FORENSIC ENTOMOLOGY: THE USE OF INSECTS IN DEATH INVESTIGATIONS

FUNCTIONAL COMPETENCIES

- ☐ **A**cquiring and Analyzing Information - Conducting Investigations and Collecting Evidence [Collecting Information]
- ☐ **P**artnership - Inter/Intra-Agency and Multi-Disciplinary Cooperation [Partners]

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INTRODUCTION

Forensic (or medico-legal) entomology(1) is the study of the insects associated with a human corpse in an effort to determine elapsed time since death. Insect evidence may also show that the body has been moved to a second site after death, or that the body has been disturbed at some time, either by animals, or by the killer returning to the scene of the crime.

However, the primary purpose of forensic entomology today is to determine elapsed time since death.

Forensic entomology was first reported to have been used in 13th Century China and was used sporadically in the 19th Century and the early part of the 20th Century, playing a part in some very major cases. However, in the last 15 years, forensic entomology has become more and more common in police investigations. Most cases that involve a forensic entomologist are 72 h or more old, as up until this time, other forensic methods are equally or more accurate than the insect evidence. However, after three days, insect evidence is often the most accurate and sometimes the only method of determining elapsed time since death.

There are two main ways of using insects to determine elapsed time since death :

1. using successional waves of insects
2. using maggot age and development.

The method used is determined by the circumstances of each case. In general, the first method is used when the corpse has been dead for between a month up to a year or more, and the second method is used when death occurred less than a month prior to discovery.

The first method is based on the fact that a human body, or any kind of carrion, supports a very rapidly changing ecosystem going from the fresh state to dry bones in a matter of weeks or months depending on geographic region. During this decomposition, the remains go through rapid physical, biological and chemical changes, and different stages of the decomposition are attractive to different species of insects. Certain species of insects are often the first witnesses to a crime. They usually arrive within 24 h of death if the season is suitable *i.e.* spring, summer or fall in Canada and can arrive within minutes in the presence of blood or other body fluids. These first groups of insects are the Calliphoridae or blowflies and the Muscidae or houseflies. Other species are not interested in the corpse when the body is fresh, but are only attracted to the corpse later such as the Piophilidae or cheese skippers which arrive later, during protein fermentation. Some insects are not attracted by the body directly, but arrive to feed on the other insects at the scene. Many species are

involved at each decomposition stage and each group of insects overlaps the ones adjacent to it somewhat. Therefore, with a knowledge of the regional insect fauna and times of carrion colonization, the insect assemblage associated with the remains can be analyzed to determine a window of time in which death took place. This method is used when the decedent has been dead from a few weeks up to a year, or in some cases several years after death, with the estimated window of time broadening as time since death increases. It can also be used to indicate the season of death *e.g.* early summer. A knowledge of insect succession is required for this method to be successful.

The second method, that of using maggot age and development can give a date of death accurate to a day or less, or a range of days, and is used in the first few weeks after death. Maggots are larvae or immature stages of Diptera or two-winged flies. The insects used in this method are those that arrive first on the corpse, that is, the Calliphoridae or blowflies. These flies are attracted to a corpse very soon after death. They lay their eggs on the corpse, usually in a wound, if present, or if not, then in any of the natural orifices. Their development follows a set, predictable, cycle.

The insect egg is laid in batches on the corpse and hatches, after a set period of time, into a first instar (or stage) larva. The larva feeds on the corpse and moults into a second instar larva. The larva continues to feed and develop into a third instar larva. The stage can be determined by size and the number of spiracles (breathing holes). When in the third instar, the larva continues to feed for a while then it stops feeding and wanders away from the corpse, either into the clothes or the soil, to find a safe place to pupate. This non-feeding wandering stage is called a prepupa. The larva then loosens itself from its outer skin, but remains inside. This outer shell hardens, or tans, into a hard protective outer shell, or pupal case, which shields the insect as it metamorphoses into an adult. Freshly formed pupae are pale in colour, but darken to a deep brown in a few hours. After a number of days, an adult fly will emerge from the pupa and the cycle will begin again. When the adult has emerged, the empty pupal case is left behind as evidence that a fly developed and emerged.

Each of these developmental stages takes a set, known time. This time period is based on the availability of food and the temperature. In the case of a human corpse, food availability is not usually a limiting factor.

Insects are 'cold blooded', so their development is extremely temperature dependent. Their metabolic rate is increased with increased temperature, which results in a faster rate of development, so that the duration of development decreases in a linear manner with increased temperature, and *vice-versa.*

An analysis of the oldest stage of insect on the corpse and the temperature of the region in which the body was discovered leads to a day or range of days in which the first insects oviposited or laid eggs on the corpse. This, in turn, leads to a day, or range of days, during which death occurred. For example, if the oldest insects are 7 days old, then the decedent has been dead for at least 7 days. This method can be used until the first adults begin to emerge, after which it is not possible to determine which generation is present. Therefore, after a single blowfly generation has been completed, the time of death is determined using the first method, that of insect succession.

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PROCEDURE

The first and most important stage of the procedure involved in forensic entomology involves careful and accurate collection of insect evidence at the scene. This involves a knowledge of the insects behaviour, therefore it is best performed by an entomologist. **I am always willing to come to a scene if it is possible**. Unfortunately, the entomologist is often not called until after the body has been removed from the death site. I usually see the remains at the morgue, and in some cases, do not actually see the remains at all, so my evidence is **dependent** on **accurate** collection by the investigating officers.

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COLLECTING, PRESERVING AND PACKAGING SPECIMENS

Collection

Samples of insects of all stages should be collected from different areas of the body, from the clothing and from the soil/carpet *etc*. Insects will often congregate in wounds and in and around natural orifices.

The two main insect groups on bodies are flies (Diptera) and beetles (Coleoptera).

Both types of insect look very different at different stages of their lives.

Flies can be found as:-

- **eggs** (in egg masses usually)
- **larvae or maggots** (in a range of sizes from 1-2 mm to 17 mm)
- **pupae and/or empty pupal cases**
- **adults.**

Eggs - are very tiny, but are usually laid in clumps or masses, and are usually found in a wound or natural orifice, but may be found on clothing *etc*. They can be collected with a child's paint brush dipped in water or with forceps. Half should be preserved in 75% alcohol or 50% isopropyl alcohol. The rest should be placed in a vial with a little damp tissue paper to prevent dehydration. If it will be more than a few hours before the entomologist receives them, they should also be given a small piece of beef liver. Make sure there is tissue or sawdust present if liver is added, to prevent drowning. They need some air. Newly emerged maggots can escape through holes, so a paper towel held over the top of the vial with a rubber band is excellent, as long as the vial stays upright! (No lid other than the paper towel is needed).

Maggots - collect a range of sizes. Maggots will be found crawling on or near the remains and may be in **maggot masses**. The masses generate a lot of heat, which speeds up development. Therefore, please note:-

- the site of maggot masses
- the temperature of each mass (thermometers can be acquired cheaply at drugstores) or if no thermometer available, please estimate size of mass.
- label which maggots come from a particular mass.

Large maggots are usually older so are most important, but smaller maggots may belong to a different species so both large and smaller maggots should be collected, with the emphasis on larger maggots.
Collect samples of maggots from different areas of the body and the surrounding area, and keep them separate.

As mentioned before, third instar larvae leave the food source to find a suitable area in which to pupate. They may wander some distance from the body so the soil for a metre or two around the body should be carefully sifted. Some may burrow down into leaf litter, so the soil below the corpse should be checked for several centimetres. If the remains were on a slope, the body fluids will seep downhill and insects will be found here, feeding on the fluids. This means that a very intensive search of the corpse, the clothes and the surrounding area must be made in order to get the entire picture.

When collected, a **proportion** of the larvae should be preserved **immediately** for two reasons. Firstly, to show the entomologist, if s/he is not present at the scene, what stage the larvae were when collected, as if they are then placed on meat, they will continue to develop, giving a misleading impression to the entomologist when they are examined. Secondly, to produce as evidence in court.
If there are lots of maggots on the body, preserve approximately half of all sizes.
If there are only 20-30, preserve 1 or 2
Preserve the specimens by immersing them in hot water for a few minutes, then putting them in 70% alcohol or 50% isopropyl alcohol. If no hot water available, put straight in preservative. **Don't forget that most should be kept alive**. A sample should contain about 100 maggots (of each size if possible).
The living specimens should be placed in a vial, with air and food, as for the eggs. There should be only enough maggots to cover the bottom of the vial. Too many in one vial will drown.

Pupae and Empty Pupal Cases - these are extremely important and are easy to miss. They are often found in clothing, hair or soil near the body. Pupae like dry, secure areas away from the wet food source in which to pupate so pockets, seams and cuffs are likely hiding places. If the remains are found indoors, they may have traveled some distance and be under clothing, rugs, boxes *etc*. They range from 2-20 mm, and are oval, like a football. They are dark brown when completely tanned. An empty pupal case is very similar but is open at one end, where the adult fly has emerged. They need some air, so secure a paper towel over vial as for eggs, as although the pupae are immobile, if they emerge during transit, an adult can get out of anything! A piece of tissue in the vial will help to avoid breakage as they are quite vulnerable. This can be *very slightl*y moistened with water, but be careful not to drown them. The moisture isn't necessary if the journey is not long**. Do not preserve pupae!** They won't grow, so the reasons for preserving larvae do not apply, and it is almost impossible to identify a pupa until it emerges as an adult. I also cannot determine its exact age until I find out the day on which it emerges. If a pupa is found when a pale colour, it is just entering pupation, so please keep that specimen separate and label as pale coloured, as it will darken in a few hours. Such a specimen can be aged to a matter of hours.

Adult Flies - are less important. They are only of use in indicating which species of insect are likely to develop from the corpse, as you cannot determine whether an adult has developed on the corpse, or has just arrived from somewhere else to oviposit, unless it emerged only an hour or so earlier. **If an adult has crumpled wings**, it may have just emerged, so IS still important as it can be linked to the body. It should be collected, labeled as such, and kept separate. Adults can be collected by net or by using an inverted vial. They can be left in the vial without air or food, as I will kill them as soon as I receive them. The presence of empty pupal cases, however, indicates that an insect has developed on the corpse and reached adulthood. This can be very important as it indicates that at least one generation of flies of this species has completed development on the corpse.

Beetles - can be found as adults, larvae or grubs, pupae and also as cast skins. All stages are equally important. They move fast and are often found under the body, and in and under clothing. They can be placed in vials with some air. They only need to be fed if it will be more than 24h before they reach an entomologist. If necessary they can be fed extra maggots. They are cannibals so should not be placed in the same vial!!

Other Insects - other insects may be present. If you are not sure whether it's an insect, collect it anyway and place in a vial.

Other Samples - Soil and leaf litter samples will also be useful. About a coffee can size of **soil** from under of very near the body is useful. If the soil below the body is extremely wet, it is better to collect the soil from near the remains.

Labeling - Insects collected from one part of the body should be kept separate from those from another area. Different species should be kept separate as beetle larvae feed on fly larvae! If they look different, separate them. Each vial should be labeled with :-

- area of body/soil
- date and time of collection
- name of collector
- stage *e.g*. larvae, so that if the specimens are pupae when I receive them. I will know that they developed into the next stage during transit.

Handling - most specimens are fairly fragile and are probably best picked up with gloved fingers which are often gentler than forceps if you are not used to them. Very tiny or delicate specimens can be picked up using an artist's brush dipped in water or alcohol depending on what you are about to do with them. Make sure all the vials are very well sealed!

Packing - The insects should be taken to the entomologist as soon as possible. They should be couriered or hand delivered to maintain continuity. They should be packaged in a cardboard box as this has lots of air. Each vial can be taped so that it remains upright. The whole box must remain upright.

I need to know many other factors about the death site :-

Habitat -

- general - is it woods, a beach, a house, a roadside?
- vegetation - trees, grass, bush, shrubs?
- soil type - rocky, sandy, muddy?
- weather - at time of collection, sunny, cloudy?
- temperature and possibly humidity at collection time
- elevation and map coordinates of the death site
- is the site in shade or direct sunlight?
- mention anything unusual, such as whether it's possible that the body may have been submerged at any time.

Remains - I need to know :-

- presence, extent and type of clothing
- is the body buried or covered? if so, how deep and with what (soil, leaves, cloth)
- what is the cause of death, if known? in particular, is there blood at the scene?
- or other body fluids?
- are there any wounds? if so, what kind?
- are drugs likely to be involved? this may affect the decomposition rates
- what position is the body in?
- what direction is the body facing?
- what is the state of decomposition?
- is a maggot mass present? how many? this will affect the temperature on the body
- what is the temperature of the centre of the maggot mass(s)?
- is there any other meat or carrion around that might also attract insects?
- is there a possibility that death did not occur at the present site?

If the body is refrigerated at the morgue before the collection (it is much better to collect at the scene, but sometimes that is not possible) then I also need to know the exact time that the body went into the cooler, and the exact time it came out.
Photographs, or a video of the scene, the body *in situe* and the site after removal of the body are also extremely useful.

When the insects reach the insectary, the immature specimens are measured, and examined, then placed in a jar containing a suitable feeding media. In the case of blowflies, this is usually beef liver, which is placed on top of sawdust. When the insects reach the prepupal stage and leave the food source they will burrow into the sawdust to pupate. The insects are checked daily and when they pupate they are removed and placed in a petri dish with damp filter paper. The date of pupation and the date of emergence is noted for each specimen. When the adults emerge, they are killed and pinned, then placed in an insect box. Each insect has a detailed label. Any adults collected directly from the corpse are immediately killed and pinned.

The reasons for raising the immatures are two-fold. Firstly, larvae are very difficult to identify to species, but adults have many more diagnostic features. Secondly, the dates of pupation and emergence are used to help calculate the age at the time of collection.

Other important information used to determine elapsed time since death include :-

- weather records from the nearest weather station, including temperature and precipitation
- the distance between the death site and the weather station

This method of determining elapsed time since death using insect evidence can be demonstrated using an actual case. Human remains were found in mid October. Most of the head region was missing as death was due to gunshot wounds. The upper portion of the body was almost skeletonized, but the lower area, clad in

tight clothes, appeared almost fresh. There were several large maggot masses on the corpse which generate their own heat for a while due to the frenzied activity. The temperature of the largest maggot mass was 20oC, even after the body had been refrigerated at 4oC for two hours. All sizes of larvae were collected and three pupae. These were pale in colour so had only just pupated. No puparia were found. The mean temperature at the death site was 15oC.

Two species of blowfly emerged, *Calliphora vomitoria* and *Phormia regina*. Both are common species that are amongst the first to arrive on a corpse. The oldest stage of *Calliphora vomitoria* collected was just entering the prepupal stage of the third instar. This was determined from size, no. of spiracular slits (breathing holes) date of pupation and behaviour, in that the largest specimens immediately left the beef liver and entered the sawdust, indicating that they had stopped feeding. At the temperature of the deathsite, 15oC, *Calliphora vomitoria* takes a minimum of 9.3 days to reach the beginning of the prepupal stage of the third instar. So these insects were a minimum of 9 days old when collected on 12 October, meaning that they were laid as eggs on or before 4 October. As there was blood at the scene, the insects probably arrived very soon after death. Therefore death must have occurred on or before 4 October.

Using the same techniques for *Phormia regina*, the oldest specimens of which were in the pupal stage when collected, it was calculated that *Phormia regina* was oviposited no later than 3 October. Therefore, using the two insects together, it can be shown that death occurred on or before 3 October. Other police evidence later showed that death had actually occurred on 3 October.

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OTHER USES FOR INSECTS IN FORENSIC SCIENCE

- **the body may have been moved after death,** from the scene of the killing to a hiding place. Some of the insects on the body may be native to the first habitat and not the second. This will show that not only was the body moved, but it will also give an indication of the type of area where the murder actually took place.
- **the body may have been disturbed after death**, by the killer returning to the scene of the crime. This may disturb the insects cycle, and the entomologist may be able to determine not only the date of death, but also the date of the return of the killer.
- **the presence and position of wounds,** decomposition may obscure wounds. Insects colonize remains in a specific pattern, usually laying eggs first in the facial orifices, unless there are wounds, in which case they will colonize these first, then proceed down the body. If the maggot activity is centred away from the natural orifices, then it is likely that this is the site of a wound. For example, maggot activity on the palm of the hands indicates the probable presence of defense wounds.
- **the presence of drugs** can be determined using insect evidence. There is often not enough flesh left to determine drug presence, but maggots bioaccumulate so an can be analyzed to determine type of drug present.
- **insects can be used to place a suspect at the scene of a crime**. For instance, an insect inside a cocklebur was used to connect a rapist to the rape site.
- **civil cases** also sometimes use insect evidence.
- **child or senior abuse/neglect.** Some insects will colonize wounds or unclean areas on a living person. This is called cutaneous myiasis. In these cases, the victim is still alive, but maggot infested. A forensic entomologist will be able to tell when the wound or abuse occurred. For instance, in the case of neglected children, the onset of maggot infestation will give a minimum time interval since the child last had a diaper change. Such cases occur particularly in young children and seniors.

Although forensic entomology can be very effective in determining elapsed time since death, it has its limitations :-

1. The **temperature** of the death site is obviously a very important factor, but few criminals are thoughtful enough to kill their victim right underneath a weather station! In most cases, the weather records come from several miles away. We are trying to overcome this by setting up a miniature weather station at the death site after discovery, to compare these data with that from the weather station, in order to

determine the difference between the two sites, if any. Also the microclimate of the corpse itself will be slightly different from the surrounding area, especially if a maggot mass is present. Therefore, it is extremely important to know whether masses are present.

2. Forensic entomology in Canada is **seasonal**, that is, it is only commonly used in spring, summer, and fall when insects are abundant. It is of less use in winter, unless its very mild, as there are no or very few insects present. This can be a limitation, but can also be an advantage as I can sometimes show that a victim found in spring was killed the previous fall if insect evidence is present.
3. The results are not immediate, as it takes time to rear the insects. DNA evidence is now being developed to speed up identification of immature specimens.
4. The body may have been disposed of in a way that **excludes** insects *e.g.*

    a. **freezing** - if the body was frozen for a period of time before being placed outside on, for example, 8 May, the insects would only invade then, giving the misleading impression that death had occurred on 8 May. However, other forensic experts would be able to determine whether or not the body has been frozen, and insect evidence will still determine time of exposure.
    b. **burial** - if the body is buried deeply, then most insects will be excluded. However, most criminal burials are not very deep, as the aim is merely to conceal the body, and most insects will dig down to the body, particularly if there is blood soaked in the soil. Therefore, insect evidence can still be used. We have an estensive database for buried bodies in BC
    c. **wrapped** - if the body is wrapped or packaged in some way the insects may be excluded, but the wrapping must be completely secure. A body part was found sealed in a garbage bag which had been tied securely at the top, but the remains were maggot-infested, and showed severe insect damage. The adult females had probably laid their eggs at the knot, and the minute first instar larvae had crawled in.

5. More **research** is needed. Insect succession varies from geographic region to region and the species and time of colonization must be developed for all areas using this type of evidence. Research has been conducted in British Columbia in a variety of habitats, seasons and geographic areas to develop a database for this Province. It is intended that this will be extended across Canada.
6. **Drugs** - the presence of drugs may affect the development of the insects. Work is planned to determine effects of common narcotics on insects in Canada

In conclusion, **INSECTS ARE EVIDENCE!** Forensic entomology is a very useful method of determining elapsed time since death after 72 h. It is accurate to a day or less, or a range of days, and may be the only method available to determine elapsed time since death. It is vital that the insects are collected properly and its accuracy depends on this and on suitable conditions for insects.

1.Definition

- In its broadest sense, forensic entomology is the study of insects involved in any legal action, and can include urban and stored products entomology.

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