



G63 Forensic Entomology and Bacteriology for Postmortem Investigations in Romania

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After attending this presentation, attendees will understand some principles regarding the correlation of information provided by bacteria DNA and necrophagous insect taxonomic analyses, having as an objective accurate postmortem interval identification.

This presentation will impact the forensic science community by serving as a means of reference and comparison for future research regarding the identification of postmortem interval by means of bacteria and necrophagous insect dynamics.

Forensic entomology is a location-dependent science, widely developed in the United States and Europe. Throughout the years, this discipline has gained large amplitude in the field of forensic sciences, starting with classical taxonomy, DNA identification of adult and immature stages of necrophagous insect species, and entomotoxicology. Meanwhile, forensic microbiology represents a recent research direction. The objective of both fields is to determine the postmortem interval, sometimes with specification of manner and place of death.

In Romania, forensic entomology is not recognized as an additional distinct domain of forensic science and the information that could result from entomological expertise is not used by police forces. Given that this type of study is bioregion specific, research on Romanian territory is necessary and justified. Consequently, this research is interested in determining the succession of necrophagous insect species in Romania and in identifying the microbial diversity dynamics within the animal carcasses for defining microbial targets in the process of decomposition, using exposed pigs as an experimental model.

The study focuses on the characterization of necrophagous insect species' chronological succession and their stages of development while inhabiting pig carcasses exposed in an urban natural environment (Bucharest, Romania). The composition of the corresponding bacterial communities inhabiting the carcasses colon and mouth cavities, in correlation with climate condition and decomposition stages were studied. The experiment was monitored for up to 11 months during cold (November-May) and warm (July-October) periods. The baits were not accessible by vertebrate scavengers and all metrological parameters were continuously recorded.

During each experiment, both adult and immature necrophagous insect species were collected and taxonomically identified. Tissue samples were harvested from internal (4-8cm) and external sections of the pigs' colons and mouths on a weekly basis. Total bacteria genomic DNA was extracted from each sample and bacterial 16S-rDNA fragments were amplified by Polymerase Chain Reaction (PCR). Bacteria diversity was investigated by Denaturing Gradient Gel Electrophoresis (DGGE) analysis and sequencing of electrophoresis gel-extracted DNA fragments.

The results showed an accelerated activity of necrophagous insect species during warm periods. Their diversity in succession was high, with captured species of the families *Calliphoridae*, *Muscidae*, *Anthomyiidae*, *Fanniidae*, *Sepsidae*, *Phiophilidae*, *Cleridae*, *Silphidae*, *Staphylinidae*, *Dermestidae*, and *Histeridae*. The appearances of new necrophagous insect species over time and their development stages showed a correlation with the postmortem interval, meteorological parameters, and carcass decomposition stages.

Bacteria 16S-rDNA DGGE analysis revealed the presence of a high species/strains number in the mouth cavity more so than in the colon during the warm period. During the cold period, the number and representation of bacterial species was constant in both cavities. After ten weeks exposure, new bacterial species appeared in the colon cavity, while in the mouth cavity this diversity occurred after six weeks, a phenomenon associated with temperature increase.

A correlation of bacterial dynamics with the decomposition process and necrophagous insect species activity was observed. Both insect and bacterial communities' dynamics were dependent on the seasonal atmospheric conditions.

The information resulting from this taxonomy and molecular study on necrophagous insects' and bacterial communities' dynamics at two locations of exposed pig carcass in an urban Romanian location represents incipient data that can be used for accurately determining the postmortem interval and for establishing forensic entomology as an official tool in criminal investigations in this country.

Entomology, Bacteria, Forensics