1

The breadth of forensic entomology

Forensic entomology is the branch of forensic science in which information about insects is used to draw conclusions when investigating legal cases relating to both humans and wildlife, although on occasion the term may be expanded to include other arthropods. Insects can be used in the investigation of a crime scene both on land and in water (Anderson, 1995; Erzinçlioğlu, 2000; Keiper and Casamatta, 2001; Hobischak and Anderson, 2002; Oliveira-Costa and de Mello-Patiu, 2004). The majority of cases where entomological evidence has been used are concerned with illegal activities which take place on land and are discovered within a short time of being committed. Gaudry *et al.* (2004) commented that in France 70 % of cadavers were found outdoors and of these 60 % were less than 1 month old.

The insects that can assist in forensic entomological investigations include blowflies, flesh flies, cheese skippers, hide and skin beetles, rove beetles and clown beetles. In some of these families only the juvenile stages are carrion feeders and consume a dead body. In others both the juvenile stages and the adults will eat the body (are necrophages). Yet other families of insects are attracted to the body solely because they feed on the necrophagous insects that are present.

1.1 History of forensic entomology

Insects are known to have been used in the detection of crimes for a long time and a number of researchers have written about the history of forensic entomology (Benecke, 2001; Greenberg and Kunich, 2002). The Chinese used the presence of flies and other insects as part of their investigative armoury for crime scene investigation and instances of their use are recorded as early as the mid-tenth century (Cheng, 1890; cited in Greenberg and Kunich, 2002). Indeed, such was the importance of insects in crime scene investigation that in 1235, a training manual on investigating death, *Washing Away of Wrongs*, was written by Sung Tz'u. In this medico-legal book it is recorded that the landing of a number of blowflies on a particular sickle caused a murderer to confess to murdering a fellow Chinese farm worker with that sickle.

Between the thirteenth and the nineteenth century, a number of developments in biology laid the foundation for forensic entomology to become a branch of scientific study. The two most notable were, perhaps, experiments in Italy by Redi (1668) using the flesh of a number of different animal species, in which he demonstrated that larvae developed from eggs laid by flies, and the work by Linnaeus (1775) in developing a system of classification. In so doing, Linnaeus provided a means of insect identification, including identifying such forensically important flies as *Calliphora vomitoria* (Linnaeus). These developments formed foundations from which determination of the length of the stages in the insect's life cycle could be worked out and indicators of time since death could be developed.

A particularly significant legal case, which helped establish forensic entomology as a recognized tool for investigating crime scenes, was that of a murdered newborn baby. The baby's mummified body, encased in a chimney, was revealed behind a mantelpiece in a boarding house when renovation work was being undertaken in 1850. Dr Marcel Bergeret carried out an autopsy on the body and discovered larvae of a fleshfly, *Sarcophaga carnaria* (Linnaeus), and some moths. He concluded that the baby's body had been sealed up in 1848 and that the moths had gained access in 1849. As a result of this estimation of post mortem interval, occupiers of the house previous to 1848 were accused and the current occupiers exonerated (Bergeret, 1855).

The next significant point in the history of forensic entomology resulted from observations and conclusions made by Mégnin (1894). He related eight stages of human decomposition to the succession of insects colonizing the body after death. He published his findings in *La Faune des Cadavres: Application de l'Entomologie à la Médicine Légale*. These stages of decomposition were subsequently shown to vary in speed and to be dependent upon environmental conditions, including temperature and, for example, whether or not the corpse was clothed. However, the similarity in overall decomposition sequence and the value of the association of insects has been demonstrated for decomposition of the bodies of a number of animal species. This knowledge about insect succession on a corpse became the basis for forensic entomologists' estimations of the time since death of the corpse.

In the twentieth century insects were shown to be of value in court cases involving insect colonization of body parts recovered from water and not just whole corpses found on land. On 29 September 1935, several body parts, later identified as originating from two females, were recovered from a Scottish river near Edinburgh. The identities of the deceased were determined and the women were named as Mrs Ruxton and Mary Rogerson, 'nanny' for the family. The presence of larvae of the blowfly *Calliphora vicina* Robineau-Desvoidy, in their third larval instar, indicated that the eggs had been laid prior to the bodies being dumped in the river. This information, combined with other evidence, resulted in the husband, Dr Ruxton, being convicted of the murder of his wife and Mary Rogerson.

The acceptance of forensic entomology has depended upon both academics and practitioners working alongside the police and legal authorities, to refine and develop forensic entomology as a scientific study in the late twentieth and early twenty-first centuries. A list of forensic entomologists, who are members of the American Board of Forensic Entomology, the European Association for Forensic Entomology and other professional entomological and medical organizations, can be found on the website: http://www.forensic-entomology.info/forens_ent/forensic_entomologists.shtml

1.2 Indicators of time of death

In the first 72 hours after death, the pathologist is usually considered to be able to provide a reasonably accurate determination of the time of death. Historically, this has been based upon the condition of the body itself and such features as the fall in body temperature. Beyond this time, there is less medical information with which to correlate post mortem interval (PMI). So another area of expertise is required to clarify time of death. The forensic entomologist can provide a measure of the possible post mortem interval, based upon the life cycle stages of particular fly species recovered from the corpse, or from the succession of insects present on the body. This estimate can be given over a period of hours, weeks or years. The start of the post mortem interval is considered to coincide with the point when the fly first laid its eggs on the body, and its end to be the discovery of the body and the recognition of life stage of the oldest colonizing species infesting it. The duration of this stage, in relation to the particular stage of decay, gives an accurate measure of the probable length of time the person has been dead and may be the best estimate that is available.

1.3 Stages of decomposition of a body

The stages of decomposition of a body have been a topic of interest for both artists and scientists over a long period of time (Figures 1.1–1.8). There are three recognizable processes in corpse decomposition. These are autolysis, putrefaction and skeletal bone decomposition (diagenesis). In autolysis, a process of natural breakdown, the cells of the body are digested by enzymes, including lipases, proteases and carbohydrases. This process can be most rapid in organs such as the brain and liver (Vass, 2001). A ‘soup’ of nutrients is released which forms a food source for bacteria. Putrefaction is the breakdown of tissues by bacteria. As a result, gases such as hydrogen sulphide, sulphur dioxide, carbon dioxide, methane, ammonia, hydrogen and carbon dioxide are released. Alongside this, anaerobic fermentation takes place when the volatiles propionic and butyric acid are formed. The body undergoes active decay, in which protein sources are broken

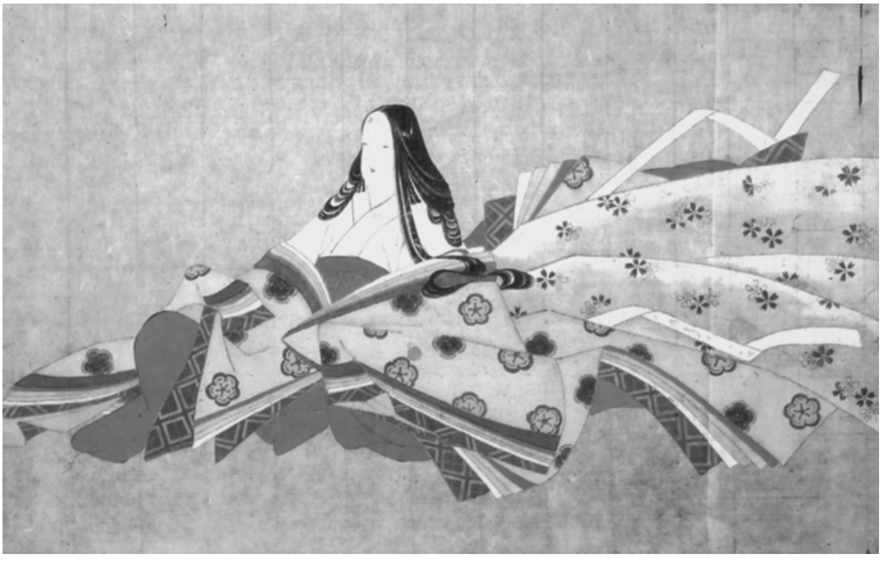

Figure 1.1 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.2 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.3 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.4 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.5 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.6 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.7 Artistic impressions of stages of decomposition of the body (Morishige, 1673–1680). The nine contemplations of the impurity of the human body, stage 1–9. Reproduced with permission of the Etnografisch Museum, Antwerp, Belgium inv.nrs: AE 4552 1/20–19/20 (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

Figure 1.8 Last stage of decomposition of a human body (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

down into fatty acids by bacteria (Vass, 2001). Fatty acids and such compounds as skatole, putrescine and cadaverine are significant members of these decomposition products (although Vass *et al.*, 2004, commented on their absence from recovered volatiles from buried bodies).

When the soft tissue is removed, skeletal material – organic and inorganic remains – are further broken down by environmental conditions and are finally reduced to components of the soil. The rate of decomposition is temperature-dependent. A formula has been proposed by forensic pathologists to estimate the time of body decomposition to a skeleton, in relation to temperature (Vass, 2001). The formula is:

$$Y = 1285/X$$

where Y is the number of days to mummification, or skeletonization, and X is the average temperature for the days before the body was found (Vass *et al.*, 1992).

1.3.1 On land

The body can be allocated to one of five recognizable post mortem conditions, which can be linked to the eight waves of arthropod colonization proposed by Mégnin (1894). No distinction from one stage to the next is obvious and Gaudry (2002), on the basis of 400 cases, considers Mégnin's first two waves to be one. Although no stage has a fixed duration, each stage can be associated with a particular assemblage of insects. The profiles of insects would appear to be universal, although the majority of research on this aspect has, until recently, been undertaken in North America (Hough, 1897; Easton and Smith, 1970; Rodriguez and Bass, 1983; Catts and Haskell, 1990; Mann, Bass and Meadows, 1990; Goff, 1993; Dillon and Anderson, 1996; VanLaerhoven and Anderson, 1999; Byrd and Castner, 2001). These stages of post mortem change are:

- *Stage 1: Fresh stage.* This stage starts from the moment of death to the first signs of bloating of the body. The first organisms to arrive are the blowflies (the Calliphoridae). In Britain these are usually *Calliphora vicina* or *Calliphora vomitoria* Linnaeus, or in early spring may be *Protophormia* (= *Phormia*) *terraenovae* Robineau-Desvoidy) (Nuorteva, 1987; Erzinçlioğlu, 1996).
- *Stage 2: Bloated Stage.* Breakdown of the body continues because of bacterial activity, or putrefaction, and this is perhaps the easiest stage to distinguish. Gases causing the corpse to bloat are generated through metabolism of nutrients by anaerobic bacteria. Initially the abdomen swells but later the whole body becomes stretched like an air-balloon (Figure 1.9). At this stage more and more blowflies are attracted to the body, possibly in response to the smell of the breakdown gases. Vass *et al.* (1992, 2004) studied the odours emanating from dead bodies that were both resting on the surface and had been buried. Their work provides clarification of the identity of some of these gases and the information supplements that provided by Mégnin (1894); Hough (1897) and Smith (1986).

  Rove beetles (Staphylinidae) may be attracted to the body at the bloat stage because of the 'ready meals' of eggs and maggots. These and other predators

Figure 1.9 Body in bloat (A colour reproduction of this figure can be found in the colour section towards the centre of the book)

can affect the interpretation of the range of insects and insect life stages present as they feed on larvae or remove puparia (Smith, 1986).

- *Stage 3: Active decay stage.* This stage is recognizable by the skin of the corpse breaking up and starting to slough from the body. This sloughing allows the decomposition gases to escape and so the inflation of the body gradually subsides as putrefaction continues. In the later stages of putrefaction fermentation occurs and butyric and caseic acids are generated. This is followed by a period of advanced putrefaction, which includes ammoniacal fermentation of the body, to which a different cohort of insects are attracted. These include the silphid beetle *Nicrophorus humator* (Gleditsch) the histerids *Hister cadaverinus* (Hoffmann) and *Saprinus rotundatus* Kugelann, and the muscid fly *Hydrotaea capensis* Wiedeman (= *Ophyra cadaverina* Curtis).
- *Stage 4: Post-decay stage.* In the later stages of decay, all that remains of the body are skin, cartilage and bones with some remnants of flesh including the intestines. Any remaining body tissue can be dried. The biggest indicator of this stage is an increase in the presence of beetles and a reduction in the dominance of the flies (Diptera) on the body.
- *Stage 5: Skeletonization.* At this stage the body is only hair and bones (Figure 1.10). No obvious groups of insects are associated with this stage, although beetles of the family Nitidulidae can, in some instances, be found. The body has clearly reached its final stage of decomposition. Any further breakdown is best described in terms of the decay of individual components of the body, such as the bones of the feet and legs, the skull and the ribs.

Figure 1.10 Post-decay stage of human decomposition. The breakdown material was retained within the polythene

1.3.2 Submerged in water

In water these same five stages still occur along with an additional stage. This additional stage is the floating decay stage, where the body rises to the water surface. At this point, besides aquatic insects such as midge (chironomid) larvae and invertebrates such as water snails, terrestrial insect species also colonize the body.

This stage is the most obvious stage and tends to be the point at which a body is noticed and recovered from the water. The period of time after death when this takes place will depend on the temperature of the water.

The relationship between time of death and physical breakdown of the body has been investigated by Giertsen (1977). He cited Casper's Dictum as a means of determining the length of the post mortem interval. This rule says that:

> '. . . at a tolerable similar average temperature, the degree of putrefaction present in a body lying in the open air for one week (month) corresponds to that found in a body after lying in the water for two weeks (months), or lying in the earth in the usual manner for eight weeks (months)'.

The reason for this difference in decomposition is that the speed at which the body loses heat in water is twice the speed at which the body will lose heat in air.

Box 1.1 Hint

Besides skeletonization, with the resultant change in the bone structure (diagenesis), two other outcomes of the decomposition process may occur. These are mummification and the generation of 'grave wax' or adipocere.

Diagenesis

When the body reaches the skeletal stage, changes to the bone called diagenesis occur. Diagenesis is defined, in chemical terms, according to *Collins Dictionary of the English Language* (Hanks, 1984), as recrystallization of a solid to form large crystal grains from smaller ones. The changes in the bone structure depend upon the breakdown of the soft tissue. This is affected by the nature of the death and subsequent treatment of the body, including the type of environment in which the body is buried.

Investigating bone can tell us about the latter stages of post mortem change because a number of features can be quantified. The amount of post mortem change can be estimated if the bone histology is investigated under the microscope, the degree of bone porosity is determined; the carbonate and protein content of the bone are calculated; the crystalline nature and content of the bone mineral made of calcium fluorophosphate or calcium chlorophosphate (apatite) is examined, and which components have leached out of or into the bone are determined.

Insect attack, both before and after the body is buried, has a role to play in causing change to the environment and hence bone diagenesis.

Adipocere

If the body is in an environment which combines a high humidity with high temperatures, the subcutaneous body fat of the face, buttocks (breasts in the female) and the extremities become hydrolysed. Fatty acids are released. These form food for bacteria, which can speed up the rate at which adipocere is made. For example, *Clostridium* bacteria will convert oleic acid (a fatty acid) into hydroxystearic acid and oxostearic acid.

Two types of adipocere are found, depending on whether the fatty acids combine with sodium or with potassium. If sodium from the breakdown of intercellular fluid is bound to the fatty acids, the adipocere is hard and curly. Where the cell membranes break down and potassium is released, a softer adipocere results, which is often termed 'pasty'. An indication of submergence in cold water is the uniform cover of adipocere over the body (Spitz, 1993).

Mummification

If water is removed from skin and tissue, that tissue becomes desiccated and mummification will occur. This happens particularly where a body is kept in an environment with a dry heat, little humidity and where the airflow is good. Chimneys are examples of good locations with these features. In mummified corpses in temperate conditions, the extremities become shrivelled and the skin tends to be firm but wrinkled and to have a brown colouration. The internal organs, such as the brain, will decompose during mummification.

More research is needed to explore decomposition in various types of water body and in a number of locations, so that a comprehensive picture of the potential indicators of submerged post mortem interval can be clarified. Research by Keiper and Casamatta (2001), Hobischak and Anderson (2002) and Merritt and Wallace (2001) has provided a starting point.

Whilst the major contribution of forensic entomology to solving crimes could be considered to be in relation to suspicious death, it has a part to play in investigating other crimes in which the victims may be alive or dead.

Figure 1.11 Rabbit exhibiting myiasis. Reproduced with kind permission of Frances Harcourt-Brown

1.4 Indicators of physical abuse

Insects are of value as forensic indicators in cases of neglect or abuse. Some insects, for example the greenbottle *Lucilia sericata* (Meigen), are attracted to odours, such as ammonia, resulting from urine or faecal contamination. Adult flies of this species tend to be attracted to an incontinent individual; a baby that has not had its nappy changed sufficiently often, or incontinent old people who have not been assisted in maintaining their bodily hygiene.

Flies may lay their eggs in clothing or on skin. These eggs, if undiscovered, will hatch into maggots (larvae) which start feeding upon flesh, or on wounds, ulcers or natural entry points of the body. Over time the flesh will be eaten away and the region may be further infected by bacteria as well as being invaded by other insects.

Such an insect attack can also happen to animals. In particular, rabbits, pigs, dogs and sheep can be the victims of fly strike (Figure 1.11) because of urine or faecal material attached to their fur, fleece or hind quarters through neglect, poor caging and living conditions or ill-health reflected by ‘scouring’. Such cases are considered to be instances of physical abuse, since victims are unable to remove the eggs or maggots themselves. The results can be serious, requiring attention

Box 1.2 Hint

The invasion of living tissue by insects is also of concern to the forensic entomologist. This invasion is called *myiasis* and becomes relevant where cases of misuse and abuse are involved.

Myiasis has been defined according to two criteria: the biological requirements of the fly, or where the flies attack the organism, be it human or animal. James (1947) defined biological myiasis as invasion of tissue or organs of man or animals by dipterous larvae. He acknowledged Patton’s (1922) earlier views that the presence of eggs, pupae or adults might be included, but considered that the larval stage was the ‘active stage’ of myiasis.

In medical terms, myiasis can be defined according to the location of the fly infestation. For example, it can be defined as: wound myiasis; myiasis of the nose, mouth and accessory sinuses; aural myiasis; ocular internal and external myiasis; myiasis of the rectal region and vagina; myiasis of the bladder and urinary passages; furuncular, dermal and sub-dermal myiasis; creeping dermal, sub-dermal myiasis or enteric myiasis.

Flies such as *Lucilia sericata*, *Musca domestica* Linnaeus and *Phormia regina* Meigen, the initial colonizers of the body, have all been implicated in cases of myiasis.

from veterinary surgeons and even leading to the death of the animal, or requiring its euthanasia.

Care however, has to be taken in making assumptions about the existence of physical abuse or assault prior to death. Work by Komar and Beattie (1998) in studies on dressed pigs, showed the effect of bloat was to cause the same disturbance and tearing of clothes which are characteristic of sexual assault. They considered that maggot masses were particularly important in deriving such changes to clothing.

1.5 Insect larvae: a resource for investigating drug consumption

The insect life cycle stage that feeds on the cadaver is a potential reservoir of undigested flesh from the corpse. Because, in some circumstances, the flesh from the corpse can retain some types of drugs that had been consumed by the victim before he/she died and which may even have been the cause of death, these drugs may be recovered by analysing the insects and may include opiates (Introna *et al.*, 1990), the barbiturate phenobarbital, benzodiazepines or their metabolites, such as oxazepam, triazolam, antihistamines, alimemazine and chlorimipramine, a tricyclic antidepressant (Kintz *et al.*, 1990; Sadler *et al.*, 1995).

To date there is not a great deal of information available that indicates the role of drugs, which are present in decomposing body tissue, on necrophagous larvae. Musvaska *et al.* (2001) examined the effects of consuming liver containing either a barbiturate (sodium methohexital) or a steroid (hydrocortisone) on the development of a fleshfly, *Sarcophaga* (= *Curranea*) *tibialis* Macquart. They showed that, compared with controls, the length of the larval stage was increased, whilst pupariation was more rapid. In laboratory experiments investigating the effects of heroin, Arnaldos *et al.* (2005) also showed that the length of time taken to complete individual larval stages in *Sarcophaga tibialis* was considerably longer, in contrast to those larvae which were not fed heroin.

However, heroin has been shown to increase the rate at which other species of maggots (e.g. *Boettcherisca peregrina* Robineau-Desvoidy) grow, whilst increasing the duration of pupal development (Goff *et al.*, 1991). Cocaine and one of its breakdown products has been found in small amounts in the puparium of Calliphoridae (Nolte *et al.*, 1992), so this drug is clearly sequestered in the larval body and retained in the next life stage. However, Hédouin *et al.* (2001) only showed a correlation between concentration of morphine in body tissues and that in the tissue of larvae of *Protophormia terraenovae* and *Calliphora vicina* in the third instar. In *Lucilia sericata* they found that the post mortem interval could, in reality, be 24 hours longer than expected (Bourel *et al.*, 1999).

Suicide can be investigated using forensic entomology. By analysing the maggots which had fed on the corpse and demonstrating the presence in the body of malathion, an organophosphate insecticide, Gunatilake and Goff (1989) confirmed that a 58 year-old man had committed suicide. A bottle of malathion had been found near to the corpse.

Miller *et al.* (1994) analysed chitinized insect tissue from mummified remains from which the normal toxicological sources were absent. They were able to recover amitriptyline and nortryptyline from the puparia of scuttle flies (Phoridae) and the exuviae of hide beetles (Dermestidae). Sadler *et al.* (1997), however, found that there was a variation in larval drug accumulation of amitriptyline and urged caution in directly relating the concentration harvested from the larvae to concentrations in the original source.

Paying attention to the facts known about the lifestyle of the victim may assist in interpreting the post mortem interval, using the developmental stage of the insect recovered from the body. So, all of the information known about the crime scene and pre mortem behaviour of the person should be taken into account when investigating the entomological evidence.

Insects collected with plant material destined to be used illegally can indicate the part of the world from which the plants originated. This information may be of forensic value to Customs and Excise Officers. For example, in two separate drug seizures in New Zealand, cannabis was apprehended along with eight Asian species of beetles, as well as wasps and ants. The beetles were identified by Dr Trevor Crosby as belonging to the families Carabidae, Bruchidae and Tenebrionidae. By looking at the geographic distribution of all of the insects and the level of overlap of their distributions, entomologists concluded that the cannabis came from the Tenasserim region, between the Andaman Sea and Thailand. One of two suspects confessed on the basis of this evidence (Crosby *et al.*, 1986).

1.6 Insect contamination of food

Many societies consume insects as part of their diet (Figure 1.12). For example, aquatic beetles such as the giant water bug, *Lethocerus indicus* Lepeletier Serville, are eaten as a delicacy across south-eastern Asia. Chocolate-covered bees have been sold in speciality shops in the UK, and in North America some shops sell canned, fried grasshoppers (DeFoliart, 1988; Menzel and D'Aluisio, 1998), whilst Thai cooked crickets in tins are available via the world-wide web.

However, the presence in food of insects that are eaten unintentionally, or could be eaten along with the food, is considered unacceptable to the consumer and a source of contamination. For example, the saw-toothed grain beetle, a stored product pest, may be found in cereal packages; wire worms may be sold along with freshly cut lettuces, or may be processed into lettuce and tomato sandwiches; whilst in many countries, fish and meat which is left in the open to dry can become infested with beetles or flies, either in the drying process or later on a market stall. These are then eaten and have the potential to cause illness. Forensic entomologists may therefore find themselves being asked for an expert opinion in civil cases

Figure 1.12 A tequila bottle label illustrating the Maguey worm, *Aegiale hesperiatis* Walker (Lepidoptera), which authenticates the drink. Reproduced from a letter in *Antenna* **6**(3) (1982) with kind permission of Dr B. Lawrence and the Royal Entomological Society of London

relating to the food industry, where food has been contaminated by insects living in close association with man (such insects are described as ‘synanthropic’).

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Useful websites

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