SIMPLIFIED DNA BARCODING STRATEGY FOR FORENSICALLY RELEVANT BLOW AND FLESH FLIES

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Accurate insect identification is critical to their use in estimation of time of colonization (TOC) and post-mortem interval (PMI) during medicolegal death investigations. Insect specimens are currently identified by evaluating morphological characteristics as indications of particular taxonomic groups; however, this process is limited because immature life stages typically lack distinguishing morphologies. These deficiencies may be addressed through molecular identification by DNA “barcoding” wherein DNA sequences from unknown samples are matched to references. This technology enables identification of immature specimens, may be performed without specialized forensic entomology training, and requires equipment common to forensic genetics laboratories. DNA barcoding has been demonstrated in numerous entomological surveys of forensically relevant species; however, the technology has not been implemented for medicolegal death investigations. This is due in part because of deficiencies in the technology: no single primer set is capable of distinguishing all of the diverse species important to forensic investigations. Instead, multiple primer sets and sequencing reactions are utilized to maximize the species that may be identified. We propose a simplified DNA barcoding strategy for identifying insects commonly encountered in casework at Harris County Institute of Forensic Sciences (HCIFS), in particular, blow and flesh flies of families Calliphoridae, Sarcophagidae, and Phoridae. The strategy comprises sequencing and phylogenetic analysis of a single barcoding fragment amplified from the mitochondrial COI locus. Using verified reference specimens and samples collected during past casework, we show that the DNA barcoding strategy enables statistically supported identification of immature samples, for example, pupal exuvia, first and second stage larva, and adult fly legs, and resolves closely related species including members of the blowfly genus *Lucilia.* Future work will include elucidating sequence variations of local blow and flesh fly populations to provide further statistical support for identifications, and validating the DNA barcoding assay for casework application.