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IMPACT OF PLASTIC BAG CONCEALMENT ON INSECT SUCCESSION PATTERNS AND DECOMPOSITION RATE OF RABBIT CARCASSES

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ABSTRACT

Insect succession on decomposing remains is widely used in estimate the post-mortem interval (PMI) in forensic investigations. However, perpetrators may conceal the body to destroy evidence and delay discovery, thereby limiting insect colonisation and altering the rate of decomposition. Despite its forensic relevance, the quantitative effects of plastic bag concealment on insect colonisation patterns, decomposition dynamics and fly developmental duration remain poorly understood, particularly under tropical conditions. This study investigated the effect of plastic bag concealment of rabbit carcasses on insect succession patterns and decomposition rate. Fifteen carcasses were divided into three groups: plastic bag-enclosed indoors, plastic bag-enclosed outdoors and exposed outdoor controls across five experimental trials. Insects were collected and identified, and decomposition progression and blowfly development from oviposition to adult emergence were recorded. A total of 2005 insects were collected, predominantly *Chrysomya megacephala*, *Chrysomya rufifacies*, and *Sarcophaga* sp. Blowfly species *Hemipyrellia ligurriens* and *Hypopygiopsis violacea* as well as ants and carrion beetles were observed only on exposed carcasses. Exposed carcasses decomposed in approximately eight days, whereas enclosed carcasses required 18-20 days. Fly oviposition on concealed carcasses was by 28-32 hours outdoors and 49-52 indoors, and development from egg to adult emergence was significantly delayed in concealed carcasses (291–340 hours) compared to exposed controls (197-205 hours). These findings demonstrate that plastic bag concealment delays insect colonisation, decomposition, and blowfly development, emphasizing the importance of considering enclosure effects in forensic PMI estimation for concealed remains.

Keywords: Forensic entomology; post-mortem interval (PMI); tropical environment; *Chrysomya megacephala*; *Chrysomya rufifacies*

ABSTRAK

Corak sesaran serangga pada mayat yang mereput digunakan secara meluas dalam penganggaran selang masa kematian (PMI) dalam pemyiasatan forensik. Walau bagaimanapun, pelaku jenayah mungkin menyembunyikan mayat untuk memusnahkan bukti, dan melambatkan penemuan, dan seterusnya mengehadkan kolonisasi serangga serta mengubah

kadar pereputan. Walaupun mempunyai kepentingan dalam bidang forensik, kesan kuantitatif penyembunyian mayat menggunakan beg plastik terhadap corak kolonisasi serangga dinamika pereputan dan pertumbuhan lalat masih kurang difahami, terutamanya di dalam keadaan tropika. Kajian ini menyiasat kesan penyembunyian bangkai arnab menggunakan beg plastik terhadap corak sesaran serangga serta kadar pereputan. Sebanyak 15 bangkai dibahagikan kepada tiga kumpulan, iaitu bangkai yang dibungkus beg plastik dalam persekitaran tertutup, bangkai dibungkus beg plastik dalam persekitaran luar serta bangkai terdedah di luar. Sebanyak 2005 ekor serangga telah dikumpulkan, dengan spesies dominan ialah *Chrysomya megacephala*, *Chrysomya rufifacies* dan *Sarcophaga* sp. Spesies langau lain, *Hemipyrellia ligurriens* dan *Hypopygiopsis violacea*, dan juga semut dan kumbang hanya ditemui pada bangkai yang terdedah. Bangkai yang terdedah mengalami pereputan lengkap dalam tempoh kira-kira lapan hari, manakala pereputan bangkai yang dibungkus beg plastik di persekitaran luar dan dalam masing-masing memerlukan antara 18 hingga 20 hari. Kelewatan oviposisi lalat pemakan bangkai didapati pada bangkai yang disembunyikan iaitu selama 28–32 jam di persekitaran luar dan 49–52 jam di persekitaran dalaman, dan kadar pertumbuhan lalat dari peringkat telur sehingga dewasa lebih panjang secara signifikan pada bangkai dibungkus (291-340 jam), dibandingkan dengan bangkai terdedah (197-205 jam). Penemuan ini menunjukkan bahawa bangkai dibungkus di dalam beg plastic terutamanya di persekitaran dalaman melambatkan proses kolonisasi serangga dan pereputan secara signifikan, di mana boleh memberikan kesan kepada anggaran minimum PMI di dalam kes-kes forensik yang melibatkan mayat yang disembunyikan.

Kata kunci: Entomologi forensik; selang masa selepas kematian (PMI); persekitaran tropika; *Chrysomya megacephala*; *Chrysomya rufifacies*

INTRODUCTION

Forensic entomology involves the use of insects and other arthropods associated with decomposing remains to assist in criminal investigations, particularly in estimating the minimum post mortem interval (minPMI) (Sharma et al. 2025). Estimation of the minPMI often relies on predictable patterns of insect colonisation and development on decomposing remains. However, these processes may be influenced by several intrinsic and extrinsic factors that can alter the rate and duration of decomposition. Intrinsic factors are associated with the characteristics of body itself such as age, body size, cause of death and body integrity, whereas, extrinsic factors involve environmental conditions such as temperature, ventilation, humidity and physical barrier including clothing, wrapping or concealment that may alter the microenvironment surrounding the remains (Campobasso et al. 2001; Malainey & Anderson 2020; Matuszewski 2021).

Blowflies (Diptera: Calliphoridae) are typically among the earliest insects to colonise decomposing remains. They are attracted to volatile compounds released during the early stages of decomposition and may arrive within minutes after death (Façanha et al. 2024). Adult flies deposit eggs or larvae on carcass, and the developing larvae went through a predictable developmental stage while feeding on decomposing tissues. These developmental patterns are commonly used to estimate the time since the remains were first exposed to the environment (Obafunwa et al. 2025). As decomposition progresses, insect succession becomes more important information for estimating minPMI (Matuszewski & Mądra-Bielewicz 2025).

In actual forensic cases, perpetrators may attempt to conceal or dispose of bodies to avoid detection and delay decomposition. Bodies or body parts may be wrapped or enclosed in

materials such as plastic bags, suitcases or containers (Geissenberger et al. 2024; Kotzé at al. 2021). Such concealment can restrict insect access to the remains, and alter the surrounding microenvironment, potentially delaying insect colonisation and modifying succession patterns. Consequently, these changes may ultimately affect the rate of decomposition and complicate the estimation of post mortem interval in forensic investigations (Alfsdotter & Petaros 2021; Pittner et al. 2020).

Although insect succession on exposed carcasses has been widely documented, studies have examined the effects of physical concealment on insect colonisation and decomposition processes remain limited. For instance, Taleb et al. (2022) investigated insect activity on carcasses wrapped in plastic bags placed outdoors under full sunlight. However, limited studies have quantitatively examined how plastic bag concealment influences insect colonisation patterns and decomposition dynamics across differential environmental settings, particularly under tropical conditions. Comparisons between concealed carcasses placed in outdoor and indoor environments remain insufficiently explored, despite their potential importance for forensic investigations.

In addition to documenting insect succession, understanding the developmental duration of key blowfly species under different concealment conditions is critical, as concealment can delay both colonization and larval growth, potentially affecting PMI estimations. Therefore, this study also aimed to investigate the duration from oviposition to adult emergence for the dominant blowfly species, *Chrysomya megacephala* and *Chrysomya rufifacies*, in addition to changes in insect succession patterns and decomposition rate of concealed rabbit carcasses in both indoor and outdoor environments. By analysing the associated insect communities and decomposition rate of rabbit carcasses this study provides insights into how physical concealment influences insect colonisation and highlights the implications of these changes for post-mortem interval estimation in forensic investigations.

MATERIALS AND METHODS

Study Site

The experiment was conducted at the Faculty of Built Environment, Universiti Malaysia Sarawak, Kota Samarahan, Sarawak (1°28'04"N 110°26'50"E, 6 m) (Figure 1). The study site was situated at the front of the faculty building, adjacent to the roadside. It included a security post and a small field measuring approximately 630 m². The experiment was conducted prior to the opening of the faculty, resulting in minimal human interaction. The faculty building was surrounded by peat swamp and a small river located about 200 meters northwest of the study site.

Figure 1. Study site was located at Faculty of Built Environment, UNIMAS (1°28'04"N 110°26'50"E, 6 m) as shown by red placemark on the map. The faculty was surrounded by swamp and a small river

Experimental Design

Fifteen domestic rabbits (*Oryctolagus cuniculus*), each weighing between 1.2 to 1.5 kg, were locally sourced for this study. The rabbits were prepared at the Entomology Research Laboratory, Faculty of Medicine and Health Sciences, UNIMAS, located 1.8 km southwest of the study site. They were sacrificed by cervical dislocation, a procedure carried out by a Medical Laboratory Technologist trained in animal handling, with animal ethics approval (UNIMAS/AEC/R/F07/050).

Rabbit carcasses were assigned to three decomposition conditions: concealed–outdoor, concealed–indoor and exposed–outdoor controls. The concealed carcasses were placed inside black plastic garbage bags, measuring 47 cm x 90 cm, which are commonly used as the household waste-disposal bags. Black bags were specifically chosen because they are frequently used in real forensic cases, where perpetrators conceal bodies or body parts using readily available domestic waste materials (Garcia 2026; Scholl & Moffatt 2017).

The concealed-indoor carcass was kept in a security guard's house, with all windows closed and small openings taped to simulate decomposition study in a closed environment. Meanwhile, the outdoor concealed carcass was placed on the ground, >20 m from the security guard post, while the exposed carcass was positioned >20 m from both the guard post and the outdoor concealed carcass. Figure 2 illustrates the experimental setup of this study. To prevent disturbance by scavengers such as dogs, cats, and other small mammals, while allowing free access for necrophagous insects, a slotted welded wire mesh cage (45 cm length x 42 cm width x 60 cm height) was used for both the outdoor concealed carcass and the exposed carcass. Five trials were conducted for this study throughout March 7, 2019 until September 22, 2020.

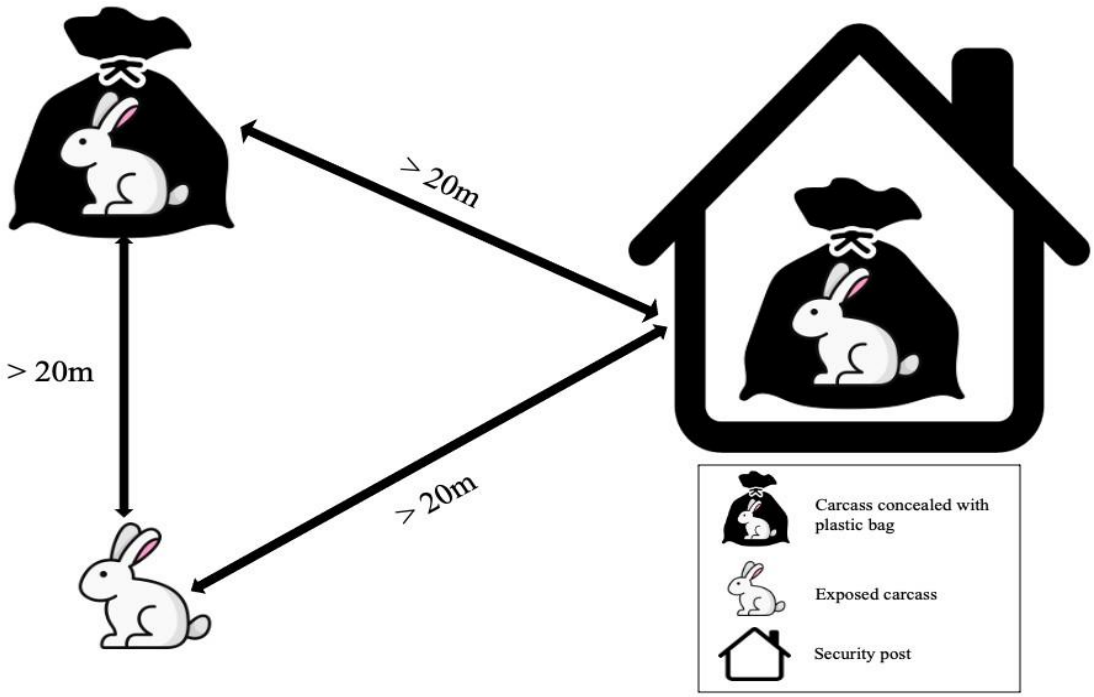

Figure 2. An illustration of the experimental setup and position of plastic bag-concealed rabbit carcasses in the study

Environmental and Insect Data Collection

Environmental data, including temperature, humidity, and rainfall, were recorded at the study site using a data logger equipped with a rain gauge meter (AMTAST, USA) at the study site. The carcasses were visited twice daily, in the morning and afternoon. Physical changes occurred during decomposition process was recorded, insects visiting and infesting the carcasses were collected. Flying insects were captured using a sweep net; while crawling adults were collected by hand with forceps. All insects were killed in a jar containing cotton balls soaked in ethyl acetate to preserve their appearance. The insects were then transferred to the lab, pinned, labelled, and examined under a stereomicroscope. Approximately 30 fly larvae colonizing the carcasses were collected. Half of the larvae were killed using hot water (80°C) and preserved in 70% ethanol, while the other half were reared to adulthood to facilitate species identification.

Data Analysis

The relative abundance of entomological data collected on carcasses in each group was calculated and tabulated in a table using Microsoft Office 2020. Since the trial of experiments were conducted in different months, the total number of insects collected in each group was compared across trials to determine seasonal variation and consistency of colonization patterns. The comparison of insect abundance between trials was carried out using one way Analysis of Variance (ANOVA) in SPSS version 27. Meanwhile, the environmental data between indoor and outdoor was compared using t-test analysis in SPSS version 27.

The comparison of total developmental stages of blowflies in exposed and concealed carcasses was compared using one-way ANOVA and followed by multiple comparison analysis, Tukey HSD test when significant difference was found. The infestation duration of

flies on carcasses was tabulated using Microsoft Office 2020. R statistical software (version 4.5.2) was used to assess the compositional dissimilarity of insects occurring on the carcasses across three different groups. The similarity of insect composition between groups was analyzed using the Bray-Curtis Dissimilarity Index (vegan package). This dissimilarity data was then visualized using a Principal Coordinate Analysis (PCoA) plot (ggplot package). Furthermore, a Permutational Multivariate Analysis of Variance (PERMANOVA) was performed to statistically compare the significant difference in insect composition between the groups, which revealed a highly significant difference (P<0.001).

RESULTS

Insect Composition and Abundance on Decomposing Rabbit Carcasses.

A total of 2005 individual insects were collected from carcasses across five experimental trials (Table 1). Three predominant species of flies were identified infesting the carcasses wrapped in plastic bags: *Chrysomya megacephala* (Family: Calliphoridae), *Chrysomya rufifacies* (Family: Calliphoridae), and *Sarcophaga* sp. (Family: Sarcophagidae). The highest number of insects collected was *Chrysomya megacephala* (50.7%), followed by *Chrysomya rufifacies* (33.7%) and *Sarcophaga* sp. (2.5%). Similarly, these species were predominant in control carcasses, with relative abundances of 46.8% and 28.5%, respectively, followed by ants, which contributed 19.9%. Other insects collected included *Hypopygiopsis violacea*, *Hemipyrellia ligurriens*, ants and carrion beetles, which were only present in exposed carcasses (control group).

The findings indicate that the highest number of insects was collected from the control group (total = 1108), followed by the concealed-outdoor group (total = 519) and the concealed indoor group (total = 316). The Bray-Curtis Dissimilarity were calculated on samples and visualised using a PCoA plot. PCoA plot showed clear separation of insect composition and abundance among the three groups of carcasses. Control samples clustered along the positive axis of PC1 indicating a distinct different insect community compared to concealed treatments. Meanwhile, the samples in concealed-indoor were on the negative end of PC1. The sample demonstrating a consistent but substantially different community compared to concealed-outdoor which separated primarily along PC2 (Figure 3). These results indicate a clear difference of insect composition between indoor and outdoor concealment conditions. PERMANOVA analysis confirmed that insect communities differed significantly among the groups, with the model explaining 98.36% of the variation (F=360.16, P=0.001).

Statistical analysis was conducted to compare the abundance of insects across all groups of carcasses over the five experimental trials. The analysis revealed no significant difference in the number of insects collected in control group [F (4,10) = 0.008, $P>0.05$], the concealed-outdoor group [F (4,10) = 0.016, $P>0.05$], and the concealed-indoor group [F (4,10) = 0.021, $P>0.05$] in between trials.

Table 1. The composition of insects associated with carcasses decomposition in control and plastic bag-enclosed group

Order/Family	Species	Control	Plastic Bag-Enclosed Carcasses		Total Insect	Relative Abundance (%)
			Outdoor	Indoor		
Diptera Calliphoridae	*Chrysomya megacephala*	519	**291**	**206**	1016	50.67
	Chrysomya rufifacies	316	201	159	676	33.71
	Hemipyrella ligguriens	22	0	0	22	1.09
	Hypopygiopsis violacea	15	0	0	15	0.77
Sarcophagidae	*Sarcophaga* sp.	10	40	0	50	2.49
Hymenoptera Formicidae	Unidentified	221	0	0	221	11.02
Coleoptera	Unidentified	5	0	0	5	0.25
		1108	**532**	**365**	**2005**	**100.0**

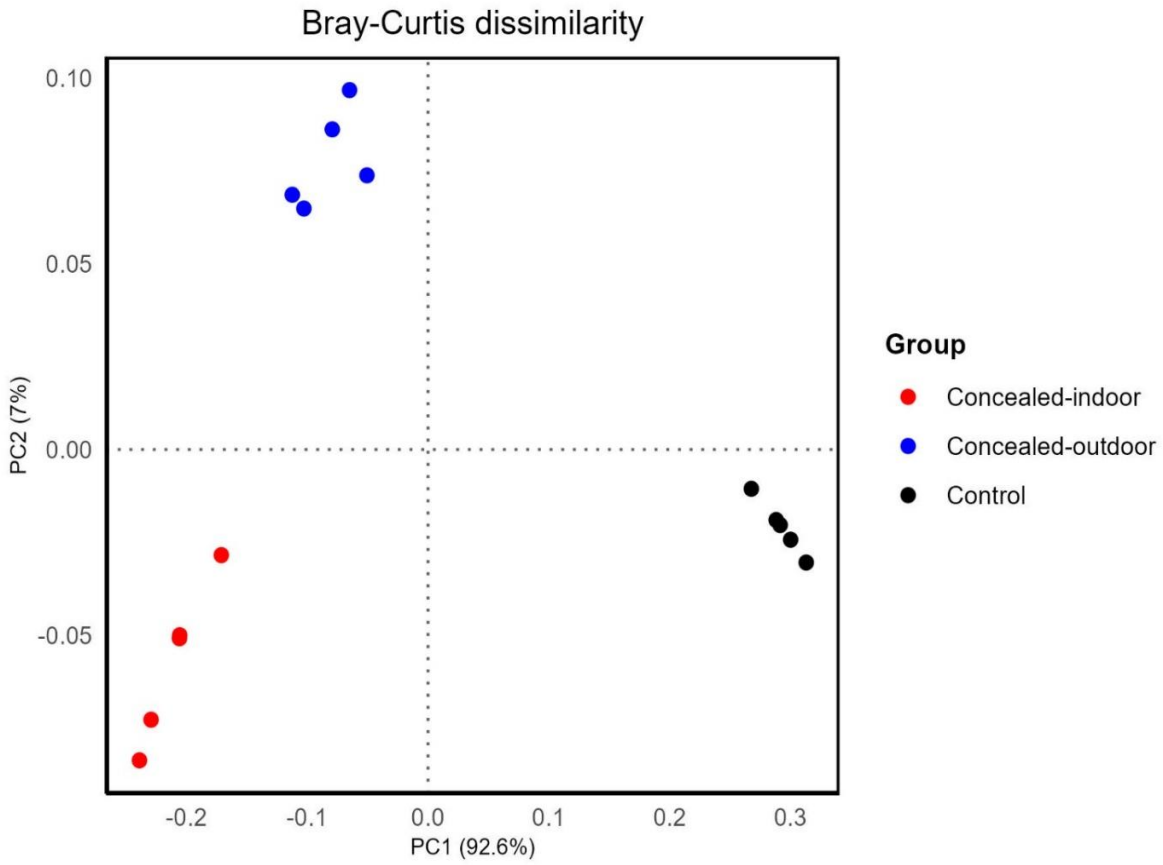

Figure 3. PCoA plot of insect communities on carcasses based on Bray-Curtis dissimilarity. Control (black), concealed-indoor (green) and concealed-outdoor (red) samples show clear separation along PC1 and PC2. PERMANOVA confirmed significant differences among groups ($F = 360.16$, $R^2 = 0.9836$, $p = 0.001$)

Decomposition of Rabbit Carcasses and Insect Activity

The decomposition process followed the expected sequence of fresh, bloated, active decay, advanced decay and dry stages across all carcasses. Concealment delayed progression: the fresh stage lasted for 1 day in control carcasses, 2 days for outdoor-concealed and 3 days for indoor-concealed carcasses. Bloated and active decay stages were similarly prolonged in concealed carcasses, with larvae first appearing later than in exposed carcasses. Pupation of third-instar larvae occurred earliest in control carcasses (day 5) and latest in indoor-concelaed carcasses (day 16). Dominant species varied slightly with concealment, with *C. megacephala* and *C. rufifacies* consistently present, while *Sarcophaga* sp. appeared later. In exposed carcasses, in addition to the two predominant *Chrysomya* sp., other insects such as *H. violacea*, *H. ligurriens*, ants (Formicidae), and coleopteran beetles were also present, whereas these taxa were absent in concealed carcasses. Overall, concealment significantly delayed decomposition and altered the timing of insect colonization. The decomposition stages in this study were described in Table 2, and the insect succession and abundance during each stage of decomposition were displayed in Table 3.

Environmental Condition

The daily ambient temperatures and relative humidity in both outdoors and indoors shows fluctuations throughout the study period. The environmental parameters recorded outdoors

were significantly different from those recorded indoors (*P<0.05*). Overall, the mean daily outdoor ambient temperatures show greater variability, ranging 30.5 to 35.9°C, whereas indoor temperatures remain stable, ranging from 28.1 to 32.5°C (Figure 4). The mean daily outdoor relative humidity ranged from 76.2 to 85.9%, while indoor relative humidity ranged from 79.0 to 87.8% (Figure 5).

Rainfall occurred intermittently throughout the study period. The highest recorded rainfall was during the third trial, accumulating to 209.8 mm. The least amount of rainfall was observed during the fifth trial, with only 82.8 mm recorded. Figure 6 shows the trend of rainfall from the first to the fifth trial.

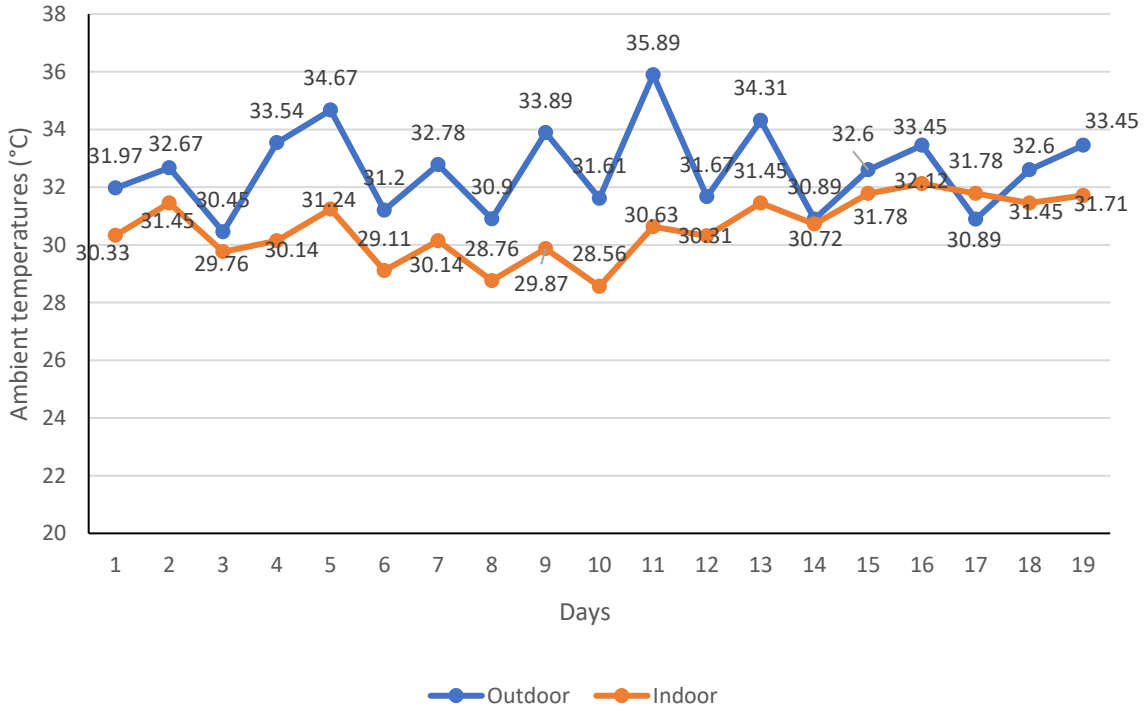

Figure 4. A graph showing ambient temperatures between outdoor and indoor environment. Outdoor ambient temperatures were significantly (*P<0.05*) higher than indoor temperatures

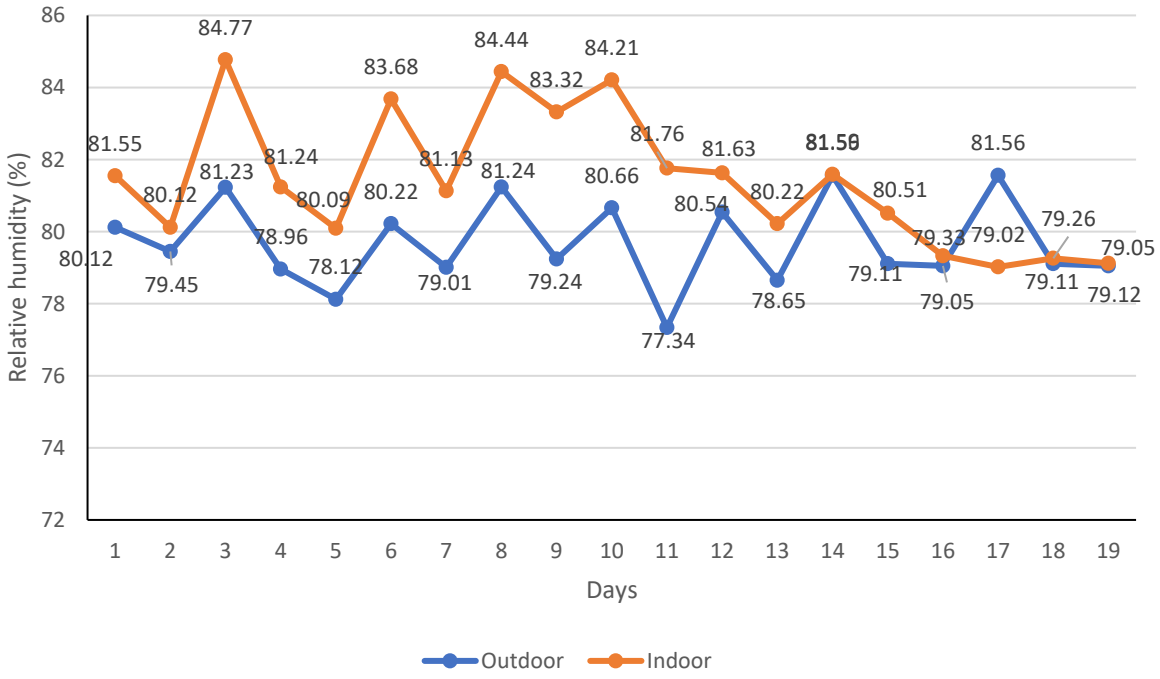

Figure 5. A graph showing relative humidity between outdoor and indoor environment. Indoor relative humidity was significantly ($P<0.05$) higher than outdoor humidity

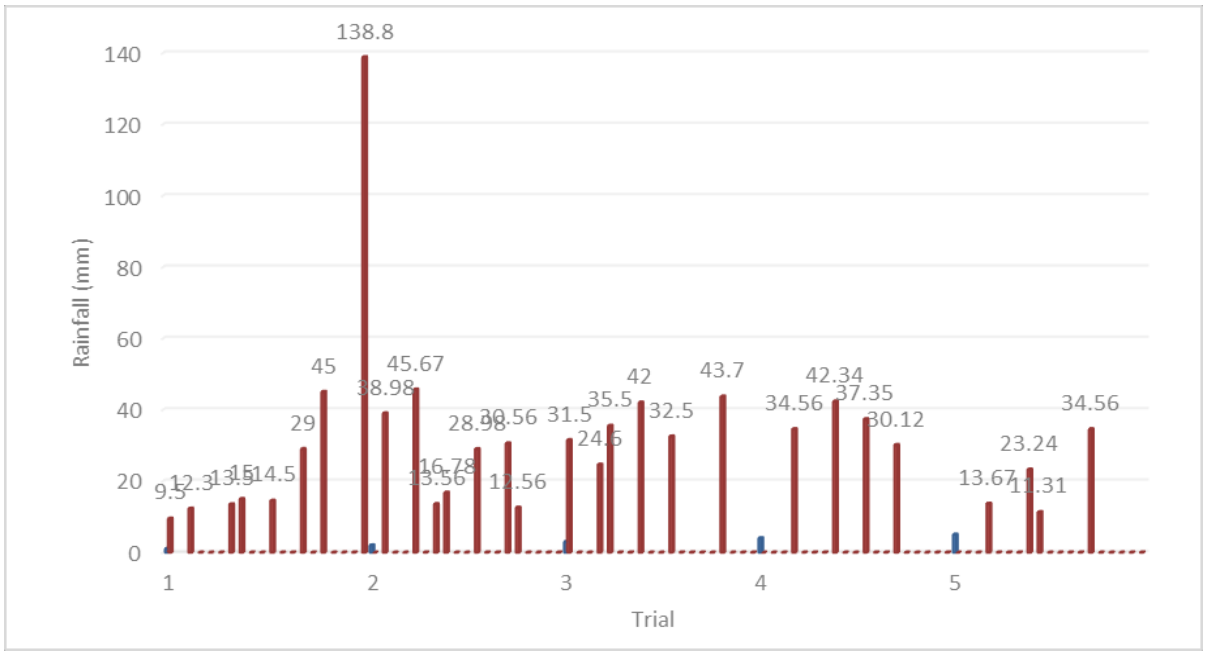

Figure 6. A graph showing rainfall recorded over study period. The highest rainfall recorded was 138.8 mm during the first trial of experiment

Duration of Blowflies Development on Concealed Rabbit Carcass

In this study, *C. megacephala* took approximately 2-5 hours to oviposit on the control carcasses (Table 4). There was a delay in the arrival of *C. megacephala* on the concealed carcasses, with the first eggs observed 28-31 hours and 49-51 hours after the carcasses were placed in outdoor and indoor locations, respectively. The time for the eggs to hatch was similar across groups, taking around 24-27 hours in the control group, and 18-21 hours and 22-24 hours for the outdoor and indoor concealed carcasses, respectively (Table 4). The mean development duration of *C. megacephala* in the control group was 197.4±1.82 hours. A longer development time was observed for *C. megacephala* on the concealed carcasses, taking approximately 291.6±1.67 hours and 339.6±1.82 hours for the outdoor and indoor groups, respectively.

A one-way ANOVA analysis was conducted to compare the duration from the first observation of eggs to adult emergence of *C. megacephala* across all groups. The analysis revealed a significant difference in development duration between the groups [$F_{(2,12)} = 8350.7$, $P<0.001$]. The development duration of *C. megacephala* was significantly delayed in indoor-concealed carcasses compared to outdoor-concealed and exposed carcasses. Similarly, the development duration was significantly delayed in outdoor-concealed carcasses compared to exposed carcasses.

Another forensically important blowfly infesting the carcasses was *C. rufifacies* (Table 5). Eggs were first observed 3-5 hours after the placement of exposed carcasses (control group). In contrast, *C. rufifacies* took longer to arrive at the concealed carcasses, with eggs observed 28-32 hours and 50-52 hours after placement in outdoor and indoor locations, respectively. First instar larvae were observed on control carcasses 25-27 hours after the first observation of eggs. For outdoor and indoor concealed carcasses, it took approximately 1820 hours and 22-25 hours, respectively, for the eggs to hatch. Detailed durations for each stage of development in all groups are provided in Table 5. Overall, the mean development duration for *C. rufifacies* from oviposition to adult emergence was 204.8±1.30 hours for the control group, compared to longer durations of 293.8±1.10 hours and 340.0±1.22 hours for the outdoor and indoor-concealed carcasses, respectively.

A one-way ANOVA analysis was conducted to compare the duration from the first observation of eggs to adult emergence of *C. rufifacies* across all groups. The analysis revealed a significant difference in development duration between the groups [$F_{(2,12)} = 16099.1$, $P<0.001$].

Table 2. Duration (days) of each decomposition stage observed in the different carcass treatment groups

Stage	Control (Exposed)	Outdoor-Concealed	Indoor-Concealed	Key Observations/Insects
Fresh	Day 0–1	Day 0–2	Day 0–3	Adult *C. megacephala* & *C. rufifacies* on control; no flies on concealed carcasses
Bloated	Day 1	Day 2	Day 3	Inflated abdomen; eggs on exposed vs. exterior of bag; odour attracts flies
Active Decay	Day 2–5	Day 5–9	Day 6–10	Larvae feed inside; maggot masses; tissue liquefaction; odour strong
Advanced Decay	Day 5–8	Day 10–15	Day 11–16	Reduced odour; pupation begins; ants & beetles present
Dry/Skeletonized	Day 8–9	Day 15	Day 16	Only bones and fur remain; indoor drying slower; some larvae pupate inside bag

Table 3. The insect succession pattern and abundance during each decomposition stages of carcasses in control and concealed groups

Group	Order	Family	Genus/Species	Decomposition Stages/Day-n				
				Fresh	**Bloated**	**Active Decay**	**Advanced Decay**	**Dry**
				1-2	**2-3**	**3-5**	**5-7**	**7-9**
Control	Diptera	Calliphoridae	*C. megacephala*	4	11	40	22	6
			C. rufifacies	2	7	21	13	3
			H. ligurriens	-	-	2	1	-
			Hy. Violacea	-	-	2	-	-
		Sarcophagidae	*Sarcophaga* sp.	-	-	2	-	-
	Hymenoptera	Formicidae	Unidentified	2	5	19	6	4
	Coleoptera	Unidentified	Unidentified	-	-	-	-	-
				1-3	**4-5**	**6-10**	**11-15**	**16-21**
Carcass Enclosed (Outdoor)	Diptera	Calliphoridae	*C. megacephala*	1	4	31	11	2
			C. rufifacies		3	23	7	1
		Sarcophagidae	*Sarcophaga* sp.		-	3	2	0
				1-4	**5-6**	**7-12**	**13-18**	**19-24**
Carcass Enclosed (Indoor)	Diptera	Calliphoridae	*C. megacephala*	-	3	22	9	1
			C. rufifacies 0	-	2	19	4	1

Table 4. The development stages and duration of *Chrysomya megacephala* in control and concealed carcass groups

Group	***C. megacephala* Development Duration (Hours)**							**Average Developmental Duration (Mean±SD)**
	Eggs	**1st Instar Larvae**	**2nd Instar Larvae**	**3rd Instar Larvae**	**Post-Feeding**	**Pupae**	**Adult**	
Control	**Day 1** 2-5	**Day 2** 24-27	**Day 2** 7-9	**Day 3** 16-17	**Day 4** 23-25	**Day 5** 23-25	**Day 9** 95-97	197.4±1.82
Plastic Bag-Enclosed Outdoor	**Day 2** 28-31	**Day 3** 18-21	**Day 3** 7-8	**Day 4** 17-19	**Day 5** 29-30	**Day 6** 20-22	**Day 13** 164-168	291.6±1.67
Plastic Bag-Enclosed Indoor	**Day 3** 49-51	**Day 4** 22-24	**Day 5** 23-25	**Day 6** 23-26	**Day 7** 24-26	**Day 8** 22-25	**Day 15** 168-170	339.6±1.82

Table 5. Observation on developmental duration of *Chrysomya rufifacies* collected from rabbit carcasses enclosed in plastic bag at indoor and outdoor locations during the five trials of experiment

Group	***C. rufifacies* Development Duration (Hours)**							**Average Developmental Duration (Mean±SD)**
	Eggs	**1st Instar Larvae**	**2nd Instar Larvae**	**3rd Instar Larvae**	**Post-Feeding**	**Pupae**	**Adult**	
Control	**Day 1** 3-5	**Day 2** 25-27	**Day 2** 7-8	**Day 3** 16-17	**Day 4** 27-29	**Day 5** 26-28	**Day 9** 96	204.8±1.30
Plastic Bag-Enclosed Outdoor	**Day 2** 29-32	**Day 3** 18-20	**Day 3** 7-8	**Day 4** 17-20	**Day 5** 28-30	**Day 6** 20-22	**Day 13** 168-169	293.8±1.10
Plastic Bag-Enclosed Indoor	**Day 3** 50-52	**Day 4** 22-25	**Day 4-5** 23-25	**Day 5-6** 23-25	**Day 6-7** 24-25	**Day 8-10** 21-25	**Day 15** 168-170	340.0±1.22

DISCUSSIONS

Forensic entomology commonly estimates the post-mortem interval using two principal approaches: insect developmental data and insect successional patterns (Byrd & Tomberlin 2020). Both approaches can be affected by physical barriers that restrict insect access to remains (Malainey & Anderson 2020). In the present study, plastic bag concealment significantly influenced insect colonisation, decomposition rate and fly development on rabbit carcasses placed in outdoor and indoor environment. Concealed carcasses decomposed considerably more slowly than exposed carcasses, highlighting the importance of considering concealment effects when interpreting entomological evidence in forensic investigations.

Decomposition stages generally follow a predictable sequence of stages characterised by distinct physical changes and insect activity depending on the animal models and geographic region (Payne 1965; Villet 2011). In present study, the decomposition stages were categorised into five stages which are fresh, bloated, active decay, advanced decay, and dry remains, based on description by Goff (1993). Although this sequence was consistent across all treatments, the rate of progression differed substantially between exposed and concealed carcasses, indicating that concealment modified the normal decomposition course. One major factor contributing to the slower decomposition observed in concealed carcasses was the reduced diversity and abundance of necrophagous insects. Exposed carcasses attracted wider assemblage of insects, including blowflies, sarcophagid flies, ant and carrion beetles. In contrast, concealed carcasses were visited primarily by two blowfly species, *C. megacephala* and *C. rufifacies*, with the lowest species richness recorded in indoor concealment. These findings support previous studies indicating that physical barrier such as plastic wrapping restrict the dispersal of decomposition odours and limit access to the carcass (Brownlow et al. 2023; Teo et al. 2021). Reduced insect activity can consequently slow tissue breakdown and prolong decomposition duration.

Blowflies are typically among the first insect to colonise decomposing remains, particularly during the early stages when carcasses release volatile compounds that attract necrophagous species (Grassberger & Frank 2004). Likewise, a study conducted in the coastal region of Mukah, Sarawak, found that *C. megacephala* was the predominant blowfly species colonizing rabbit carcasses (Mohd Allif et al. 2024). In this study, *C. megacephala* and *C. rufifacies* rapidly colonised exposed carcasses and became the dominant species throughout decomposition. Similar patterns have been documented in other regions, although the identity of primary colonisers may vary geographically. For example; *C. albiceps* has been reported as the earliest coloniser in Nigeria (Emeka 2020), while sarcophagid flies dominate early colonisation in some parts of China (Shi et al. 2009). These differences highlight the importance of regional entomological data in forensic casework.

In contrast to exposed carcasses, concealment significantly delayed blowfly oviposition. Eggs were deposited within minutes to hours on exposed carcasses, but were delayed by more than a day on concealed carcasses and nearly two days when carcasses were concealed indoors. Similar delays have been reported in experimental studies where remains were wrapped or otherwise enclosed (Campobasso et al. 2001; Taleb et al. 2022). One explanation for this delay is the restricted release of decomposition odours and the limited physical access to the carcass. Necrophagous flies rely heavily on volatile compounds produced during decomposition to locate suitable oviposition sites, and barriers that restrict gas diffusion can significantly reduce carcass detectability (Campobasso et al. 2001; Greenberg 1991). Experimental studies have further demonstrated that reduced odour diffusion surfaces and

smaller access points can substantially delay fly access and oviposition even when insects are present in the surrounding environment (Charabidze et al. 2015). Consequently, concealment disrupts host-finding behaviour and delays colonisation (Amendt et al. 2011; Brownlow et al. 2023), which may lead to underestimate of PMI if concealment is not taken into account during forensic investigations.

Beyond delaying colonisation, concealment also influenced the developmental duration of blowfly larvae, which is the predominant flies infesting on the carcass in this study. *C. megacephala* and *C. rufifacies* was significantly prolonged in concealed carcasses, particularly those located indoors. This extended developmental period can be partially attributed to delay oviposition, as larval development begins later when colonisation is postponed. In addition, enclosed environments may create altered microclimatic conditions such as reduced oxygen availability, restricted airflow, and accumulation of decomposition by-products, which can be negatively affect insect growth and development (Brownlow et al. 2023; Pakosh & Rogers 2009). These findings suggest that concealment influences decomposition through two interacting mechanisms: delaying the initial colonisation of necrophagous insects and altering the subsequent developmental rate of larvae. Both factors should therefore be considered simultaneously when estimating PMI in cases involving concealed remains, particularly in tropical environments where insect activity and decomposition processes are typically rapid.

Environmental conditions also differed between indoor and outdoor settings, contributing to the variation in decomposition observed in this study. Indoor environments typically experienced reduced temperature fluctuations, limitedairflow, and restricted insect access, all of which may slow decomposition processes (Anderson 2011; Voss et al. 2008). Previous research has demonstrated that carcasses placed indoors can decompose substantially more slowly than those outdoors (Brownlow et al. 2023; Cammack et al. 2016). Similar observations were reported by Zuha et al. (2016), where rabbit carcasses placed indoors decomposed more slowly due to stable temperatures and reduced insect colonisation.

Temperature is critical factor governing insect growth and decomposition processes. Because insects are poikilothermic organisms, their metabolic and developmental rates depend strongly on environmental temperature (Byrd & Tomberlin 2020). Warmer conditions generally accelerate larval growth, whereas cooler temperatures slow development (Angilletta et al. 2004). In the present study, blowfly development occurred more rapidly on exposed carcasses where temperatures were higher than in indoor concealed environments. Similar temperature-dependent development patterns have been reported for *C. megacephala* (Gabre et al. 2005) and *C. rufifacies* (Day & Wallman 2006).

Humidity may also influence decomposition dynamics. Extremely low humidity can lead to desiccation and mummification, whereas excessive moisture may slow tissue breakdown by promoting wet decomposition (Campobasso et al. 2001; Goff 2009). In the present study, plastic bag concealment likely trapped moisture within the enclosure, prolonging decomposition. When larvae eventually penetrated the plastic bags, the formation of maggot masses further increased internal temperature and humidity through metabolic heat and moisture production (Weger et al. 2020), creating a highly saturated microenvironment.

Overall, the findings of this study demonstrate that plastic bag concealment significantly alters insect colonisation patterns, delays blowfly oviposition, prolongs larval development, and slows the overall decomposition process, particularly under indoor conditions. These effects have important implications for forensic investigations, as

concealment may lead to delayed insect activity and potentially inaccurate PMI estimations if not properly considered.

CONCLUSIONS

The study highlights the impact of plastic bag concealment on insect succession patterns, fly development duration and carrion decomposition rate. The findings indicate that carcasses concealed in plastic bags decomposed significantly slower than exposed ones. In between outdoor and indoor environment, the decomposition of indoor-concealed carcasses was further delayed compared to outdoor-concealed carcasses. Additionally, the delay of decomposition rate might be due to alteration of succession pattern and colonisation of insects. These results emphasize the importance of considering physical concealment when interpreting entomological evidence. However, due to the differences in body size and mass between animal model and human cadavers, PMI estimation based on this model should be interpreted with caution.

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AUTHORS DECLARATIONS

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Conflict of Interest
The authors declare that they have no conflict of interests.

Ethics Declarations
Approval from Animal Ethic Committee, the Faculty of Medicine and Health Sciences, UNIMAS, Kota Samarahan **(**UNIMAS/AEC/R/F07/050) was obtained prior to the commencement of this study.

Data Availability Statements
My manuscript has no associated research data.

Author's Contributions
Robin Maramat was responsible for investigation, writing of the original draft and reviewing and editing manuscripts. Marlini Othman contributed to statistical analysis and was involved in reviewing and editing the manuscripts. Nor Aliza Abdul Rahim was responsible for conceptualization, supervision and reviewing and editing the manuscript. All authors read and approved the final manuscript.

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