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# THE INFLUENCE OF METHOMYL AND OPIOIDS ON THE DECOMPOSITION OF RABBIT CARCASSES AND INSECT SUCCESSION

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| Article info  | Abstract  |
|---|---|
| Received:       2024-07-04         Accepted:       2024-09-03         Published:       2024-12-31   | Internal decomposition starts in corpses after death<br>due to microorganisms. It is accompanied by the<br>arrival of forensic insects to the corpses in<br>succession shortly after death according to the starges   |
| <b>DOI-Crossref:</b> 10.32649/ajas.2024.151519.1310   | succession shortly after death according to the stages<br>of decomposition. Their arrival is affected by several  |
| Cite as:<br>Metab, A. H., and Ahmed, Q. H.<br>(2024). The influence of<br>methomyl and opioids on the<br>decomposition of rabbit<br>carcasses and insect succession.<br>Anbar Journal of Agricultural<br>Sciences, 22(2): 1411-1427.<br>©Authors, 2024, College of<br>Agriculture, University of<br>Anbar. This is an open-access<br>article under the CC BY 4.0<br>license<br>(http://creativecommons.org/lice<br>nses/by/4.0/). | factors, including temperature, humidity, and<br>geographical location. The exposition of rabbits to<br>toxin chemicals such as chemical pesticides and<br>drugs before death can delay the decomposition of<br>corpses or affect forensic insects. This study<br>examined the impact of methomyl and morphine on<br>the arrival of insects and the decomposition period<br>for treated buried rabbits in the rural area of the holy<br>city of Kadhimiya in Baghdad. Five species of<br>dipterans have been recorded as belonging to four<br>families, namely Calliphoridae, Sarcophagidae,<br>Muscidae, and Ulididae, and six species of<br>coleopterans belong to five families, that is<br>Histeridae, Dermestidae, Cleridae, Staphylinidae,<br>and Nitidulidae. Moreover, six hymenopteran<br>species belonging to three families were recorded,<br>i.e., Pteromalidae, Chalcididae, and Formicidae. The<br>study results show that rabbit bodies decompose<br>faster with morphine treatment, followed by the<br>control treatment and then the methomyl treatment<br>by 15.17, 17.17, and 20.17 days after death,<br>respectively. Also, insects are found in rabbit |

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corpses according to the stages of decomposition and, as such, can be used as forensic evidence to determine the cause of death.

**Keywords:** Forensic insects, Insect succession, Morphine, Rabbit carcasses, Postmortem.

# تأثير الميثوميل والافيون على تحلل جثث الأرانب وتعاقب الحشرات عليها

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## الخلاصة

تمر الجثث بعد الموت بمراحل تحلل عديدة نتيجة التحلل الداخلي بغعل الاحياء المجهرية ويرافق ذلك وصول الحشرات الى الجثث بعد الموت بفترة وجيزة وبتعاقب حسب مراحل التحلل. تتأثر وصول الحشرات الى الجثث بعدة عوامل منها درجات الحرارة والرطوبة والموقع الجغرافي. هنالك دور للمواد الكيميائية التي تعرض لها الارانب قبل الموت مثل المواد السامة كمبيد اللانيت او المخدرات كالمورفين في تأخير تحلل الجثث او تؤثر على وصول الحشرات المواد السامة كمبيد اللانيت او المخدرات كالمورفين في تأخير تحلل الجثث او تؤثر على وصول الحشرات الجنائية. هدفت هذه الدراسة الى تأثير مبيد اللانيت والمخدر المورفين في تأخير على وصول على وصول الحشرات الجنائية. هدفت هذه الدراسة الى تأثير مبيد اللانيت والمخدر المورفين على وصول الحشرات وفترة التحلل للأرانب المعاملة والمدفونة في المنطقة الزراعية في مدينة الكاظمية المقدسة. سجلت 5 على وصول انواع من النباب من رتبة شائية الاجنحة والتي تعود الى 4 عوائل وهي Sarcophagidae ، Calliphoridae بهديد الارانية في مدينة الكاظمية المقدسة. مجلت 5 انواع من النباب من رتبة غمديه الاجنحة والتي تعود الى 4 عوائل وهي Nucidide ، Staphylinidae ، Staphylinidae ، Staphylinidae ، Staphylinidae ، أنواع من رتبة غمدية الاجنحة والتي تعود الى 5 عوائل وهي Nitidulidae ، معاملة والمحت على النباب من رتبة عمديه الاجنحة والتي تعود الى 5 موائل وهي Nitidulidae ، معائل وهي المادانية ثم معاملة المواغ من رتبة غمائية الاجنحة والتي تعود الى 5 موائل وهي Nitidulidae ، Staphylinidae ، Cleridae ، الحام ، أوضحت الدراسة ان الجثث تحللت بشكل أسرع في معاملة المورفين تلتها معاملة المقارنة ثم معاملة الميد الكيميائي بعد عشائية الاجنحة والتي تعود الى 5 موائل وهي Nitidulidae ، Pteromalidae ، والتي تعود الى 5 موائل وهي الدراسة ان الجثئ معاملة المورفين تلتها معاملة الموازي تناية ما معاملة المقارنة ثم معاملة المينيائي بعد مشائية الاجنحة والتي تعود من ما لموت على التوالي. بينت الدراسة ان الجثن تما معاملة المينياني بعد مرام من الموت على المورفي علية العولي ولافي ما رتبة معاملة الموازي تماما المينيا معاملة المورفي ما مالي المينياني معاملة المون ما معاملة المورفين تلتها معاملة المقارية ثم معاملة المينياني بعد مرام المرامة ان الحثث. أمرحا موى مالموت على التوالي. بينت الدراسة ان الحشرات

كلمات مفتاحية: حشرات جنائية، تعاقب الحشرات، مورفين، جثث الارانب، فترة ما بعد الوفاة.

# Introduction

Forensic entomology can provide excellent evidence in support of forensic investigations (47). The use of insects in legal and criminal investigations is the field of forensic entomology, now known as medico-legal entomology, and controlled experiments can be used to study the expected sequence in which insects colonize

corpses (20). Human and animal carcasses are considered a food source and reproductive environment for forensic insects with about 400 species identified as found on corpses (29). Careful analysis of the results of insects living in the body can help reveal critical information in criminal investigations (15). Scientists use forensic entomology to estimate postmortem intervals, and this has emerged as a distinct and highly recognized branch of forensic science in which entomological evidence is deemed admissible by courts worldwide (35). Traditionally, the time of death has been determined and estimated by viewing the external appearance of cadavers, considering other conditions such as temperature, body coldness, eye changes, skin color, sagging muscles, stiffness, and others (21). However, any of these characteristics may be unobservable due to decomposition and deformation of carcasses. Forensic entomology is an essential evidential tool in studying insect succession waves, allowing for the calculation of the postmortem interval period (PMI) (43).

Insect analysis as a forensic tool has gained greater prominence due to insects being the first organisms to reach a decomposed body or crime scene (23). The succession and departure of insects on a cadaver is characterized by a unique sequence known as the insect succession wave, which develops on cadavers over time (29 and 30).

Information related to succession waves, which change based on geographic location, season, and environmental surroundings, as well as the developing larval stage, can be used to determine the elapsed time since death and the probable cause of death (3 and 46). Arthropods occupy an important position in the environment of corpses, where they are active and multiply and thus play a significant role in their decomposition (18). The presence of insect species such as adults, larvae, and pupae of flies (Dipteran) and beetles (Coleopteran) depends on biological preferences in terms of behavior and nutrition, as well as on the stage of decomposition and geographical location (50). The stages of cadaver decomposition were identified as fresh, bloated, active, advanced, and dry decomposition (24). Therefore, the typical sequence of insects occurs when each group is attracted to a specific stage of decomposition (48).

Necrophagous species of Coleoptera (beetles) and Diptera (flies) are highly recommended for entomotoxicological studies since they are the first to colonize a corpse and are frequently found at crime scenes. As the first arthropods to find and oviposit on a corpse, Calliphoridae, often known as blowflies (Order: Diptera), are common and crucial insects for forensic analysis (12 and 23). Forensic entomology depends mainly on the order of dipterans and coleopterans, which are the most important in the succession and sequence in the stages of decomposition of corpses (15). Therefore, the aims of this study are to record and determine the seasonal abundance of forensic insects found on rabbit corpses in the summer to provide insect data for criminal investigations in rural areas.

#### **Materials and Methods**

Location of the experiment: The study was conducted on an agricultural region containing date palm and citrus trees belonging to the holy city of Kadhimiya, which is located north of Baghdad governorate and on the west side of the Tigris River on longitude 44.317103 east and latitude 33.390097 north (Fig 1). Metal cages of 50x 50x50 cm dimension were placed and covered with a metal mesh with 2 cm holes that allowed forensic insects to enter. Each cage contained one rabbit carcass representing one replicate for each treatment. The cage was prepared for burial at a depth of 10 cm. The temperature and humidity data were obtained from the Ministry of Agriculture, Iraqi Agrometeorological Center, Abu Ghraib Climate Station.

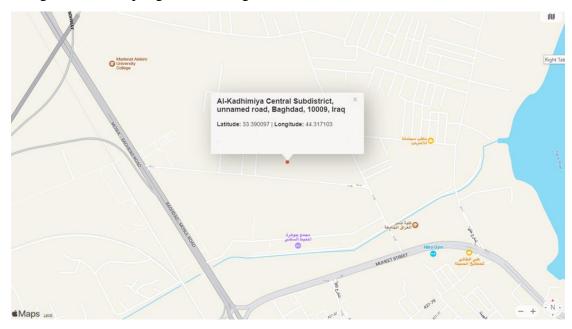


Fig. 1: Location of the experiment in an agricultural area within the holy city of Kadhimiya.

The experimental animals used in this study were disease-free domestic rabbits (*Oryctolagus cuniculus* L.) obtained from the Ministry of Health, National Center of Drug Control and Research (NCDCR). Nine rabbits were used and reared under animal house conditions at a temperature of  $25\pm2^{\circ}$ C, relative humidity of  $50\pm10$ , and a photoperiod of 14 h:10 h (light: dark). The average weight of the rabbits ranged from 1.75 - 2 kg. They were divided into three groups based on the experimental treatments (morphine, methomyl, and control treatments). Each group contained three rabbits and represented three replicates for each treatment.

Killing method and experiment design. The experiment was conducted in September 2023, using the pesticide Lannate-90% (active ingredient Methomyl) belonging to the carbamate pesticide group (WE-Young, Industrial and Trading Company, China). This pesticide has been used for many years and is widely employed in agriculture to control many agricultural insect pests. Methomyl was used because it is highly toxic to humans when directly or indirectly exposed to it at high concentrations and can lead to acute poisoning or death. Morphine was obtained from the Ministry of Health in Iraq and is considered one of the essential opioids belonging to the opioid group; it is used to relieve chronic and acute pain in hospitals and works directly on the central and peripheral nervous systems and the digestive system. It has several adverse side effects, the most important of which is addiction, with poppies being the primary source of morphine. Diethyl ether (ether groups) was also used. It is an anesthetic used in surgical halls and one of the common organic solvents in organic chemistry. It has a distinctive smell and was used after dosing rabbits with methomyl and morphine to accelerate their killing. It was also used in killing rabbits in the same way as the control treatment (3 and 10).

The first group of rabbits was injected with methomyl at a concentration of 3 mL/rabbit after dissolving 700 mg of methomyl in one liter of distilled water by intravenous injection (ear veins). The rabbits were injected after they were placed in a catcher designed for rabbits to prevent movement. Each rabbit was left after the infusion for 15 minutes and re-injected with the same concentration. The second group of rabbits was similarly injected with morphine at the concentration of 15 mg/kg of rabbit weight (each vial contained 10 mg of morphine). The concentration of methomyl and morphine was selected based on the lethal dose for each item, as commonly used in cases of suicide or misuse involving them (overdose). After injection, the rabbits were placed in a sealed plastic container-sized  $50 \times 50 \times 50 \times 50$  cm, and a cotton piece saturated with diethyl ether compound was attached inside the container to accelerate their death. The third group of rabbits was treated with diethyl ether compound only as a control treatment.

The nine rabbit corpses (three for each treatment) were transferred to the experimental agricultural site in Kadhimiya. Each rabbit carcass was placed inside the metal cage, buried in the soil at 10 cm depth, and covered with a piece of carton. The cages were distributed about 10-12 meters apart and secured to prevent predators from feeding on the carcasses. Observations were recorded on all cages, including the treatment name, the date of death, and placing of the treatment. Then, the arrival of insects to the rabbit carcasses was recorded daily, and the stages of decomposition followed for each treatment.

Insect Specimen Collection and Classification. A sticky white trap sized 30x20 cm was hinged at 1-meter height over each cage site to collect flies. The traps were inspected daily in the morning and evening to monitor the arrival of flies to the carcasses from the early stages of wet decomposition until dry decomposition. Meanwhile, beetles were collected using forceps, a soft brush, and a spoon from above and beneath the carcasses. Insect specimens (larvae) were placed in a glass vial sized 10 mL containing 70% ethanol alcohol and transferred to the laboratory. Flies and beetles were mounted and pinned on cork pieces and sorted by species to dipterans and coleopterans, and ants and wasps belonging to Hymenoptera were also collected. All insect specimens were morphologically identified using the taxonomic keys (4, 9, 16, 37 and 45). The insect specimens were then sent to the Natural History Research Center and Museum at the University of Baghdad to confirm the diagnosis. A Canon PowerShot ELPH115 digital camera was used to shoot photos of rabbit carcasses and insect specimens during the decomposition study.

Decomposition of rabbit carcasses and insect succession table. Based on the stages of carcass decomposition, forensic insect data were collected from the white sticky traps for the dipterans and manually for coleopterans and sorted in a table. The postmortem period (PMI) was determined according to the number of insects and distribution of the different insect species collected. Four stages were identified in the decomposition process: fresh, bloated, active decomposition, advanced, and dry decomposition. The criteria for the established decomposition of rabbit carcasses were correlated with the changes in carcasses observed under the experiment area conditions. Moreover, the specific characteristics of the carcasses were used to determine the stages in which the remains were found.

Statistical analysis. A Completely Randomized Block Design (CRBD) was used with three replications for each treatment. The analysis of variance (ANOVA) was conducted using Microsoft Excel and SPSS software statistics for Windows, version 26.0 (Armonk, NY: IBM Corp, Released 2019). The average and standard deviations (SD) were compared at a 5% significance level.

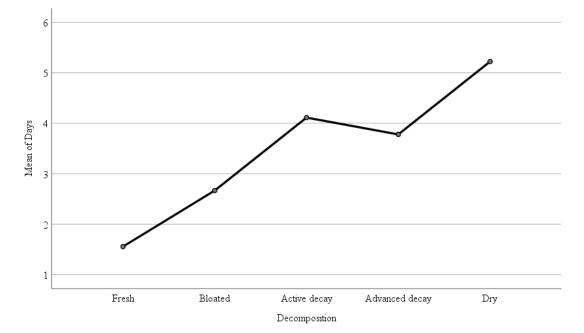
#### **Results and Discussion**

As seen in Table 1 and Figure 2, the rabbit bodies in the control treatment began to show the first signs of bloating after 2.33 days, while those in the methomyl and morphine treatments took longer, reaching 3.33 and 2.50 days, respectively. On the other hand, the period of active (wet) decomposition stage after the bursting of carcasses and exit of body fluids was 3.33 days in the morphine treatment, and 4.67 and 4.50 days in the methomyl and control treatments, respectively. There were no significant differences in the periods for the advanced decomposition stage at 3.33, 3.83, and 4.00 days for the morphine, methomyl, and control treatments, respectively. In addition, no significant differences were seen from the dry decomposition to the skeletal stages of the carcasses, averaging 4.83 days for both morphine and control treatments. In contrast, a considerable difference was recorded for methomyl treatment at 6.50 days. There were significant differences between morphine, methomyl, and control treatments in the number of days for the carcasses to reach the skeleton stage at 15.17, 20.17, and 17.17 days, respectively.

Insects provide essential entomological evidence in forensic investigations and are mainly used for estimating post-mortem periods. Forensic insects are attracted to corpses shortly after death and begin laying their eggs in the mouth, nose, ear, eyes, and anal regions of corpses and on wounds, as they participate with other factors in the analysis of corpses. Insects are attracted to corpses according to the different stages of decomposition as characterized by their high activity in each stage, namely fresh, bloated, active, advanced, and dry decomposition (Table 1). All these stages differ in the number of decomposition days according to the treatments carried out on the rabbit carcasses. Temperature, humidity, and geographical location can play an important role in accelerating the decomposition process in conjunction with the presence and abundance of insects in the environment of the carcasses. The decomposition stages are closely related to the succession of insects (2 and 9). The results show that the period for the decomposition of rabbit carcasses in the morphine treatment was shorter than for the methomyl and control treatments. This could be due to the rapid development of insects and the faster completion of their life cycles in morphine treatment, as indicated by (19 and 33). Poisoning with chemical pesticides has a lethal effect on insect larvae, especially the young fly larvae attracted to rabbit carcasses, which is why the period of analysis of the carcass was delayed (23).

| Table 1: Duration of rabbit carcass decomposition (average days $\pm$ SD) related |
|---|
| to the temperature and humidity in the holy city of Kadhimiya, Baghdad.           |

| Date       | Stage of       | Morphine  | Methomyl        | Control       | Max   | Relative |
|------------|----------------|-----------|-----------------|---------------|-------|----------|
|            | decomposition  | ±SD       | ±SD             | ±SD           | temp. | humidity |
| 03/09/2023 | Fresh          | 1.17±0.64 | 1.83±0.65       | $1.50\pm0.86$ | 41.42 | 54.86    |
| 04/09/2023 |                |           |                 |               | 43.09 | 47.21    |
| 05/09/2023 | Bloated        | 2.50±0.86 | 3.33±0.82       | 2.33±0.81     | 45.71 | 43.48    |
| 06/09/2023 |                |           |                 |               | 46.06 | 35.22    |
| 07/09/2023 |                |           |                 |               | 43.63 | 34.76    |
| 08/09/2023 |                |           |                 |               | 43.55 | 40.12    |
| 09/09/2023 | Active decay   | 3.33±0.82 | 4.67±0.81       | 4.50±0.87     | 41.59 | 51.96    |
| 10/09/2023 |                |           |                 |               | 41.97 | 47.12    |
| 11/09/2023 |                |           |                 |               | 40.15 | 53.72    |
| 12/09/2023 |                |           |                 |               | 40.80 | 51.59    |
| 13/09/2023 |                |           |                 |               | 43.89 | 42.08    |
| 14/09/2023 | Advanced       | 3.33±0.81 | 3.83±1.19       | $4.00\pm0.00$ | 38.67 | 43.77    |
| 15/09/2023 | decay          |           |                 |               | 38.18 | 37.49    |
| 16/09/2023 | _              |           |                 |               | 39.38 | 45.71    |
| 17/09/2023 |                |           |                 |               | 39.91 | 37.86    |
| 18/09/2023 | -              |           |                 |               | 40.38 | 42.92    |
| 19/09/2023 | Dry            | 4.83±0.65 | $6.50 \pm 0.86$ | 4.83±0.65     | 40.16 | 36.81    |
| 20/09/2023 |                |           |                 |               | 40.38 | 44.03    |
| 21/09/2023 |                |           |                 |               | 42.13 | 42.78    |
| 22/09/2023 |                |           |                 |               | 42.4  | 41.89    |
| 23/09/2023 |                |           |                 |               | 42.58 | 36.93    |
| 24/09/2023 |                |           |                 |               | 41.49 | 47.84    |
| Total deco | mposition days | 15.17     | 20.17           | 17.17         |       |          |



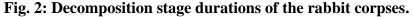


Table 2 shows that the presence of different species of insects belonging to the order Diptera, Coleoptera, and Hymenoptera on the rabbit carcasses varied according to the stage of decomposition. Each stage had particular species of insects, with the first three days characterized by the *Chrysomya megacephala* and *C. albiceps* species

of the family Calliphoridae that were almost the first arrivals in the corpses during the early hours after death. They are considered the most important in criminal investigations as insect evidence and in determining the postmortem period through the development of their species.

| Table 2: Forensic insect species on rabbit corpses according to the stages of |  |  |  |  |  |  |
|---|--|--|--|--|--|--|
| decomposition.  |  |  |  |  |  |  |

| Insect Species        | Decomposition stages/Postmortem days |         |   |        |   |          |   |   |     |    |    |    |     |
|-----------------------|--------------------------------------|---------|---|--------|---|----------|---|---|-----|----|----|----|-----|
| _                     | Fresh                                | Bloated |   | Active |   | Advanced |   |   | Dry |    |    |    |     |
|                       | 1                                    | 2       | 3 | 4      | 5 | 6        | 7 | 8 | 9   | 10 | 11 | 12 | 12+ |
| Chrysomya albiceps    | +                                    | +       | + | +      | + | +        | + | + | +   | +  | -  | -  | -   |
| Chrysomya megacephala | +                                    | +       | + | +      | + | +        | + | + | +   | +  | -  | -  | -   |
| Sarcophaga vagans     | -                                    | -       | + | +      | + | +        | + | + | -   | -  | -  | -  | -   |
| Musca domestica       | +                                    | +       | + | +      | + | +        | + | + | +   | +  | +  | +  | -   |
| Physiphora alceae     | -                                    | -       | - | +      | + | +        | + | + | +   | -  | -  | -  | -   |
| Saprinus subnitescens | -                                    | -       | - | +      | + | +        | + | + | +   | +  | -  | -  | -   |
| Dermestes maculatus   | -                                    | -       | - | +      | + | +        | + | + | +   | +  | -  | -  | -   |
| Dermestes frischii    | -                                    | -       | - | +      | + | +        | + | + | +   | +  | -  | -  | -   |
| Necrobia rufipes      | -                                    | -       | - | -      | - | -        | - | + | +   | +  | +  | +  | +   |
| Aleochara curtula     | -                                    | -       | - | -      | - | -        | + | + | +   | +  | +  | -  | -   |
| Carpophilus sp.       | -                                    | -       | - | -      | - | -        | + | + | +   | +  | -  | -  | -   |
| Nasonia vitripennis   | -                                    | -       | - | +      | + | +        | + | + | +   | -  | -  | -  | -   |
| Brachymeria podagrica | -                                    | -       | - | +      | + | +        | + | + | +   | -  | -  | -  | -   |
| Dirhinus hesperidum   | -                                    | -       | - | +      | + | +        | + | + | +   | -  | -  | -  | -   |
| Monomorium sp.        | +                                    | +       | + | +      | + | +        | + | + | +   | +  | +  | +  | +   |
| Crematogaster sp.     | -                                    | -       | + | +      | + | +        | + | - | -   | -  | -  | -  | -   |
| Camponotus xerxes     | +                                    | +       | + | +      | + | +        | + | + | +   | +  | +  | +  | +   |

(-) insect species not recorded, and (+) insect species recorded in the stage of decomposition of the rabbit carcasses.

The *Musca domestica* species of the Muscidae family was observed throughout the stages of decomposition, but its presence varied in numerical density depending on the stage of decomposition (Table 3). The species was found in the early stages of decomposition, especially in the active and advanced stages, with their numbers decreasing to a few individuals in the dry decomposition stage. The *Sarcophaga vagans* species from the Sarcophagidae family was noted in the first three stages (fresh, bloating, and active or wet decomposition). The *Physiphora alceae* species of the Ulidiidae family was also observed in the damp decomposition stage. It disappeared with the end of advanced decomposition or the beginning of dry decomposition, where it was observed to feed and reproduce on the manure produced by intestinal decomposition. This was identical to the presence of the species of ants, *Camponotus xerxes* from the Formicidae family and *Crematogaster* sp., from the beginning of carcass decomposition to the dry decomposition stage.

The density of forensic insects increased significantly after the bloating stage because of inflated and ruptured bodies. The skin breaks up, slogging from rabbit carcasses and releasing the decomposition gases. At this stage, the beetle species of the order Coleoptera, which belong to three families, were recorded successively. These were the *Saprinus subnitescens* species of the Histeridae family, and most common on cadavers, followed by the *Dermestes maculatus* and *D. frischii* species of

the Dermestidae family on cadavers at various stages of decomposition (Table 3). The *Necrobia rufipes* species belonging to the family Cleridae were the least commonly found on corpses.

|    |                     | Scientific name       | Family        | Order       |
|----|---------------------|-----------------------|---------------|-------------|
| 1  | Blow flies          | Chrysomya albiceps    | Calliphoridae | Diptera     |
| 2  | Blow flies          | Chrysomya megacephala | Calliphoridae | Diptera     |
| 3  | Flesh flies         | Sarcophaga vagans     | Sarcophagidae | Diptera     |
| 4  | House flies         | Musca domestica       | Muscidae      | Diptera     |
| 5  | Band-eyed Wingwaver | Physiphora alceae     | Ulidiidae     | Diptera     |
| 6  | Clown beetle        | Saprinus subnitescens | Histeridae    | Coleoptera  |
| 7  | Skin beetle         | Dermestes maculatus   | Dermestidae   | Coleoptera  |
| 8  | Skin beetle         | Dermestes frischii    | Dermestidae   | Coleoptera  |
| 9  | Checkered beetle    | Necrobia rufipes      | Cleridae      | Coleoptera  |
| 10 | Rove beetle         | Aleochara curtula     | Staphylinidae | Coleoptera  |
| 11 | Sap Beetles         | Carpophilus sp.       | Nitidulidae   | Coleoptera  |
| 12 | Parasitic wasp      | Nasonia vitripennis   | Pteromalidae  | Hymenoptera |
| 13 | Parasitic wasp      | Barchymeria podagrica | Chalcididae   | Hymenoptera |
| 14 | Parasitic wasp      | Dirhinus hesperidum   | Chalcididae   | Hymenoptera |
| 15 | Pharaoh ant         | Monomorium sp.        | Formicidae    | Hymenoptera |
| 16 | Acrobat ant         | Crematogaster sp.     | Formicidae    | Hymenoptera |
| 17 | Giant desert ant    | Camponotus xerxes     | Formicidae    | Hymenoptera |

 Table 3: Forensic insect species on rabbit carcasses according to the stages of decomposition.

The advanced putrefaction stage involving ammoniacal fermentation of the rabbit body attracted different species of insects. (6) indicated the presence of the *Aleochara curtula* species of the family Staphylinidae, which feeds on the tissues of the cadavers and on the larvae of blowflies (23). Beetles were also seen during the wet decomposition to the end of the dry decomposition stages, as noted by the presence of adult beetles and larvae of the *D. maculates*, *D. frischii*, and *N. rufipes* species. In contrast, the *Carpophilus* sp. species of the Nitidulidae family were recorded on rabbit carcasses as they exist and feed on carcasses in advanced decomposition, consistent with the findings of (37).

(37) also noted species of insect parasites found on the carcasses of rabbits, such as *Nasonia vitripennis* of the Pteromalidae family, which parasitize the pupae of the Muscidae family and Calliphoridae, as the female *N. vitripennis* lays eggs on the pupae of these types of flies. The life cycle of *N. vitripennis* can be used to estimate the postmortem period, consistent with the findings of (25). Similarly, the species *Barchymeria podagrica* and *Dirhinus hesperidum* parasitize several families belonging to the Diptera. Previous studies have recorded their parasitism on Calliphoridae, Sarcophagidae, and Muscidae families, which have medical and forensic importance (1).

Table 4 shows no significant differences in the population densities of flies in the sticky traps at the active decomposition stage, with the average for *C. albiceps* at 8.50, 9.67, and 9.00 individuals for the control, methomyl, and morphine treatments, respectively. In contrast, there were significant differences in the control treatment for the species *C. megacephala* at 8.67 individuals compared to 6.00 each for the

methomyl and morphine treatments. The species *Sarcophaga vagans* had the least numbers compared to the others in the stage of active decomposition, averaging 3.67, 2.67 and 1.67 individuals for the control, methomyl, and morphine treatments, respectively. For the species *Physiphora alceae*, the number of individuals was 4.67, 3.00, and 2.67 for all three treatments. The main species of flies visiting the carcasses at the active decomposition stage was *Musca domestica* with no significant differences in their average numbers at 13.67, 13.00, and 12.00 for the control, methomyl, and morphine treatments, respectively.

The results indicate that the *Saprinus subnitescens*, *Dermestes frischii*, and *Dermestes maculatus* species had numbers ranging from 6-10 individuals for all treatments. In contrast, *Necrobia rufipes* recorded fewer numbers than the other beetle species, averaging 5.00, 2.67, and 3.00 for the control, methomyl, and morphine treatments, respectively. Many previous studies have indicated that forensic insects are related to the temperatures of corpses, humidity, and geographical location. Higher temperatures lead to faster decomposition of corpses due to the effectiveness of microorganisms in conjunction with insects that lay their eggs and hatch into larvae which then feed on the body tissues. In this study, the first appearance and population density of forensic insects in the rabbit carcasses were recorded in all three treatments during September 2023. The duration of the decomposition under different coefficients was proportional to temperature, consistent with the study by (2).

| Table 4: The average density of insects on rabbit carcasses based on the |  |
|--|--|
| comparison of methomyl and morphine treatments, September 2023.          |  |

| Insect Species        | Treatment | Mean±SD    | Coefficient of variation | 95% Credible<br>Interval |       |  |
|-----------------------|-----------|------------|--------------------------|--------------------------|-------|--|
|                       |           |            |                          | Lower                    | Upper |  |
| Chrysomya albiceps    | Control   | 8.50±0.29  | 1.24                     | 8.00                     | 9.00  |  |
|                       | Lannate   | 9.67±0.88  | 0.16                     | 5.87                     | 13.46 |  |
|                       | Morphine  | 9.00±0.58  | 0.11                     | 6.52                     | 11.48 |  |
| Chrysomya megacephala | Control   | 8.67±0.33  | 0.07                     | 7.23                     | 10.10 |  |
|                       | Lannate   | 6.00±0.58  | 0.17                     | 3.52                     | 8.48  |  |
|                       | Morphine  | 6.00±0.00  | 0.00                     | 6.00                     | 6.00  |  |
| Sarcophaga vagans     | Control   | 3.67±0.33  | 0.16                     | 2.23                     | 5.10  |  |
|                       | Lannate   | 2.67±0.33  | 0.22                     | 1.23                     | 4.10  |  |
|                       | Morphine  | 1.67±0.33  | 0.35                     | 0.23                     | 3.10  |  |
| Musca domestica       | Control   | 13.67±0.88 | 0.11                     | 9.87                     | 17.46 |  |
|                       | Lannate   | 13.00±0.58 | 0.08                     | 10.52                    | 15 48 |  |
|                       | Morphine  | 12.00±0.58 | 0.08                     | 9.52                     | 14.48 |  |
| Physiphora alceae     | Control   | 4.67±0.88  | 0.33                     | 0.87                     | 8.46  |  |
|                       | Lannate   | 3.00±0.58  | 0.33                     | 0.52                     | 5.48  |  |
|                       | Morphine  | 2.67±0.33  | 0.22                     | 1.23                     | 4.10  |  |
| Saprinus subnitescens | Control   | 9.67±0.88  | 0.16                     | 5.87                     | 13.46 |  |
|                       | Lannate   | 7.00±0.58  | 0.14                     | 4.52                     | 9.48  |  |
|                       | Morphine  | 7.67±0.33  | 0.08                     | 6.23                     | 9.10  |  |
| Dermestes frischii    | Control   | 10.00±1.16 | 0.20                     | 5.03                     | 14.97 |  |
|                       | Lannate   | 7.67±0.33  | 0.08                     | 6.23                     | 9.10  |  |
|                       | Morphine  | 6.67±0.33  | 0.09                     | 5.23                     | 8.10  |  |
| Dermestes maculatus   | Control   | 8.67±0.33  | 0.07                     | 7.23                     | 10.10 |  |
|                       | Lannate   | 10.00±0.58 | 0.10                     | 7.52                     | 12.48 |  |
|                       | Morphine  | 9.00±0.58  | 0.11                     | 6.52                     | 11.48 |  |
| Necrobia rufipes      | Control   | 5.00±1.16  | 0.40                     | 0.03                     | 9.97  |  |
|                       | Lannate   | 2.67±0.33  | 0.22                     | 1.23                     | 4.10  |  |
|                       | Morphine  | 3.00±0.58  | 0.33                     | 0.52                     | 5.48  |  |

Corpses exposed to the sun decompose faster in the presence of higher temperatures. During September, the rabbit carcasses decomposed faster because the forensic insects were highly attracted to them in the warm and hot environment. (11) noted that temperature affects insect activity due to suitability and control overgrowth and reproduction rates. The nature of the forensic insects' attraction to carcasses varies with each stage of decomposition. During the fresh decomposition stage, five species of dipteran flies were observed in the carcasses, similar to the findings of (49). The first fly at the fresh stage of rabbit decomposition was *M. domestica*, as also noted by (8). The second species during the active decomposition stage was C. Albiceps followed by C. Megacephala. The adult meat fly Sarcophaga vagans was another dipteran species collected during the fresh decomposition stage. The reduced attraction of insect species during the active decomposition stage can be explained by the lack of odors emitted from the carcasses, making them extremely difficult to detect. It is also due to the presence of toxic compounds that kill newly hatched larvae feeding on cadavers treated with chemical pesticides or the bioaccumulation of pesticides in soil that affect emerging pupae (8 and 32).

There was a relationship between the percentage of decomposition of rabbit carcasses and the species of insects attracted to them. This explains the slow decomposition of carcasses in the methomyl treatment compared to the morphine or control treatments. This study showed that the number of forensic insect species present was mainly in the order of dipterans involving five species of flies. In comparison, the second group attracted to the carcasses belonged to the order of Coleopterans with four species of beetles recorded.

## Conclusions

This study showed no significant differences between the rabbit carcasses treated with methomyl and morphine and the control treatment in attracting forensic insects of the order Diptera and Coleoptera. Dipteran flies were the first to arrive at the carcasses on the first day after death, followed by beetle species of the Coleopteran order. The results also show that the duration of decomposition of the carcasses was extensive in the morphine treatment, followed by the control, and finally, the chemical pesticide treatment. The number of insect species on the carcasses varied from flies and beetles in the active decomposition stage, with various types of flies and beetles recorded in large numbers in the sticky traps and those collected by hand. This facilitates the process of collecting and adducing forensic evidence in the investigation of crimes.

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