



BIOMORPHOLOGICAL, ECOLOGICAL AND ETHOLOGICAL PROPERTIES OF DIPTERA (ARTHROPODA: INSECTA) SPECIES IN DECOMPOSITION PROCESS

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ABSTRACT

The existence of insects around the world dates back to 400 million years. People have existed for 300 thousand years. There are more than one million insect species have been identified on earth. The functional properties of insects are examined depending on many different disciplines. Forensic Entomology is the most current of these. The experimental part of this study was conducted between the dates of April 2018 and August 2018 in the Yahyalı locality of Kayseri. Within the scope of the study, Diptera (Arthropoda: Insecta) species, which have a forensic importance, were investigated in terms of their ecological, biomorphological and ethological characteristics. Forensic entomology is the science in which biological information and data related to insect-arthropods are used in cases of misdemeanor and civil lawsuits. The main reason for the use of arthropods in criminal investigations is that they are one of the living species that detect and accept the body as soon as possible, exist in every stage of decay, and some insect species are particularly specific to certain environments and habitats. In the crime scene investigations, the ecology, biomorphology and ethology of insects are used to find out when, how and where the death occurred. Within the scope of the study, in the research conducted; The ethological properties of Diptera individuals in the decomposition process and within the framework of ecological succession were investigated by letting 5 *Rattus rattus* (Linnaeus, 1758) samples per month in the periods are protected. In addition, it is aimed to contribute to forensic entomological researches and analysis of forensic cases by making biomorphological - ecological evaluations.

Keywords: Diptera, Ethology, Ecology, Fauna, Criminology

1. INTRODUCTION

The science that studies insects is called entomology. The existence of insects around the world dates back to 400 million years. There are more than one million insect species have been identified on earth [1-8]. In parallel with their large numbers, ecological diversity is also high for insects.

From the functions specified; Along with the fact that animal carcasses are in the decomposition process, they take part in the clarification of legal processes [6, 7, 9]. Forensic entomology is the science that investigates the ways and methods of arthropod use in unexpected sudden deaths, unexplained traffic accidents, determining the place and time of death. Forensic entomology refers to a regular decomposition event by predicting the death time by the occurrence of adult individuals who come to the corpse after the death of the living creatures in the Arthropoda branch and Insecta class [1, 5, 10-16]. In forensic entomology science, there is a certain succession of insects that come upon the body. Depending on this temporal succession, the developmental periods of insects are examined and the death time interval (post-mortem interval: PMI) is determined [6, 11, 13, 17, 18] Insects on the body are affected by many variables and perform their colonization accordingly. These variables are meteorological events, the cause and form of death of the corpse, ecological phenomena. [11, 13, 18].

The main reason for the use of arthropods in forensic events is that they reach the body in a short time and that they are in ecological and periodic succession in the decomposition process make them the most reliable evidence [1, 10, 12, 13, 15, 16].

In the 13th century, Sung Tzu discovered insects by chance to search for the killer of a worker whose throat was cut. He caught the culprit due to the presence of Calliphoridae members in a sickle without blood traces [2, 4, 9, 10, 13, 17-28]. In 1855, Dr. He made the first modern forensic entomological studies by Bergeret. He used *S. carnaria*'s larva as evidence in a courtroom [11, 19, 24-26, 29]. In 2001, Goff proved that forensic insects were the best method in PMI [17, 19, 20, 21, 27, 28, 30-32]. In 2014, Çavuşoğlu detected traces of forensic insects in the decayed human corpses [33].

Ethology is the sub-branch of zoology that studies animal behavior. Ethology encompasses laboratory and field studies carried out in close cooperation with certain disciplines such as evolution, neuroanatomy and ecology [34]. Ethology has been concerned with the basic studies such as revealing the behavioral biology of the species whose environmental conditions are largely shaped by humans, determining the behavioral needs, as well as the development of "biological descriptors" related to

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animal welfare and the examination of the effects of different feeding and feeding conditions on behavior [35]. With the evaluation of ethological features, the position of forensic entomology gains value in legal processes.

Vertebrate animal bodies provide nutrients for many organisms. Insects come at the beginning of these organisms. These form a seasonal and continuous “ecological succession”, in which sequential populations colonize and disappear during the decomposition process of the carcass. The timing and nature of succession depends on the size of the carcass, seasonal and environmental climatic conditions and non-biological environmental factors such as soil type. The creatures participating in succession vary depending on whether they are on or in the carcass, just below the carcass or being in the soil near the carcass. At the same time, even in different geographic areas with a similar climate, each succession will also have different species. This is due to the fact that only a few species have a very wide distribution, and therefore each biogeographical region has its own carcass fauna. However, certain taxa that are specialized with carcasses are also determined. [36-40].

The first hierarchy in the decomposition of lechin is the fresh phase (Photo 1.(a).). At this stage, microorganisms and insects that existed in the body begin to decay the body. The second hierarchy begins as the swelling phase (Photo 1.(b).) within a few days. After about 2 weeks, the third stage begins as the active decay stage (Photo 1.(c).), where strong decay odors are formed. At the end of this phase, the carcass becomes almost a dry carcass and only the bones remain in the carcass in response to the fourth stage, the slow dry decay phase (Photo 1.(d).) [17, 27, 40–46].

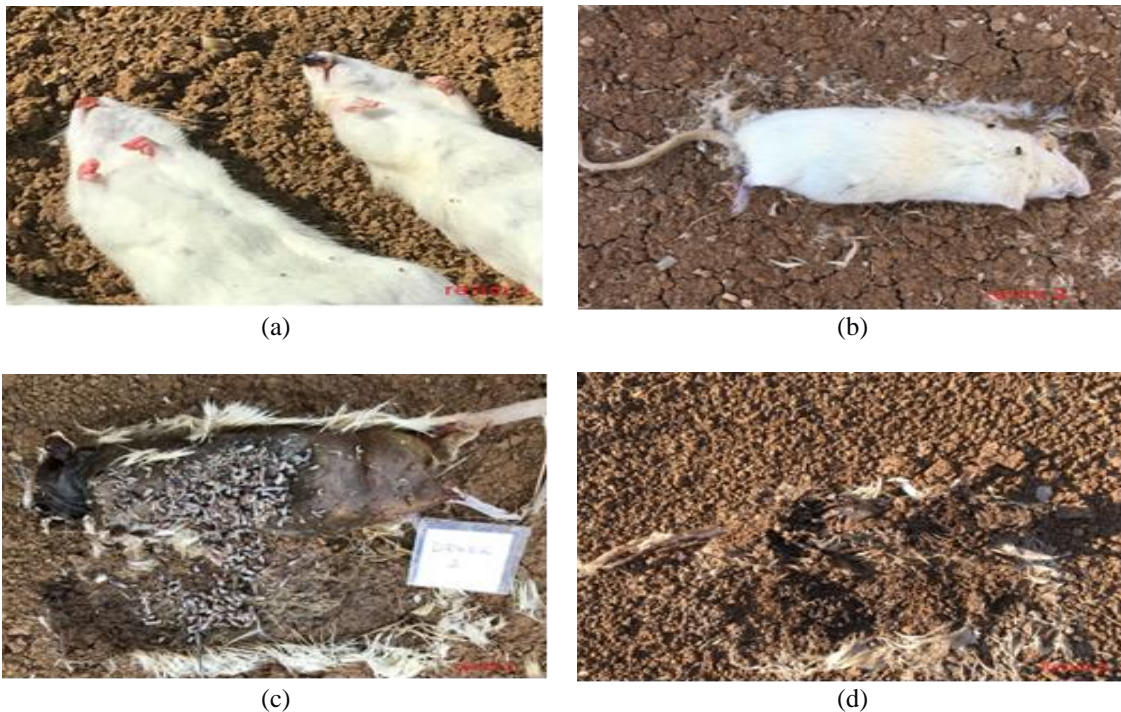


Photo 1. (a) Fresh stage, (b) Swelling stage, (c) Decay stage, (d) Drying stage.

Diptera is one of the most populated insect order. They prefer to live in common living spaces with people. They are important in ensuring ecological balance. They are called biplanes. However, while the first pair of wings was found, the second pair of wings turned into a structure called barbell. This transformed structure keeps the Diptera in balance [4, 47, 48]. It is the order that has the best flight ability among insects. There are also wingless types. The layer of the outer kit is not very hard [4]. Adult dipters have sizes between 7 and 10 mm. They can be of various colors. Depending on the antenna types, undercarriages appear. The lower part, whose antennae are long, such as the filiform type, is called nematocera. It is called Brachycera, another subset with its antennae with 3 segments and arista hair at the end [4-7, 17, 28]. The mouth structures are of a licking - absorbent or introducing - absorbent type. They carry two large honeycomb eyes and 3 ocel eyes. They show Holometabol metamorphosis. The larvae are rimless leg type. In the case of the pupa, it is stored in a sheltered place, so as not to prey. They are the fastest species in experimental studies and forensic cases. After death, they reach the body within 1-2 minutes under suitable conditions. They are present in almost all stages of decomposition, except during the drying phase. The reproductive type is ovipar and vivipar. In the case of vivipar, the adult does not lay eggs, but live breeding is in question. Some Diptera species are breeding types when they cannot trust the environment [5-7, 13, 17, 19, 20, 21, 23, 24, 26-28, 47, 48].

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2. MATERIAL AND METHOD

Our research covers April 2018 - August 2018. As experimental animals 25 *Rattus rattus* (Linnaeus, 1758), was used for this study. Decomposition length of *Rattus rattus* was taken into consideration to determine our working process. Accordingly, it was determined that the decomposition ended in 30 days by evaluating and observing the ecological environment [10-30,33].

Every month, 5 rats were left by on the experimental land. Daily examinations were made and samples were collected (egg, larval periods, pupa, adult). The skeletons remaining at the end of the month were collected and disposed in appropriate waste units. The ground was ventilated and ground was prepared for new rats. Since we leave the rats on the ground, a special cage arrangement has been prepared and installed to protect it from other factors. Digital temperature and humidity meter were used to measure the ambient temperature. A mercury thermometer was used to measure the temperature of the soil at 10 cm.

Our study was conducted between April and August 2018 in a land where steppe vegetation is observed in Yahyalı locality of Kayseri. It has been used by obtaining the necessary permissions for our land, which is a private property, for easy monitoring of inactive and daily periods away from the residential area. A sheltered area has been chosen so that the land used will not disturb the environment and people. The land is located at a height of 1200 m from the sea at 38 ° 6'36 "N, 35 ° 21'10" D coordinates.



Photo 2. Experiment area

3. RESULTS AND DISCUSSION

3.1. *Calliphora vicina* (Robineau-Desvoidy, 1830)

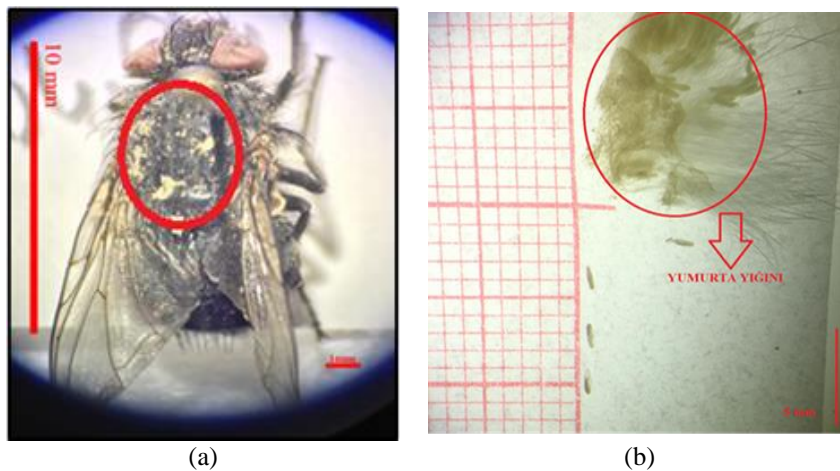
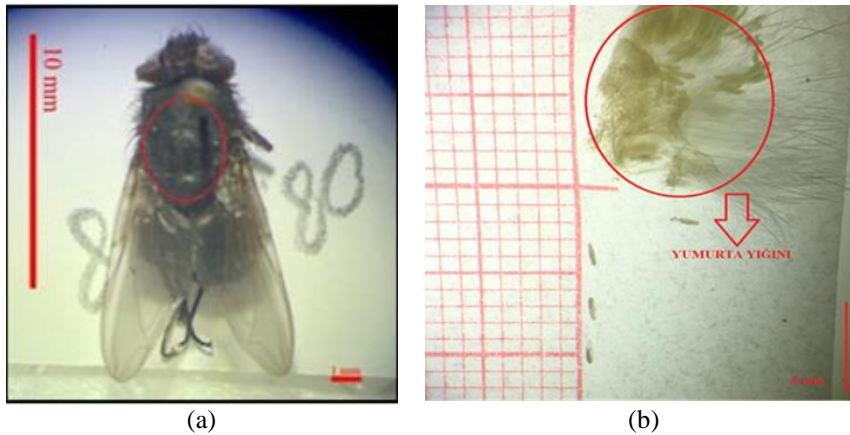


Photo 3. (a) Dorsal view of *Calliphora vicina*, (b) Egg view belonging to Calliphoridae family

Table 1. Evaluation of *Calliphora vicina* (Robineau-Desvoidy, 1830) [4, 28, 47, 49].

Decomposition	Biomorphology	Ecology	Ethology
Fresh stage	Its eggs are 1-2mm in size and adults are 5-13mm in size. Abdomen is metallic blue.	They can live within 17-25 °C. It is generally seen in spring and autumn seasons.	Migration takes place because the life of the species is forced in a temperature above 25 °C.
Swelling stage	Thorax part is dusty blue. The surface of the gena (cheek) part of the head has orange-yellowish black hair.		It has harmonized city life.
Decay stage	The stem - vein vein on the wing is hairless. The thorax has an acrostical hair pattern.		Due to their biological rhythms, day length is a factor. It has also a relationship with myiasis.

3.2. *Calliphora vomitoria* (Linnaeus, 1758)

**Photo 4.** (a) Dorsal view of *Calliphora vomitoria*, (b) Egg view belonging to Calliphoridae family**Table 2.** Evaluation of *Calliphora vomitoria* (Linnaeus, 1758) [4, 28, 47].

Decomposition	Biomorphology	Ecology	Ethology
Fresh stage	Its eggs are 1-2mm in size and adults are 5-13mm in size. Abdomen is metallic blue. Thorax part is dusty blue.	They can live within 15 - 25 °C. It is generally seen in spring and autumn seasons.	Migration takes place because the life of the species is forced in a temperature above 25 °C.
Swelling stage	The gena part of the head part has orange hairs on the black surface.		They make intense sound while flying.
Decay stage	The stem - vein vein on the wing is hairless. The thorax has an acrostical hair pattern		Adapted to rural life. Due to their biological rhythms, day length is a factor. Life activities are active in shade and wooded areas. it has also a relationship with myiasis.

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3.3. *Lucilia sericata* (Meigen, 1826)

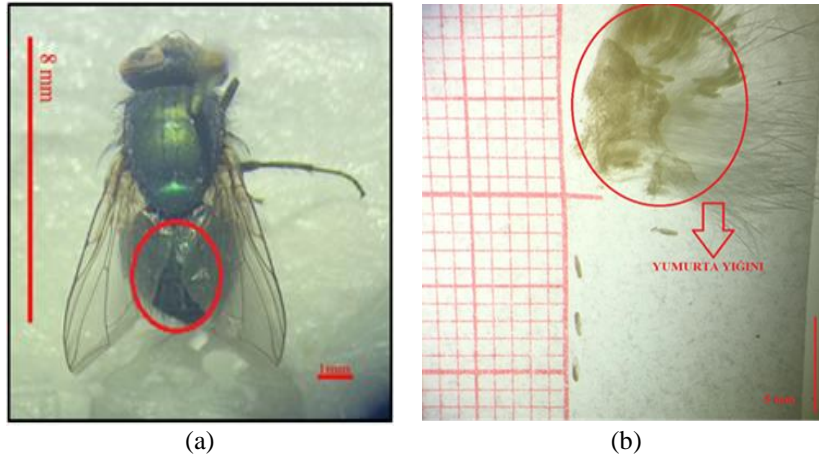


Photo 5. (a) Dorsal view of *Lucilia sericata*, (b) Egg view belonging to Calliphoridae family

Table 3. Evaluation of *Lucilia sericata* (Meigen, 1826) [50, 51]

Decomposition	Biomorphology	Ecology	Ethology
Fresh stage	Their eggs are 1-2mm in size, and adults are 5-9mm in size.	They can experience temperate air within 16 - 27 °C. It is generally seen in spring and autumn seasons.	Flies are the first to come after death.
Swelling stage	Abdomen metallic is blue-green. The stem - vein vein on the wing is hairless.		They love fresh carcasses.
Decay stage	1. The calypter under the double wings is hairless. The basicosta at the point where the wing is attached is yellow in color.		It is colonized in open and sunny places. It has also a relationship with myiasis.

3.4. *Lucilia cuprina* (Wiedemann, 1830)

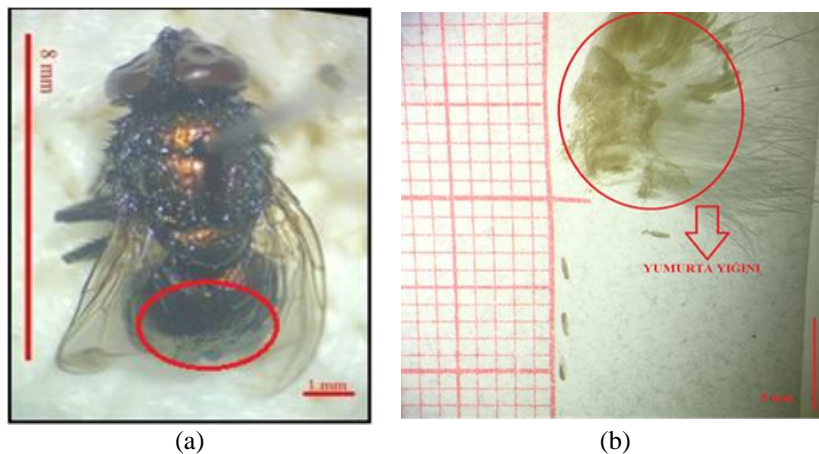


Photo 6. (a) Dorsal view of *Lucilia cuprina*, (b) Egg view belonging to Calliphoridae family

Table 4. Evaluation of *Lucilia cuprina* (Wiedemann, 1830) [4, 52-55]

Decomposition	Biomorphology	Ecology	Ethology
Fresh stage	Their eggs are 1-2mm in size, and adults are 5-9mm in size.	They can experience hot air within 17 - 40 °C..	Larvae and adults are found on rotten sugar plants, on top of carrion.
Swelling stage	Abdomen is metallic yellow-copper.		When they are disturbed, they run away immediately.
Decay stage	The stem - vein vein on the wing is hairless. 1. The calypter under the double wings is hairless. The basicosta at the point where the wing is attached is yellow in color.		In a relationship with myazis. They don't enter the houses much.

3.5. *Sarcophaga haemorrhoidalis* (Fallen, 1817)



Photo 7. Dorsal-lateral view of *Sarcophaga haemorrhoidalis*

Table 5. Evaluation of *Sarcophaga haemorrhoidalis* (Fallen, 1817) [28, 56, 57]

Decomposition	Biomorphology	Ecology	Ethology
Fresh stage	Adults are 7-14 mm in size.	They usually live in shady areas	Adults are fed with liquefied rotten materials.
Swelling stage	The checkered pattern in the abdomen and the yellow - red in genitalia and the three black bands in the thorax are distinctive points.		Their larvae feed on carrion and feces.
Decay stage	.		Adults and larvae are vectors. Females give birth to larvae to human carcasses indoors. Its larvae cause myalul. They are the first flies to arrive in the body after heavy rain.

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In April, the average temperature value was measured as 12.7 °C, humidity 57.8%, soil temperature 5.5 °C, and the amount of precipitation was 0.4 kg / m². During this month; *Calliphora vicina*, *Calliphora vomitoria*, *Sarcophaga haemorrhoidalis* dominated. The detected species are compatible with the data of Dinar, Topçular, Şabanoğlu and Özdemir [27, 28, 47, 48]. Depending on the temperature data, Calliphoridae larvae shortened the skin change and the process of getting out of the pupa. In this context, there was no difference with the literature [25, 27, 28, 47, 48].

In May, the average temperature value was measured as 15.6 °C, humidity 67.6%, soil temperature 6.1 °C, and the amount of precipitation as 1.9 kg / m². During this month; *Calliphora vicina*, *Calliphora vomitoria*, *Lucilia sericata*, *Lucilia cuprina*, *Sarcophaga haemorrhoidalis* have been identified. The detected species are compatible with the data of Dinar, Topçular, Şabanoğlu and Özdemir [27, 28, 47, 48]. Unlike the previous month, *L. sericata*, *L. cuprina*, was identified. The reason for this was that due to the high density of soil, as a result of the high amount of precipitation during this month, some insects woke up early from sleep and hid in a sheltered area, *Rattus rattus* corpses. Precipitation did not affect the temperature lowering direction. Even the temperature has increased compared to the previous month. Accordingly, *C. vomitoria* reduced her dominance. It has been determined that *L. sericata* is the dominant species. With the effect of rainwater, the corpse was dissolved and skin change times were extended because Calliphoridae larvae could not be fed. Again, with the effect of rain water, the process of entering and exiting the pupa has been prolonged due to the high humidity in the soil. In this context, it contradicts the current literature studies [41, 55, 58].

Average temperature value was measured as 19.1 °C, humidity 64.7%, soil temperature 7.8 °C, rainfall amount 1.2 kg / m² in June. During this month; *Lucilia sericata*, *Lucilia cuprina*, *Sarcophaga haemorrhoidalis* detected. It is in line with the data of Smith, Yuca, Karapazarlıoğlu, Özdemir, Şabanoğlu, Dinar and Topçular [26-29, 47, 48, 55]. Insects are looking for sufficient temperature, humidity and nutrients to breed. *L. sericata*, *L. cuprina* dominated. Depending on the temperature of the calliphoridae larvae, skin change and pupae exit times were shortened. With increasing temperature, *C. vicina* and *C. vomitoria* were not observed. It is also compatible with the current literature [47, 48].

Average temperature value was measured as 22.9 °C, humidity 53.1%, soil temperature 9.9 °C in July, rainfall amount was 0.1 kg / m². During this month; *Lucilia sericata*, *Lucilia cuprina*, *Sarcophaga haemorrhoidalis* were identified. It does not contradict the data of Smith, Çoban, Yuca, Özdemir and Şabanoğlu [17, 27-29,55]. There are species that overlap with our previous 3-month study. *L. sericata*, *L. cuprina* are dominant species. With increasing temperature, the pupae of the Calliphoridae species have shortened their exit time [47, 48].

Average temperature value was measured as 21.9 °C, humidity 51.3%, soil temperature 9.5 °C, rainfall in 0.1 kg / m² in August. During this month; *Lucilia sericata*, *Lucilia cuprina*, *Sarcophaga haemorrhoidalis* were identified. It is in line with the data of Goff, Çoban, Aksoy, Karaazarlıoğlu Özdemir and Şabanoğlu [16, 17, 23, 25-28, 47, 48]. In our study, species overlapping with July were determined. *L. sericata*, *L. cuprina* have been identified as the dominant species. With increasing temperature, the pupae of the Calliphoridae species have shortened their exit time [47, 48].

The first species that came to the *Rattus rattus* corpse were dominant in *C. vicina*, *C. vomitoria*, *L. sericata*, *L. cuprina*, depending on the temperature and humidity. It was observed in the fresh, swelling and decay stages of decomposition. These species are legally important because they perceive the smell of fresh blood immediately. The work done by Şabanoğlu in Ankara coincided with the work done by Özdemir in Ankara [27, 28]. During the 5-month period in which the experimental phase was carried out, *S. haemorrhoidalis* was observed during the fresh, swelling and decay stages. A partial increase in difference was observed in May compared to other months. Because *S. haemorrhoidalis* ideally lived in rainy, closed weather. This behavior made by Şabanoğlu and his current scientific papers was similar [28, 59].

4. CONCLUSION

Depending on the successful evolution of insects, the nutrition, ecology and ethology of each differ from each other. While forensic insects show common characteristics in the field of ecology and nutrition, their ethologies differ depending on ecological factors and rotting natural material. However, they support the contribution to decomposition processes and the development of ecological succession here. These situations play an important role in the elucidation of forensic cases. There are a number of factors that affect forensic entomological examinations. These;

- Geography has different types of insects in different regions.
- Insects have different behaviors.
- While changes in ambient temperature accelerate direct sunlight and high heat succession, sheltered and cold conditions have been found to delay this period.

• Variety, burial or partial burial in situations where the body is exposed in this process significantly slows down the succession process, in this case quite different entomological success. Therefore, in our research, rats have been left in order to obtain more efficient results. The decomposition stages of the species in our study differed (Table 7.).

In our study, Diptera species were observed in fresh and swelling stages. The density of the Coleoptera species have missed the larvae of the Diptera species. The Coleoptera species created a distinction in the decomposition time of the corpse, as they consumed the larvae of the Diptera species.

As a result, it was determined that flies belonging to Calliphoridae and Sarcophagidae families, which are the first wave of decomposition, arrived in the corpse within hours. It was observed that the second and third wave insects arrived in the later stages of the decomposition. These observations are similar to the insect succession sequence made by Smith in 1986 [36]. In 2001, Anderson stated that in addition to the insect succession ranking, insects enter into different behavioral states against ecological factors [60]. In this context; It was also observed that the insects detected within the scope of the study and effective in the decay process deviated from the existing ethological evaluations depending on ecological factors. Anderson (2001) evaluates the occurrence of such situations as normal.

Table 7. Decomposition stages of detected species

Detected species	Decomposition stage detected	Decomposition stage in the literature
<i>Calliphora vicina</i> (Robineau-Desvoidy, 1830)	Fresh, swelling and decay stage	Fresh, swelling and decay stage
<i>Calliphora vomitoria</i> (Linnaeus, 1758)	Fresh, swelling and decay stage	Fresh, swelling and decay stage
<i>Lucilia sericata</i> (Meigen, 1826)	Fresh and swelling stage	Fresh, swelling and decay stage
<i>Lucilia cuprina</i> (Wiedemann, 1830)	Fresh and swelling stage	Fresh, swelling and decay stage
<i>Sarcophaga haemorrhoidalis</i> (Fallen, 1817)	Fresh, swelling and decay stage	Fresh, swelling and decay stage

Decomposition length of *Rattus rattus* was taken into consideration to determine our working process. Accordingly, it was determined that the decomposition ended in 30 days by evaluating and observing the ecological environment. Velasquez (2007), in his study on *Rattus rattus* in Venezuela, found that the decomposition process was completed in an average of 30 days within ecological factors [61]. Rabbits were used as mammals in the studies of Keskin in 2013 and Başar in 2018. The duration and process of decay of the rabbit was determined by examining ecological factors [25, 62].

Smith (1986) and Anderson (2001) grouped the species identified in the corpse as necrophage, sarcophage, coprophage, dermatophage, predator, parasite and incidental species [36, 60]. In our study, members of Calliphoridae, Sarcophagidae families belonging to the necrophage group were determined. In the studies conducted by Karapazarlıoğlu on buried pig carcass in 2012 and Şabanoğlu - Özdemir on superficial pig carcass in 2007, the insect species and decay stages of Diptera and Coleoptera were similar [26, 27, 28]. In our study, the insect species and decay stages detected on *Rattus rattus* left superficially are in the same direction.

It was determined that *C. vicina*, *C. vomitoria* was active in April and May, and not active in June - July - August. Dinar and Topçular reported that these species live in temperate climates, especially in spring and autumn [47, 48]. Accordingly, our current study data differ from the literature information.

Decomposition stages in forensic entomological studies conducted by Gullan, Ament, Karapazarlıoğlu, Anderson, Goff, Tüzün, Şabanoğlu, Özdemir, Greenberg, Campobassa; It occurs in 4 steps [7, 11, 13, 14, 16, 18, 27, 28, 39, 40]. Within the scope of our study, it was observed that decomposition took place in 4 stages as fresh, swelling, decay and drying stages

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