G67 Rehydrating Dried Blow Fly Larvae to Reclaim Their Usefulness in Forensic Investigations

Michelle R. Sanford, MS, Jennifer L. Pechal, MS, and Jeffery K. Tomberlin, PhD, 2475 TAMU, Department of Entomology, Texas A&M University, College Station, TX 77843*

After attending this presentation, attendees will learn methods for rehydrating dried larval insect specimens. The impact that initial preservation coupled with drying and rehydrating of larval specimens on their length and weight as it relates to estimating period of insect activity also will be discussed.

This presentation will impact the forensic community by demonstrating how studies on methods development in forensic entomology can benefit the forensics community by being used to define protocols and standard operating procedures that can be cited and used in legal proceedings.

Ethanol is commonly recommended for preserving larval blow fly specimens in forensic investigations. Alcohol is a volatile preservative that can evaporate over time resulting in the dehydration of larval specimens or the creation of *crispy maggots* which are difficult to identify and unreliable for measurements for age estimation. In this study methods recommended for rehydrating dried museum specimens were adapted and applied to *crispy maggots* of three common North American blow fly species (*Phormia regina* (Meigen), *Cochliomyia macellaria* (Fabricus), and *Chrysomya rufifacies* (Macquart)). Length and weight of the specimens were documented throughout the process. The effect of initial preservation method was also observed by collecting replicate samples and preserving in 80% ethanol, 70% isopropyl alcohol, or with fixation by hot water killing followed by preservation in 80% ethanol. Third instar larvae were collected over the course of nine months from different animal carcasses used for teaching the Texas A&M University forensic entomology course. Individual third instar larvae from each species (n = 90/species) were measured and weighed before the preservative was allowed to evaporate. Rehydration was attempted by soaking overnight in 80% ethanol, a commercial trisodium phosphate substitute solution, or 0.5% trisodium phosphate solution after which specimens were again measured and weighed. Analysis of length and weight data with analysis of variance showed that for each species the impact of rehydration and the impact of the interaction between initial preservation and rehydration treatment significantly affected final rehydrated length and weight among the different species.

For all specimens, soaking in any of the rehydration treatment solutions restored a portion of the original larval length (mean percent difference initial–final across all species and preservatives: 80% ethanol: -10.6%; trisodium phosphate: -2.9%; trisodium phosphate substitute: 1.1%) but none of the solutions were able to restore original larval weight. The original larval length and the final rehydrated larval length were used to estimate larval age using published data sets. These estimates agreed within a few hours in many cases with individual preservation by rehydration treatment combinations more closely agreeing for some species than others. A comparison between the length-based larval age estimate and the known duration of the exposure of the animal carcasses revealed that there were large differences (percent difference between estimated and actual exposure: *P. regina*: 74% lower than actual; *C. macellaria*: 51% lower than actual; *C. rufifacies*: 150% higher than actual) which probably reflect delays in, and barriers to, colonization coupled with differences in tissue types used in published studies and this experiment.

Overall the data show that *crispy maggots* can be rehydrated and suggest that their length can be measured to obtain a length-based age estimate for period of insect activity estimates. Knowledge of the initial preservation method might also aid in selecting the most appropriate rehydration method.

Studies on methods development in forensic entomology can benefit the forensic sciences community by being used to define protocols and standard operating procedures that can be cited and used in legal proceedings.

Diptera, Method, Length

 ** Presenting Author*