Forensic entomology in Germany

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Abstract

Forensic entomology (FE) is increasingly gaining international recognition. In Germany, however, the development of FE has been stagnating, mainly because of the lack of cooperation between police, forensic medicine and entomology. In 1997 a co-operative research project ‘Forensic Entomology’ was started in Frankfurt/Main at the Center of Legal Medicine and the Research Institute Senckenberg. The aim of this project is to establish FE in Germany as a firmly integrated component of the securing of evidence from human cadavers in cases of suspected homicide. For this purpose we developed a forensic insect collecting kit, and policemen are educated for greater acceptance and better application of FE. The scientific programme focusses on the investigation of the insect succession on cadavers in urban and rural habitats. This also includes new indicator groups (e.g. parasitic wasps) for a more precise calculation of the late post mortem interval. Recently a DNA-based reliable and fast identification method especially for the immature stages of necrophagous insects became part of the project. Preliminary results are reported and two case studies presented.

Keywords: Forensic entomology; Post mortem interval; Case studies

1. Review: the historical process of establishing forensic entomology in Germany

Forensic entomology – the use of insects and other arthropods in forensic investigations – has gained a lot of importance during the last 10–15 years. The Manual of Forensic Entomology by Smith [1], the entomological review by Catts & Goff [2], the installation of the International Homepage of Forensic Entomology [3] and the First International Seminar on Forensic Entomology held in Bari/Italy in 1998 may be mentioned as examples. In Germany, however, many possibilities of that discipline have hardly ever been used up to this day [4–6]. Independent research on forensic entomology seemed to be simply out of the question. The situation did not look so bad in the early 19th century, when the development of modern medicine also led to initial stages of forensic entomology in Germany [7]. Mende, for example, listed a number of animals which feed on corpses [8]. This list includes flies, beetles and other insects. Krahmer for the first time defined the possibilities and limits of FE by stating...
that rather than the decomposition of a corpse the
time of development of necrophagous insects was
calculable [9]. One important paper by Megnin [10]
provoked a stronger interest in FE also in Germany.
Nonetheless the rules mentioned in this work did not
withstand close examination and were therefore often
criticised – with justification – by German authors,
e.g. Dahl [11]. Although these authors supplied a
basis for independent examinations within FE in
Germany, ecological research was still scarcely
developed and consequently not in a position to offer
satisfactory data. Another reason for stagnation was
the lack of co-operation between ecological-oriented
entomologists and forensic examiners. This draw-
back continues on to the recent past and thus often
led to uncertainty in the determination of post
mortem intervals (PMI) by means of insects [12],
even though ecological as well as taxonomical works
already existed [13,14]. The papers by Reiter [4,15]
resulted in stronger attention to FE in the German-
speaking area and Benecke [5,6,16] raised the
knowledge of the subject further with case descrip-
tions as well as general articles. Finally, an
independent research project ‘Forensic Entomology’
was established in Frankfurt in 1997. This project,
founded in co-operation between the Center of Legal
Medicine and the Research Institute Senckenberg, is
supposed to fill a great gap. Right from the begin-
ning the research project had two main intentions.
First of all the acceptance and application of FE
should be raised. For that aim the project offers
entomology training for policemen and forensic staff.
Additionally, instructions and collection kits for
collecting insects at the death scene have been
developed [17]. Secondly, the project started a basic
research programme to increase our knowledge about
the necrophagous arthropods in Germany.

2. Research

Three important topics can be considered:

1. Examination of arthropod succession on a corpse

2. Reliable and fast identification of all stages of
development of necrophagous insects relevant for
Germany by DNA-analysis

3. Establishment of new indicator-species for the
determination of post mortem interval

2.1. Examination of arthropod succession on a
corpse

The different stages of putrefaction of a corpse
vary in attraction to necrophagous insects, so that
the dead body is characterised by a typical cadaver-fauna
depending on its prevailing conditions of decay [18].
By analysing the stages of succession a first rough
estimation of the PMI can be made [2]. But since the
putrefaction is highly dependent on exposition of the
body, climatical influences, etc., the rate of decay of
the corpse can vary considerably [19]. This is also
reflected in the insect population and may lead to
inaccurate results, when the determination of the
PMI is based exclusively on the stages of succession
found on the corpse [20]. Depending on the biogeog-
raphical region and ecological habitat, different
species of insects are involved in the decay of a
corpse. As a result, examinations on insect-succe-
sion from, e.g. Canada [21], are not applicable to the
conditions in Germany. The variety of possible
places and situations in which a body may be found
needs to be taken into account. It is easy to imagine
that the insect fauna of a corpse found in a dune area
differs fundamentally from communities of nec-
rophagous arthropods in woods. Only little is known
about the biology and ecology of necrophagous
insects of, for example, drowned bodies [22], or the
insects on cadavers during winter [23].

The main aim of this part of the research project is
the observation of insect-succession on corpses in
German urban and rural habitats.

Data are gathered from two different sources:

- Insects from human corpses

  From human bodies, of which time of death
  and other important circumstances are known,
  all species of insects in all developmental
  stages are collected. Conspicuous patterns of
  necrophagous population development and
  succession on the cadaver are considered as
  well as the surroundings (such as apartment,
ground, etc.) from which samples are taken.

- Insects from animal carcasses

  Data are also gathered from dead animals. So
  far only cadavers could be brought in. Experi-
  ments under controlled conditions with freshly
  killed animals are in preparation. Here data
  can be obtained during a completely super-
  vised putrefaction-process.
2.2. Reliable and fast identification of all stages of development of necrophagous insects in Germany by DNA-analysis

Identification-keys for necrophagous insects are based mainly on morphological characters of adult animals. To work with these keys requires entomological knowledge and appropriate taxonomical experience. Despite the existence of these keys only a few specialists are able to determine necrophagous insects correctly, which is indispensable in forensic investigations. Furthermore it is necessary to rear the larvae to gain the adult animals needed for determination. This demands adequate logistics and appropriate care for successfully rearing the larvae. Works concentrating on larvae or pupae of forensic relevance are rare [1,14,15] and often of limited applicability for the middle European area [24]. Many of the early larval stages (L1 and L2) are hardly discriminable between species and are therefore in many cases not determinable.

Both problems – the complicated assignment to a species and the often delayed identification after rearing larvae to adult insects – can be avoided by using molecular-biological methods in determining species [25,26]. Therefore, extensive sequence-analysis of relevant genes (Cytochrom-oxidase, Cytochrom b) of forensically relevant fly-species in Germany is intended to facilitate a fast and reliable determination of all stages of development. Moreover, a classification of species from insect fragments, such as empty pupae, has to be developed. Preliminary examinations were made in the laboratory to solve methodological questions: DNA-extraction, testing of primers, optimizing of amplification and preliminary sequencing analysis. These data show that until now it is possible to differentiate between the necrophagous flies Calliphora vicina, Calliphora vomitoria, Lucilia sericata and Lucilia caesar by sequence analysis of subunit I and II of cytochrome-oxidase and of cytochrome b [27].

2.3. Establishment of new indicator-species for the determination of post mortem interval

For the estimation of the PMI, attention so far has been mainly paid to blowflies, which are the first to appear on cadavers. However, further, hitherto neglected groups of insects with considerable potential for FE are existing. As an example, the possible use of so called late colonizers such as different beetles or cheeseskippers on heavily decayed corpses has not yet been adequately examined. In addition, insects which develop parasitically on necrophagous insects have barely been taken into account. These so called parasitoids are insects which, in contrast to real parasites, always kill their host. Depending on the parasitoid’s species and biology – usually Hymenoptera of the superfamilies Chalcidoidea and Ichneumonoidea – eggs, larvae or pupae of the host are being attacked. Especially pupal parasitoids of blowflies could be suitable to assessing the PMI (Fig. 1): They usually parasitize a closely defined stage of development of the host-pupa, for example a two- or three-day-old puparium. With knowledge of the biology and period of development of the parasitoid and its host, the PMI can be determined exactly by the addition of their respective periods of development. The first period to analyse is the development of the host from egg to the moment of parasitation, the second period is the parasitoids development from egg to adult. During the current research project, for the first time a PMI could be determined with the aid of the parasitoid Nasonia vitripennis (Hymenoptera: Pteromalidae). Its pupae could still be collected on the corpse when all initial colonizers (such as the host, the blowfly Protophormia terraenovae) had already left the corpse.

3. Case studies

Despite many case-reports which show the useful power of forensic entomology [2,19], there are also situations, which demonstrate the lack of a well coordinated examination between all parties involved in the securing of a crime scene. The following cases may illustrate this experience.

3.1. Case 1: corpse in a shaft

In mid-May a female body was found in a shaft over 2 m deep. The ground of the shaft was covered with soil, the opening closed with a steel top. The body had been wrapped in plastic sacks and stuffed into a travelling bag. The autopsy produced no maggots from the cadaver, but four third-instar maggots of the blowfly Calliphora vicina Robineau-
Fig. 1. Date of hatching of the blowflies *Lucilia sericata* and *Calliphora vomitoria* (Diptera: Calliphoridae), feeding on a dead rat, and the parasitic wasp *Alysia manducator* (Hymenoptera: Braconidae), which parasitize young pupae of the mentioned blowflies. Note, that the parasitoid is still recordable (in the pupae of its hosts) when the necrophagous blowflies already had left the death scene. A insect collection, e.g. on the 8th of October would not find the first colonizer. The consideration of the parasitoids could prolong the minimum PMI in that case.

Desvoidy (Diptera: Calliphoridae) were found in the plastic sacks. A soil sample from the shaft contained about 100 pupae and 24 maggots of the same blowfly species and 20 larvae (L3-stage) of the Sphaerocerid fly *Leptocera caenosa* Rondani (Diptera: Sphaeroceridae). Based on the rate of development of *C. vicina* under the known circumstances a minimum PMI of 44 days was calculated. Unlike *C. vicina*, which appears very early on a cadaver, the dung-fly *L. caenosa* colonises cadavers only after they have reached an advanced state of decomposition. Furthermore, *L. caenosa* exhibits a preference for artificial and natural cavities like caves or cellars. Investigations by Fredeen and Glen [28] have shown that at low temperatures the developmental time of that species increases disproportionately. Our calculations led to an estimate of 30 days for the period from oviposition to the development of fully grown maggots ready to pupate. The life habits of *L. caenosa* suggest that colonisation of the dead body had taken place after its deposition in the shaft. The cool temperature (about 12°C) in the shaft and the preference of *L. caenosa* for advanced states of decomposition suggests an even longer period since deposition of the body, but a perfectly accurate delimitation was not possible. The criminal investigations eventually revealed that the body had been inside the shaft since the beginning of March.

### 3.2. Case 2: dead body in a flat

On August 12th the body of a woman with stab wounds was found in a flat. The autopsy produced numerous third-instar maggots of the blowfly *Lucilia sericata* (Meigen) (Diptera: Calliphoridae). The calculation of the time since deposition of the body based on the development data available for this fly yielded 8 days. During inspection of the flat an additional 20 pupae of the blowfly *Calliphora vicina* and several third-instar maggots of the flesh fly *Liopygia argyrostoma* Robineau-Desvoidy (Diptera: Sarcophagidae) were secured. The calculations for *C. vicina* suggested a first oviposition of this species on August 2nd. The flesh fly *L. argyrostoma* lives on a
wide variety of decomposing substrates [29]. It is one of the sarcophagids which colonise cadavers only after blowflies have already begun to develop and thus is a typical representative of the second wave of colonisation (Povolný, personal communication). A biological peculiarity of *L. argyrostoma* is its ovoviviparous development, i.e. the females do not lay eggs but so called egg-larvae which immediately begin their larval development. The investigations showed that egg-larvae deposition had presumably taken place 6 days before the securing of the cadaver.

The different presumed oviposition dates of the various fly species may be explained by their different biologies. *C. vicina* colonises cadavers as early as a few hours after death, therefore the period between oviposition of this species and the discovery of the dead body comes closest to the actual period it has been lying at the place. *L. sericata*, in contrast, may appear at the body up to 2 days after death [1], *L. argyrostoma* only 3–5 days after *C. vicina* (Povolný, personal communication) (Fig. 2). These data support the estimate for the PMI of about 10 days. Criminal investigations confirmed that finding.

Both case studies clearly show that in many cases a more exact assessment of the PMI of cadavers would be possible, if the collecting of insects at the cadaver *and* the death scene would be performed in a co-ordinated and standardised way to prevent the loss of essential information.

### References


