Insects as forensic informants: the Dutch experience and procedure

J. Krikken & J. Huijbregts

National Museum of Natural History Naturalis, P.O. Box 9517, 2300 RA Leiden, The Netherlands

The principles of forensic entomology are described, and reference is made to the hierarchy in the scale of ecological processes involved. The procedural steps in The Netherlands, from crime scene investigation to the final entomology report, are reviewed. Five illustrative cases out of many are briefly described. One conclusion is that regional calibration of published life cycle data, based on few original sources, is urgently required.

Keywords: forensic entomology, carrion community ecology, post mortem interval

Insects and other arthropods can be excellent forensic indicators, being the silent witnesses of criminal events and their aftermath (Smith, 1986, latest manual Byrd & Castner, 2000). The typical death-scene investigator learns quickly that maggots and corpses go together (Haskell et al., 1997). Especially these maggots, usually blowfly larvae (Diptera: Calliphoridae), have proved to be reliable indicators of a minimum post-mortem interval (PMI) of deceased humans, i.e. of the minimum estimated time lapse between death and the actual observation of the corpse of, for instance, a murder victim. In spite of the fact that forensic entomology as a scientific discipline has been with us for more than 150 years (Mégnin, 1894), there is still little to be found in standard medico-legal, pathological and forensic texts (Simpson & Knight, 1985, Cohen, et al, 1996; Fisher, 2000, still has only one and a half page), but this may change, as there is something like a boom in forensic entomology publications (Goff, 2000; Byrd & Castner, 2000; Erzinclioglu, 2000).

In this paper we present an overview of forensic entomology as practised in The Netherlands during the past decade, with special reference to biological background and its application to the reconstruction of forensic situations where PMI and alibi questions play a primary role. During this period we have been consulted about four times per annum, mainly by the forensic technical units of Dutch district police departments, frequently on advice from forensic pathologists in the Netherlands Forensic Institute at Rijswijk ZH. Occasionally requests for entomological expertise were received from the prosecution side in criminal cases. A revised procedural overview for technical detectives in The Netherlands is in preparation (Krikken & Huijbregts, in prep.), and forensic entomology has just become part of the technical detective curriculum. We present five generalized cases in which our advice was requested.

Our expertise with carrion insects is not only based on forensic cases, but on an experience of decades dealing with the taxonomy and ecology of insects (mainly beetles and flies) associated with carrion, dung, and other decomposing organic substances, baited pitfall trapping being our favourite field technique.

BIOLOGICAL AND ENVIRONMENTAL BACKGROUND

Insects are indeed practically without exception intimate witnesses of the post-mortal phase of humans, right from the moment of death on, and sometimes even before. Unless preventative measures are taken, insects, mostly blowflies (Calliphoridae, see Erzinclioglu, 1996, for an introduction to the family), colonize the corpse within an hour after death, rapidly starting oviposition. Above 12°C, in our region, species of Calliphora and Lucilia are predominant – and they are almost everywhere. Later on other insect groups arrive, including members of other fly families, and species of Coleoptera, Hymenoptera, Lepidoptera. Immediately after the blowflies have done their work, the strictly sarcophagous species are followed by their predators and parasites. An analysis of this carrion community at a particular point in time may reveal its position on the timeline of succession – in other words, the ecological stopwatch gives us the start of the decomposition process. The analysis may also reveal specific environmental events; for
instance, allochthonous elements on the site could be indicative of transport of the corpse. In short, forensic entomology can contribute to the reconstruction of the history of a corpse.

The literature on the biology of insect faunas on carrion and associated population ecology is abundant (Putman, 1983, Smith, 1986), in addition to an ever increasing number of published forensic cases (Catts & Haskell, 1990, see also relevant web sites). Rearing experiments with blowflies under varying conditions, as well as studies of other carrion arthropods, have greatly improved the scientific basis of forensic interpretations. The description of the immature stages of carrion insects, however, is still very incomplete, in spite of the existence of substantial synoptic work (most of which is intended to support taxonomy and supraspecific identification, like Hennig’s work on dipteran larvae, 1948-1952, etcetera). Recently we had to turn to an American publication (Rattcliffe, 1972) to understand our own observations on European *Necrodes* (Coleoptera: Silphidae) in a forensic case – its life cycle is undescribed, and larval descriptions from Europe proved to be inadequate.

Important in the understanding of what happens on and around the corpse, and not immediately apparent in the current forensic entomology literature, is the recognition of the hierarchy and spatiotemporal scale of the processes involved, *i.e.* the fact that the faunal succession is made up of the numerous metamorphoses of individual species. Space plays a role in that the processes go on from landscape scale to the micro-situations on and around a corpse. The following list of the relevant processes and factors, with comments, may aid in the interpretation of entomological results.

Metamorphosis — Knowledge of both the metamorphosis of the colonizing species in time, and the activity of each stage in space, is crucial. For the common blowflies the literature is considerable, although sometimes still anecdotal, not based on experimental work. For other insects it is a matter of simply searching the literature, but, whatever the case, it remains doubtful whether the data available for species elsewhere in Europe are unreservedly applicable to our local situation. Regional calibration is urgently required. Species of *Calliphora* and *Lucilia* appear to be best known, and their indicator value is undisputed. They have a complete metamorphosis, from egg through three larval stages, to postfeeding and prepupa stages, and a final pupa stage (example in Fig. 1). Development speed is heavily temperature dependent, but normal summer temperatures produce a new generation within three weeks. Sometimes PMI estimation may be in hours rather than days, *i.e.* when eggs and minute first instar larvae are observed; this implies that the number of hours elapsed since oviposition lies between 12 and 18 h. As blowflies oviposit only during daytime hours, and some (*Lucilia*) only at high body surface temperatures (over 30°C), the oviposition period, and henceforth the PMI, may indeed be reasonably well restricted to hours rather than days.

![Figure 1. Life cycle of Calliphora vicina Robineau-Desvoidy](image-url)
Succession — There is a general pattern in the succession of insect faunas on carrion worldwide, but the details in this pattern may vary enormously between regions and from one particular site to another. In exposed situations in temperate to warm climates blowflies usually are the first colonizers of a corpse. Soon other groups arrive, starting with insects preying on blowfly offspring (like Coleoptera, e.g. Histeridae, Staphylinidae; Hymenoptera: Formicidae; also Acari, e.g. Gamasida), then parasites and hyperparasites take their turn (like Hymenoptera: e.g. Braconidae), and still later non-calliphorid sarcophagous Diptera (e.g. Piophilidae, cheese skippers) arrive. The last to arrive on the leftovers, sometimes appearing only after weeks or months, are saprophagous forms (like Coleoptera, e.g. Cleridae) and dry flesh-and-skin, bone, and keratin feeders (Coleoptera, e.g. Dermestidae, Lepidoptera: Tineidae). The occurrence of particular species may be heavily dependent on the season (see Season).

Activity cycle — The intrinsic daily activity of insect species plays a role in the interpretation of the fauna on a corpse. Calliphora and Lucilia blowflies are diurnal insects, the females limiting oviposition to the daylight hours with their higher temperatures – Lucilia prefer sunny situations. Coleoptera, e.g. carrion beetles (Silphidae), are mostly nocturnal predators and scavengers, during the daytime usually found hiding under the corpse, or buried into the underlying soil. This is why sampling, or complete sifting, of soil under a corpse is obligatory.

Season — The intrinsic seasonality of the occurrence and activity of the various insect species may be another crucial factor in forensic interpretation. The exact species composition – the actual succession – on a corpse varies according to season, although the important calliphorid occurrence remains rather stable during spring, summer and autumn, i.e. as long as average daytime temperatures do not sink below 12°C. Other dipteran groups take over during the colder season, such as Trichoceridae (winter gnats), but the decomposition speed can be extremely slow outdoors, and indeed tetrapod predators and scavengers may take over from the invertebrates. The literature containing data on the phenology of carrion insects other than blowflies is extremely scattered and limited, and regional synopses on the seasonal occurrence of species are urgently required.

Biotic environment — Two overriding extrinsic factors influencing the events on and around a corpse may be the vegetation structure (forest versus open terrain, presence of trees and shrubs producing shade, thus influencing temperature and humidity, see Abiotic environment), and elements in the non-entomological fauna (as already hinted at under Season), varying from foxes, dogs and wild boars to straightforward human intervention. The animal elements may not only influence the events on or around the corpse itself, but they may completely relocate it, shelter it, fragment it, bury it, etcetera. This may be apparent only afterwards, for instance, by meticulously sampling the soil fauna on different locations on and around the location of the corpse. Sheltering a corpse can lead to discrepancies between the decomposition phase of a body as established by the pathologist and the anticipated occurrence of blowflies, i.e. a delayed colonization, or even a complete absence of these flies. Burial of a corpse usually leads to a dominance of flies of other dipteran families, i.e. species of Phoridae (including so-called coffin flies).

Abiotic environment — The daily and seasonally changing factors of temperature, air humidity, precipitation, other weather conditions, and soil properties, are of crucial importance. The medium in which the corpse was found may be different from air and soil, such as water (consequently involving a whole suite of aquatic organisms, including insects). As the speed of development of insect larvae is a direct function of temperature, the data of the nearest weather station must be obtained. In maggot concentrations on the corpse the temperature may rise well above that of the surrounding air, and consequently whenever possible temperature records are to be taken in the large larval masses. Human body temperature is normally 37°C, and decreases 1°C every hour after death. Some Lucilia species oviposit only on surfaces of over 30°C, and the recognition of this may play a important role in PMI estimation. Other flies prefer depositing their offspring under clouded conditions, as observed in certain Sarcophagidae (flesh flies – which, by the way, have the additional complication of being larviparous).

Ante-mortar physical condition of the deceased — The anatomical, chemical and pathological condition of a person prior to death may influence the distribution and development of the insect
fauna on the corpse. Lesions, body pose, presence of drugs, and personal hygiene are all known to have played a role in forensic cases. Cocaine sniffing is known to have led to abnormal development of maggots on a victim – important to know, as the PMI estimation is usually based on the largest (oldest) available stages in the calliphorid life cycle. Even on live land vertebrates lesions may be infected with ovipositing flies (*myiasis*, Zumpt, 1965) – as humans in battle field situations have known for ages, and, conversely, the presence of numerous maggots away from natural body orifices may be indicative of lesions inflicted prior to death.

**THE DUTCH PROCEDURE**

Forensic entomology is increasingly becoming part of normal crime scene investigations in The Netherlands – certainly whenever, on or around a victim, insect activity is conspicuous. Technical detectives collect forensic evidence on the crime scene (in Dutch “Plaats Delict”, P.D.), and now frequently and systematically, pick up insect and other arthropod material, on their own, or on the advice of the forensic pathologist. Occasionally the state prosecutor in a criminal case asks for a special *pro justitia* report, and the forensic entomologist then acts as an expert witness.

The sequence of actions at, and following, a normal crime scene investigation is usually as follows:

**Crime scene investigation** — Detectives sample the site of the corpse according to detailed instructions given in a forensic entomology checklist, which they obtain by fax, from the Internet, or, shortly, from a publication (Krikken & Huijbregts, in prep.).

**Consulting the entomologist** — They contact the forensic entomologist as soon as possible by phone for consultation, receiving further sampling, recording, sample preservation, packing, and delivery instructions.

**Delivery of documentation** — Detectives report to the entomologist according to detailed checklist instructions, delivering also pictures, sketches of the corpse and sampling locations, topographic maps, meteorological data, sample lists with codes; occasionally autopsy reports.

**Delivery of physical evidence** — Fresh and preserved samples are delivered to the entomologist personally, as soon as the autopsy has been completed, labelling being consistent with the documentation under **Delivery of documentation**.

**Handling of samples and documentation** — The entomologist, on receiving the samples decides what to preserve, what (maggots and other immature stages) to attempt to rear to the adult stage, and what (soil samples) to analyse further, e.g. by means of Berlese extraction.

**Provisional reaction** — Detectives, on delivering the material, receive oral comments, and if the analysis is likely to take long, they may receive a provisional conclusion in writing, usually within a month after the delivery of the evidence.

**Analysis material and extraction of samples** — Sample extraction on Berlese funnels and other analytic techniques are conducted; Berlese extraction is usually limited to two weeks under room temperature conditions, subsequent sorting and identification (not necessarily to species level) may take another week.

**Results in spreadsheet format** — After completing the rearing, extraction, and identification efforts the results are entered in a standardized spreadsheet, to be attached to the final reaction.

**Final reaction** — A brief standardized report to the technical detectives is then drafted, taking into account all the material available; this report contains conclusions aiding in PMI estimation and/or in the reconstruction of forensically relevant circumstances.

**Follow-up research** — Further research may be conducted on request, such as an intensive microscopic analysis of additional sample material, a more detailed identification of organisms recognized (insects and other invertebrates) found or still to be recovered from remaining soil samples, and, involvement of additional experts. Tariffs are set for this follow-up work.

The forensic biology checklist mentioned under **Crime scene investigation** was developed on the experience of the past decade and the literature (Catts & Haskell, 1990). The list contains detailed instructions on the administrative, environmental, and sampling data to be delivered, as well as on the actual entomological evidence required – maggots in the first place! It includes advice on how to actually sample the site of the corpse, and how to preserve, pack, transport, and deliver the evidence; finally, there is a list of what field equipment and supplies to use.
The Berlese extraction mentioned under Analysis material and extraction of samples is one of various soil biology techniques that can be applied to forensic samples (Dunger & Fiedler, 1997). It should be noted that in The Netherlands, autopsy reports as mentioned under step Delivery of documentation rarely mention PMI estimates.

Although in most cases detectives follow the checklist instructions meticulously, certainly after having been made aware of the biological rationale of the various items, some serious points of attention always remain to be repeated at step Consulting the entomologist above, such as:
- Absence or scarcity of particular insect species may be as good for drawing conclusions as is their presence and abundance; selective sampling may result in considerable bias if not explicitly reported.
- Deliver the samples without delay, adequately labelled, and keep live material, including soil samples, airy; store hand-picked specimens in small labelled containers (vials) with a gauze cover or perforated lid, samples of earth, humus, and litter in fine-meshed bags instead of closed jars.
- In case a corpse is suspected to be more than three days old (e.g. on pathological evidence), sample also at some distance from the corpse, in outdoor situations take soil samples at 4-8 m, accounting for larval migration; always sift the soil under the corpse for maggots and other insects, apart from taking normal soil samples.

FIVE DUTCH CASES

Five illustrative cases of unnatural death dealt with by us in the past decade have been selected here. Their descriptions are somewhat generalized for the sake of brevity. The conclusions drawn from entomological evidence were consistent with conclusions based on other types of evidence in the case concerned.

Early development of blowfly larvae on a dead body in a park, indicative of a short PMI, pointing to death having occurred before dusk the previous day:

On a warm summer day, a young girl went missing during the afternoon, reputedly after 15:00. The mother reported the event at 19:00. At noon the next day the girl was found dead in the park where she was last seen by her mother. Detectives noticed egg masses on the body, and brought in samples and pictures, including some on which Lucilia were clearly visible on the body. The eggs had started to hatch, and considering a development of at least 12 h and the fact that oviposition is a daytime affair, the death of the girl must have occurred before dusk the previous day.

Allochthonous presence of a body in a closed indoor situation, the likelihood of transportation of the deceased indicated by the mass presence of Lucilia adults, normally characteristic of warm, exposed situations:

A male corpse was found in an upstairs room, with windows blinded. There were masses of dead adult Lucilia around, as well as Anthrenus larvae (Coleoptera: Dermestidae). Detectives asked what this meant, and it was concluded that the body had initially been outdoors, in a warm sunny situation, having been moved later to its present location. Lucilia oviposits on high temperature surfaces. The combination of dead Lucilia adults and stage 3 Anthrenus larvae indicates a PMI of several weeks.

The estimation of a PMI of two weeks based on the presence of full-grown Calliphora larvae and pupal remains, agreeing with approximate date last seen alive by his partner of a murder victim:

In July, a male corpse, shot through the head, was found under scrub along a motorway parking place by passing tourists making a sanitary stop at night. The next day the pathologist sampled insects, including full-grown larvae and pupal remains of Calliphora. This indicates a PMI of about two weeks. Identification of the victim proved difficult, but after having traced his address in neighbouring Belgium it was established that he was last seen alive about 15 days previously.

Skeleton with loose skull, found in winter, with presence of numerous empty pupal cocoons of Protophormia under the bones, indicative of long-term in situ decomposition of the deceased:
Human remains, almost completely skeletonized, were found in a small forest in January, not far from the forest edge. The skull was detached from the rest of the skeleton, and dogs or foxes may be responsible for this. Saprophagous isopods and millipedes were present, and hand sifting of the soil under the skeleton turned up numerous empty pupal cocoons of *Protophormia*, a blowfly pupating on or close to a corpse, contrary to *Calliphora* and *Lucilia*. It was concluded that the PMI must go back to at least the previous summer, amounting to 6 months or more, and that the whole decomposition process had taken place on that same spot.

*Unusual absence during warm summer of mass development of blowflies on victim’s body, indicative of a delayed infection of flies:*

The prosecution delivered to us an autopsy report describing a female corpse beyond the rigor mortis stage, found in an exposed situation during summer, at the edge of a lake. Drowning was certainly not the cause of death. A few insect eggs, most probably of blowflies, stuck to her hairs, as was also visible on autopsy photographs. The question was: could we add anything to the reconstruction of the case? The total absence of maggots on the body was considered odd, and inconsistent with the approximate lapse of time implied by the pathologist. It was concluded that the body must have been sheltered, delaying the colonization by blowflies. This was confirmed. Witnesses declared having seen the suspect, who had been released from custody, moving a container to the spot where the victim was found.

**CONCLUSIONS**

In The Netherlands forensic entomology is increasingly seen as a useful tool in reconstructing the history of corpses found on crime scenes. The principles found in the rapidly developing body of literature can be applied with equal success, but there is scope for refinement and regional calibration. The data used in the interpretation of the PMI stem from few original sources, experimental research is still limited, and our biological knowledge of the lesser known carrion insect species leaves much to be desired.

**REFERENCES**


