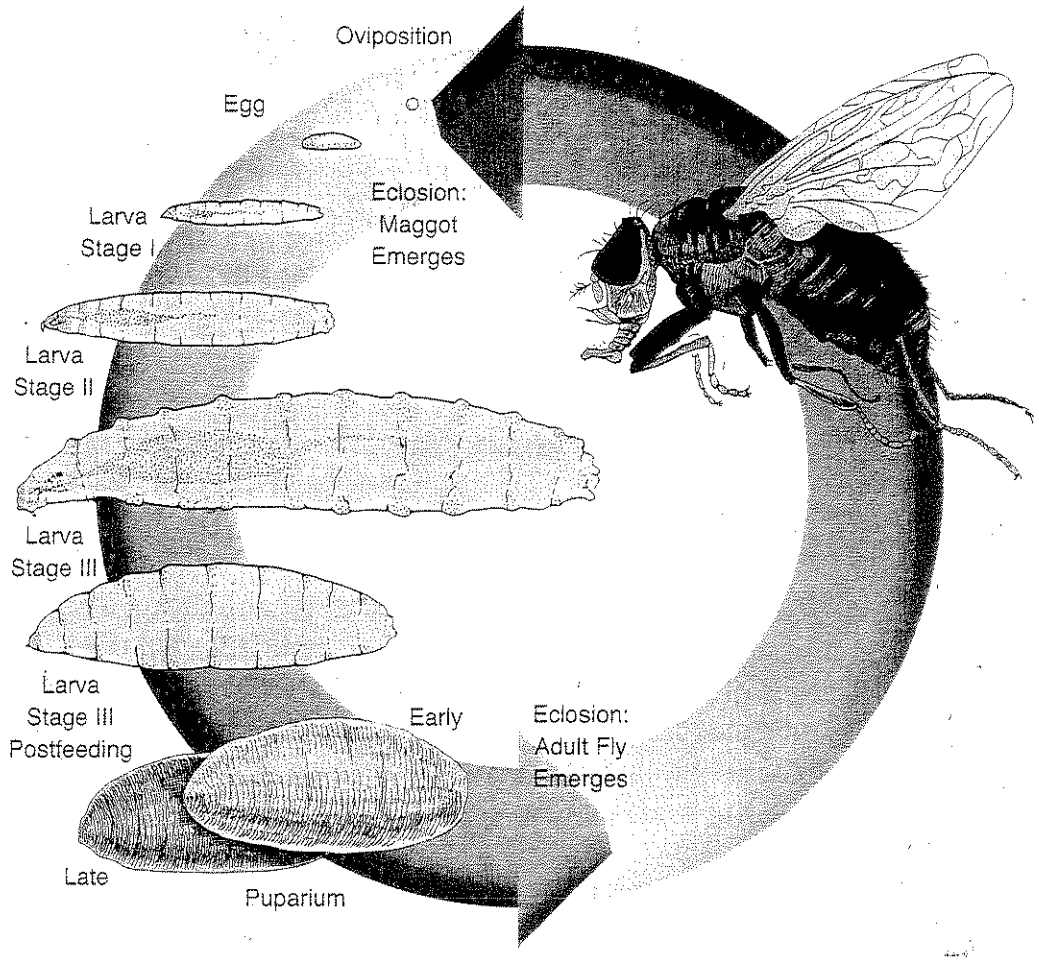
exercise 16

Forensic Entomology

The order of natural events following a death at an outdoor location begins with the appearance of any number of fly species. The arrival of insects, such as the blow fly, to a death scene is inevitable and almost immediate. The progression of these events is more or less predictable, which makes the analysis of these events useful to investigators seeking to account for the time that has passed since death, called the **post-mortem interval (PMI)**. The application of this knowledge—the profile of insect activity since death—to criminal investigation is called forensic entomology.

Flies will begin colonizing the orifices and any wounds present on the body of the recently deceased. Fly activity, especially egg-laying, sends a strong olfactory signal to other flies in the area so a body can quickly become swarmed. During this period of commotion, eggs are deposited continuously, producing young of varying ages present at the same time. The most important specimen collected by a forensic entomologist is the oldest life stage present on the body. This specimen acts as a stopwatch that began ticking the moment the first egg was deposited on the body. Using that species' life cycle timetable with local temperature and weather information, a forensic entomologist is able to approximate the PMI.

The life cycle of a blow fly begins with **oviposition**, or egg-laying. Development of the egg ends with an immature fly, called a larva or maggot, eclosing (hatching) from the egg. The larva molts (sheds its skin) three times during the larval phase. The individual between each of the molts is called an instar. Thus, there are three larval instars, each larger than the last. The larva will shed its skin three times, each time with a different set of appendages. Many species then proceed to the **migration** phase, wherein a larva stops feeding and travels several feet away from the corpse. The purpose of this travel is to allow the larva to pupate in a location some feet away from the environment which it used to call home. **Pupation** is marked by a hard shell the maggot secretes in which it will change from the larval to adult stage. The larval stage is protective in nature and some flies will remain as pupae for days



to weeks. After further development, an **adult** fly emerges from the pupa and begins the cycle anew. This describes the life cycle of the dozens of species we commonly refer to as "house flies." Blow flies and flesh flies are in this group, and these are the species to which we limit our exploration.

THE LIFE CYCLE

After eggs are laid, the duration of a typical blow fly life cycle is dependant on the temperature. It requires a certain time period, expressed as "degree-days" or "degree-hours," for the larva to develop. Degree days/hours are the number of days/hours multiplied by the number of degrees above a threshold temperature for that species (usually 50° Fahrenheit). This means that any time interval, say overnight, that is below 50° Fahrenheit does not count toward the life cycle—it is too cold for the eggs/larvae/puparia to grow. For example, at 70° Fahrenheit, eggs

will hatch into larvae after approximately 12 hours. Larvae will pupate after approximately another 12 days, and adult flies will emerge from the puparia after approximately 8 more days. In other words, the complete life cycle will require 3 weeks at a constant temperature of 70° Fahrenheit.

Other than temperature, drugs or poisons in the decedent's bodily tissues can affect the growth rate of fly species. For example, arsenic, a heavy metal found in rat poison, slows down the growth rate of all fly species. More specifically, narcotics like cocaine can accelerate the growth rate, especially in the *Calliphora vomitoria* species. Factors like these are always important to the calculation of the post-mortem interval and must be carefully considered.

THE STAGES OF DECOMPOSITION

Decomposition of a mammal begins at the "fresh" stage and progresses until the body appears bloated. The flies that arrive during this phase are there within minutes, and are most commonly the blow flies and flesh flies, from genus *Calliphora* and *Sarcophaga*, respectively. The "bloated" stage occurs because of the activity of bacteria which produces gases inside the body. It is during this phase that house flies, from genus *Musca*, begin to appear and deposit eggs. The "decay" stage begins with the splitting of the skin to allow the gases to escape. The body then takes on a compressed appearance and exudes a powerful odor. By the end of this phase, the flesh is absent and only bone, skin, and hair remain. It is at this time that the larvae begin their migration away from the food source to find a suitable location to pupate. In the "post-decay" stage, beetles begin to colonize the body and consume the dry, less nutrient-rich components.

An important clue to the condition of a body at death is the location of the larval mass on a body. If an individual dies without sustaining a wound to the flesh—for example, by carbon monoxide asphyxiation—then the flies would initially lay their eggs in the orifices of the body. This grants the most convenient access through the tough skin. In this situation, the larvae are seen in the face/head and the anal/genital region first. If the larval mass is present in the chest or abdomen, this indicates there was an "unnatural orifice" in that area at the time of death. For example, if the decedent received a fatal stab wound to the abdomen, then a larval mass would be present in the trunk area, as well as the head and perineal regions.

EXERCISE 1 – ESTIMATING THE POST-MORTEM INTERVAL[18, 19]

In this exercise each team will be presented with "entomological evidence" collected from four death scenes. This "evidence" is simulated larvae and pupae, symbolized by pipe cleaners of varying colors and lengths. Teams use a ruler to measure the length of each "maggot." These data are then used in conjunction with Table 1 to identify the species present at the death scene and determine the post-mortem interval.

Color Key for Pipe Cleaners

White = *Sarcophaga*
Blue = *Musca*
Yellow = *Calliphora*
Pink = *Piophila*
Brown = pupa (species can only be determined by length of pupa)

Table 1 L=larva, E=egg, A=adult, P=pupa—Stage and Length in Millimeters for Each Species as Developed Under Constant 72° Fahrenheit.

	SPECIES			
Days after Death	***Musca Domestica***	***Calliphora Vomitoria***	***Sarcophaga Carnaria***	***Piophila Nigirceps***
1	~	E	L9–11	~
2	E	L9–11	L12–16	~
3	E	L9–11	L17–20	~
4	L6	L12–16	L21–25	~
5	L6	L12–16	L26–30	E
6	L7–11	L17–20	L31–35	E
7	L12–16	L17–20	L36–40	L3
8	L17–20	L21–25	L41–44	L3
9	L21–25	L21–25	L44–46	L4–6
10	L26–30	L26–30	L44–46	L7–9
11	L31–35	L26–30	P38–40	L10–13
12	P26–29	L31–35	P38–40	L14–16
13	P26–29	L31–35	P38–40	P13–15

(continued)

[18] Please refer to the *Instructor's Manual* for background information and instructions for the preparation of the materials used in this exercise.

[19] Adapted from "Of Maggots and Murder—Forensic Entomology in the Classroom" by Lisa Carloye, in *The American Biology Teacher, vol. 65*(5), May 2003.

Table 1 *Continued*

	SPECIES			
Days after Death	***Musca Domestica***	***Calliphora Vomitoria***	***Sarcophaga Carnaria***	***Piophila Nigirceps***
14	P26–29	P31–34	P38–40	P13–15
15	P26–29	P31–34	P38–40	P13–15
16	P26–29	P31–34	P38–40	P13–15
17	P26–29	P31–34	P38–40	P13–15
18	A30–32	P31–34	P38–40	P13–15
19		P31–34	A42–45	A16–18
20		P31–34		
21		A36–38		

Table 2 Factors Affecting the Development of Four Fly Species. Temperature Changes as from the Standard of 72° F, Given in Number of Days.

		SPECIES			
		Musca Domestica	***Calliphora Vomitoria***	***Sarcophaga Carnaria***	***Piophila Nigirceps***
Temperature (in°F)	55	Delayed 4	Delayed 4.5	Delayed 4	Delayed 3
	65	Delayed 2	Delayed 3	Delayed 2	Delayed 1
	80	Aaccelerated 1	Accelerated 2	Accelerated 1.5	Accelerated 1
	85	Accelerated 2	Accelerated 4	Accelerated 3	Accelerated 2
Environmental factors	Habitat	Urban/rural	Urban/rural	Urban/rural	Urban
	Lighting	Full to part sun	Part sun to shade	Prefers sun	Prefers sun
	Drugs	No effect	Sensitive to drugs	No effect	No effect

MATERIALS

Container of "maggots" and "puparia," supplied by your instructor

Ruler

Reference data and worksheet

PROCEDURE

1. Examine the "maggots" and "puparia" in your container. This represents the entomological evidence from a crime scene. Be careful to work with the evidence from one case at a time—do not mix up your cases!
2. Measure each "maggot" and record its length and color.

3. Compare these to the data provided in Table 1 to identify the species present in your "evidence" sample.
4. Based on the entomological evidence provided, you should make a conclusion about the earliest possible date of death in this case. Outline your findings in your notebook, citing specific examples from your analysis.
5. Answer the questions following each of the following cases. Document your findings (answers) in your notebook.

Case A

Elderly male discovered in his home—apparently deceased for some time.

Maggots are apparent in his face and neck region.

Local temperature has been a consistent 70–75°F for the last 2 weeks.

Questions:

1. Approximately how long has this man been deceased?
2. Did you find maggots of different ages on the body?
3. Why or why not?

Case B

Young male recovered in his college dorm room.

Maggots are evident in the man's head and chest regions.

The weather report shows daytime temperatures of 74–95°F with sunny skies.

The windows of his room are closed and the university keeps the building at a constant temperature of 72°F.

Questions:

1. Approximately how long has this man been deceased?
2. What effect, if any, does the outside temperature have on your estimation of the PMI?
3. How does the fact that the windows are closed influence your evidence sample?
4. How do you explain the absence of *Calliphora vomitoria*?
5. Do you suspect foul play? Explain.

Case C

Adult female found in a city park.

The weather report indicates that the recent weather has been a heat wave with daytime temperatures at 84–86°F.

The toxicology report indicates the presence of cocaine in the woman's blood.

Questions:

1. Approximately how long has this woman been deceased?
2. What effect, if any, do the toxicological findings (cocaine in the woman's bodily fluids) have on your estimation of the PMI? Explain.
3. What effect, if any, does the daytime temperature have on your estimation of the PMI?
4. Do you suspect foul play? Why or why not?

Case D

Adult male found by a road cleanup crew.

Maggots found in his face and neck region.

The subject was under tall trees on the side of an interstate highway in a rural area.

The weather has been partly sunny and 70–74°F for most of the week.

Questions:

1. Approximately how long has this man been deceased?
2. What effect, if any, does temperature have on your estimation of the PMI?
3. Does the death scene location, when compared to the entomological evidence, suggest foul play? Do you need more information to make this determination?

EXERCISE II – Rearing Maggots

In this exercise, you will observe the initial colonization activity of blow flies and subsequent larval development using a simulated corpse composed of fresh beef or chicken liver. Since fly activity is seasonal and temperature-dependent, it is necessary to perform this lab during a warm week, with a daytime temperature of *at least* 65°F (18°C).

Safety

Materials

One (1) three-pound coffee can, with lid

One hole paper punch

¼ lb. fresh beef or chicken liver

5-inch square of aluminum foil
Vermiculite
Timer
Outdoor thermometer
Specimen jars
Marker
Isopropyl alcohol
Magnifying hand lens

A reference collection is a set of life stages from one or more species of flies kept for the purposes of comparison to recovered specimens. Your instructor will find a useful reference collection kit—*Maggots from Murder*, available at http://www.maggotsfrommurder.com, to help you determine the growth stage of the maggots you recovered.

Procedure

1. In the lab, remove the lid from your coffee can and make ten or fifteen holes with the one-hole paper punch. These holes will provide access to the food source for the adult flies.
2. Add approximately ½ inch of vermiculite to the bottom of the can.
3. Fashion a tray of aluminum foil around the liver. This will help to contain the liver during larval development.
4. Put the liver in the can and affix the lid.
5. Select a shady spot outdoors and away from any heat sources for your simulated death scene.
6. Note the time and date when you place the can on the ground.
7. Retrieve the can after 24 hours.
8. Using your magnifying lens, remove a few eggs from the surface of the "body" and transfer them to a specimen jar containing isopropyl alcohol. Label the jar with the contents, date, time, and your initials.
9. Sketch the eggs in your notebook.
10. Place the can in a well-ventilated area in your laboratory and observe the progress of the larvae over the next 3 weeks. Record the temperature of the room every day.

Plan Your Experiment

Plan the following observations:

1. Appearance of larvae
2. Appearance of migrating larvae
3. Formation of puparia
4. Appearance of adult flies

The room temperature will determine the time of these events relative to the time of oviposition. *It is up to you to estimate when each of these phases will occur.*

Make Your Observations

1. For each of the phases previously described (from egg to adult), remove a specimen of the appropriate life stage and rinse with water to remove as much of the substrate as possible.
2. Transfer to a specimen jar containing isopropyl alcohol.
3. Label the jar with the contents, date, time, and your initials.
4. Using your stereoscopic microscope, examine the specimen and sketch it in the best detail you can.
5. Measure the approximate dimensions of each specimen and include this information in your report.