Group ii), 16.7 ±1.138 mm (Group iii) and 6.4 ± 0.378 mm (Group iv). The weevils of Group iii were brown in colour, while the rest were black. Microscopic examination revealed that the banana weevils have two different spot patterns; Group I and Group iii had spot marks on either side of the elytra. The other two groups had spot marks spread all over the elytra. Genomic DNA was isolated from single insects by using the Malcolm method. Two universal primers which were designed from the 5' end of the 12S gene in mitochondrial DNA of insects was used in the PCR. Ten weevils from each phenotype were sequenced in both directions. The 355 bp PCR products were directly sequenced and the computer analysis confirmed similar sequences in Group iii and Group i. This result indicates that they belong to the same species and comparison with specimens confirmed that both groups belonged to *Odoiporus longicollis.* The black weevils in Group ii and Group iv showed different sequences and specimen comparison revealed that Group ii belongs to *Cosmopolites sordidus.* Group iv weevils were identified as banana saprophytes. Further studies are necessary to identify its real taxonomic position. According to the data base BLAST, the closest sequence homology to the weevils was *Chrysolina americana* (Chrysomelidae). The sequence similarity between C. *sordidusand* 0. *longicollis* was 81.1%. The saprophytes show the highest similarity with C. *sordidus* (74.5%). Mt DNA sequencing has confirmed the identification of C. *sordidus* and 0. *longicollis* among the banana weevils and established their phylogenetic relationships and position in the dendrogram.