

Poster: Taxonomy and Morphology

Development of a multi-entry identification key for economically important fruit fly (Diptera: Tephritidae: Dacinae) larvae

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Background: Identification of fruit fly larvae are typically difficult due to the limited morphological characteristics present. However, this is the stage at which fruit flies are intercepted at ports of entry when importing fruits and vegetables. Molecular tools are useful, but DNA analyses take time and are expensive, compared to morphological identifications. This project aims to use available information from literature and our own research to build a multi-entry identification key for thirteen tephritid species, including species complexes, such as the *B. dorsalis* complex and *Ceratitis* FAR complex, that are of economic concern for the European Union.

Methods: Third instar larvae were obtained from a variety of different regions and hosts, including South Africa, La Reunion, India, Spain, China, Greece, Israel, Australia and the IAEA colonies in Austria. Thirteen species or representatives of species groups were obtained, based on availability, including *Ceratitis*, *Dacus*, *Bactrocera* and *Zeugodacus* spp. The cephalopharyngeal skeletons were dissected out, cleared in a 10% NaOH solution, dehydrated with 70-100% alcohol and mounted in Euparal on glass slides. Images of at least 20 larvae/species were captured using a compound microscope fitted with a camera and 400x magnification, measurements taken of 1) distance between ventral apodeme and apical tooth, 2) ventral angle between apical tooth and ventral apodeme, 3) distance between dorsal apodeme and ventral apodeme, 4) ventral apodeme and pre-apical tooth, and 5) apical tooth and pre-apical tooth. The number of tubules in the front spiracles were counted and the position of the spiracles in relation to the cephalic skeleton was noted. Data were analysed using a one-way ANOVA with multiple variables and discriminant analysis with classification functions of the effective principal components.

Results: For development of the multi-entry key and mobile application, the LUCID software was used. A matrix was compiled for 13 species and included 9 characters for which significant inter-specific differentiation was preliminarily detected. The key starts with options for separating Tephritidae larvae from other commonly encountered insect larvae in fruit, including Drosophilidae, Tortricidae, Pyralidae, Nitidulidae and Lonchaeidae. Finally, the key was converted into a mobile application by LUCID, for both Android and Apple devices.

Conclusion: This is the first time a multi-entry key for tephritid larvae of economic significance has been developed. While the characters rely mostly on measurements, it does require some knowledge of dissecting out and treating the mouthparts, so that measurements of specific distances can be made. However, as nothing else exists for this list of species, it will be a valuable tool for enabling non-molecular identifications of fruit fly larval pests in fruit.

Keywords: Morphological identification, *Ceratitis capitata*, *Bactrocera dorsalis*, *Bactrocera tryoni*, *Zeugodacus curcurbitae*, quick detection