Role of entomology in forensic sciences

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Forensic entomology is the application of insect biology in criminal investigations. Insects are important agents in the biological breakdown of corpses and often provide valuable clues in criminal investigations[1,2]. Entomological evidence is particularly useful in estimating the time of death in cases where the post-mortem interval (PMI) is prolonged and the value of other methods is limited. Insects also provide spatial or temporal information of forensic material which cannot be obtained otherwise. This field of study has unfortunately not received much attention; in North America, this field of science is a little over 20 years old[3]. It utilizes the basic concepts of insect systematics, ecology, toxicology, physiology and molecular biology. This article highlights the different strategies of entomological approaches that are used besides emphasizing the need to incorporate this discipline in the academic curriculum at the Master's level under Applied Entomology/Zoology.

Insect diversity on the carrion

A knowledge of the insect species associated with different habitats may provide information regarding the history of the remains. Several species of insects belonging to the orders Diptera, Coleoptera, Lepidoptera and Hymenoptera are associated with decaying matter, including corpses[1,4,5]. The Dipterans are predominantly the largest group both in terms of species diversity and density followed by the beetles (Coleoptera) belonging to families Histeridae, Staphylinidae, Dermestidae, Rhizophagidae, Ptinidae and Tenebrionidae. Wasps and ants are predaceous, preying upon insects that invade the carrion. The diversity of these insect species has been found to vary in different ecosystems depending on the habitat of the carrion as well as that of buried and unburied cadavers. Such difference in diversity paves way for possible suspicion/prediction about any crime. For example, in a particular study dealing with pig carrion which had been designed to mimic a homicide scene, observations revealed a complete absence of maggot masses in all the buried carcasses in contrast to that of unburied or pitfall trap records[6]. This reflects that presence of maggot by chance in any one of the buried carcasses is a case of suspicion of delayed burial of the body. If that is the case, the minimum PMI can be calculated on the basis of density as well as diversity of the different stages of flies. In recent years, several mathematical models have been developed based upon observations on development times for stages and instars in experimental conditions and interpolating these data against on-site conditions to estimate PMI[7].

Succession of species

Decomposing bodies undergo biological, chemical and physical changes and at each stage of decomposition, they are invaded by a specific species of insect – often in a predictable sequence[6]. These changes are dependent on interrelated factors such as climate, situation and insect access[5]. For instance, blowflies are the first colonizers of the carrion and include several important genera, viz. *Calliphora*, *Lucilia*, *Cochliomyia* and *Phormia*. The sarcophagids arrive a few days later. A characteristic pattern of the maggots of these species is their movement in large numbers which presumably facilitates their combined ammoniacal excretory products to condition the tissues and counter the acidic effects of rigor mortis[8]. As these species develop and the process of decay sets in, the odour of the corpse changes becoming more ammoniacal and putrescent[5]. This results in attraction of other Dipteran species, particularly the Phorids and later, the Stratiomyids and species succession begins. Beetles colonize corpses later than the flies probably defining specificity with a particular stage of decomposition. It may also be an adaptation to reduce competition or the fact that Dipterans are strong fliers and arrive at the site of the corpse earlier[9]. Histerid and Staphylinid beetles are the first to arrive followed by beetles belonging to Dermestidae, Rhizhophagidae, Ptinidae and Tenebrionidae. Several species of Lepidopterans may be associated with the corpse at different stages of decomposition, being attracted by the urine, excreta and nitrogenous fluids[5]. Occurrence of these insects in different stages and their density further provide clues about the approximate time that has elapsed since the death of the animal[10]. However, we need to re-evaluate our judgement based on this criterion to determine PMI. It needs to be appreciated that fertilization in Diptera is often quite complex and it would be appropriate to base the estimate on the age of the larval stage that is present in largest numbers, i.e. the stage representing the progeny resulting from eggs fertilized during oviposition[11]. Besides, nocturnal oviposition in several Dipterans can alter the estimate of PMI by as much as 12 h (ref. 12). In addition, preservation of maggots in fixatives results in variable degrees of tissue shrinkage which may lead to erroneous estimates of PMI when the criterion is the size of the maggots. An underage error of 9.7 h was reported for maggots of *Protophormia terranovae* when stored in 70% alcohol as against 16.8 h when stored in formalin[13]. The authors suggested a need to standardize the treatment of maggots and recommended the use of boiling water for killing them (as it reduces autolysis by destroying the gut flora and digestive enzymes) and subsequent storage in 70% ethanol.

Bionomics of insects

The duration of development is another viable source to determine PMI. While the presence of blowflies and flesh flies during the initial phases of decay provides accurate estimates of PMI, it is difficult to do so in advanced stages of decomposition, i.e. when the corpse has reached post decay to dry stage. The black soldier fly, *Hermetia illucens* (Stratiomyidae) infests corpses in advanced stages of decomposition and knowledge of its life history pattern has enabled investigators to solve several cases in the US[14]. The adult females oviposit 20–30 days post-mortem and the life cycle takes an additional 55 days. Thus, a thorough knowledge of life cycle duration, age and stage of the insect occurring on the carrion along with data on other cohabiting arthropod species, under prevailing local environmental conditions, provides investigators with valuable data to form a template for dating the remains.

Genome analysis

DNA analysis of the suspect using molecular biology tools is an emerging field of forensic sciences allowing accurate evidence to help solve criminal cases such as murder, bank robbery, theft and related disputes. The use of mitochondrial DNA and 18 s RNA has been successfully employed in establishing the phylogenetic relationships between insect species[15,16]. The characterization of human DNA from blood feeding insects has advanced substantially the application of PCR and subsequently AMP-FLP analysis providing individual human locus characterization from insect species[17]. Sequencing of mt DNA from a single hair of pet animals has provided evidence to establish the suspect involved in criminal cases[18].

Corpse relocation

Forensic experts also come across situations when the body is moved from the original scene of the crime. In a landmark case in Belgium, forensic entomologists observed 5 species of staphylinids on the bones of a corpse but the absence of other insect species in spite of favourable conditions suggested that the murder was committed elsewhere[19]. In addition, the biochemical differences among geographical populations help in determining corpse relocation and in the study of population ecology of species and populations.

Detection of controlled substances

Insects also provide valuable clues in cases of deaths related to narcotics or toxins, particularly more so when the corpse itself is too putrefied to analyse. Several studies have elaborated on the detection of various toxins and controlled substances using GC-MS and HPLC and related toxicological techniques in insects found on decaying corpses[20,21]. In the cases where standard toxicological specimens are absent, detection of drugs in chitinized insect tissues from mummified corpses is a new development in the field[22].

Drug trafficking

In a typical case study on drug trafficking, police could establish the country of origin of the drug through insects[23]. On seizing the contraband material (*Cannabis*), police noticed the presence of insects which on identification revealed the drug route as well as the country of origin based on the geographic distribution of *Cannabis* as well as the associated insect species.

Other clues

Under normal conditions, insects invade the corpse through body orifices such as nose, mouth, eyes, ears, anus, etc. Wounds inflicted due to a knife or a bullet injury attract flies for feeding and egg laying. The aggregation of flies and their life stages help experts to trace the spot and predict how the murder was committed[23].

Forensic entomology in the Indian scenario

Unfortunately not many laboratories in our country deal with this specific subject. Reports of records are more often from the viewpoint of classical taxonomy unlike in other countries where continuous systematic and biological studies are undertaken to arrive at meaningful conclusions. Several variables related to decomposition are often neglected. These inter-related factors such as insect activity, ambient temperature, rainfall, burial depth, presence of carnivores, etc. need to be observed for the outcome of a successful investigation. Further interest and growth in this field can be generated only when forensic entomology is introduced in the academic curriculum of Indian Universities under Applied Zoology/Entomology.

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