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Comparative Study of Arthropod Fauna on Exposed carrions Across the Vertebrate Classes

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ABSTRACT: Arthropod fauna and succession on exposed carcasses of representative of the vertebrate classes were determined. *Parachana obscura*, *Bufo temporalis*, *Agama agama*, *Gallus gallus domestica* and *Sus scrofa* were representatives of the classes Osteichthys, Amphibia, Reptilia, Aves and Mammalia respectively. A total of 23 arthropod species were found on the carrions from 19 families, 8 orders and 4 classes of the phylum Arthropoda. The order Diptera was the predominant group, the Hymenopterans ranked second and coleopterans the third. Responsible for carrion degradation were the families Calliphoridae, Sarcophagidae, Dermestidae, Cleridae, Staphylinidae and Histeridae. These families are considered to be of forensic importance. Coleopterans were found on carrions of the higher classes; Aves, Mammalia and absent or very few on the lower classes, Reptilia and Amphibia. However, the dipterans were found on all the carrions across the classes. There were variations in the dipterans found on the carrions as they tended to increase in abundance as we go higher across the vertebrate classes.

Key words: Carrion, Arthropod, classes, flies carcass.

Introduction

Since insects can be found in a wide variety of locations, the knowledge of their presence on dead organisms can be used to predict the time of death, cause and circumstances surrounding the death of the organism. This is referred to as medico-legal forensic entomology. Forensic entomology is the application of the study of insects and other arthropods to legal issues and can be divided into three components. Urban Forensic entomology: Legal proceedings involving insects and related animals that affect man-made structures and other aspects of the human environment. Simply put, it relates to litigations and civil law actions involving arthropods in dwelling or as house and garden pest. Stored-products forensic entomology involves: Proceeding involving insects infecting stored products or contamination of a wide range of commercial products, such as insect/arthropods in candies, ketchups, soft drinks, beers etc.

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It also includes quarantine procedures on stored imported grains. Medico- Legal Forensic Entomology: encompasses that aspect surrounding felonies especially violent crimes such as murders, suicide, rape, violations in form of physical abuse, child abandonment, drug/contraband traffic (Keh, 1985), Catts and Goff (1992).

The post mortem interval (PMI) is the period between death and time of discovery of a corpse. The theory behind estimating the PMI is based on the developmental rates and the successional ecology of specific insects that on corpses. This field of study has been found very useful in legal tangles in courts of law (Catts and Goffs, 1992; Wolff *et al.*, 2001). The key element in investigations are the postmortem interval (PMI), movement of the corpse, manner and cause of death, and association of suspects with death scene, may all relate to insect/arthropod occurrence and activities.

In the absence of fresh maggot samples, forensic entomologist can use the knowledge of the cephalopharyngeal skeleton of the maggot concerned to estimate the PMI. Lui and Greenberg (1989) worked out the cephalopharyngeal skeleton of a number of calliphorids as well as the three larval instars of each calliphorids as the three larval instars of each calliphorid.

Fauna composition may also be affected by certain environmental factors which include seasonal period, soil type, altitude, relative humidity, rainfall and most importantly temperature (So and Dudgeon, 1990; Galloway *et al.*, 1989; Mann *et al* 1990).

Full-blown forensic entomology did not start till 1988 (Ekanem and Usua 1997) in Nigeria; However, the knowledge of forensic entomology has not been effectively applied. They look for clues such as bullets, knives, shells and so on and no mention of the corpse fauna. This is attributed to the lack of knowledge of forensic entomology by the investigating bodies.

The earliest record of the use of forensic entomology according to Keh (1985) is Sung Tzu's (1235 AD) discussion in a book titled "The Washing away of Wrong". In Europe, forensic entomology started about 150 years ago with works of Brouarel and Yovanovitch (Keh, 1985). However, the father of forensic entomology is a man called Megnin who carefully detailed the predictable succession of arthropods associated with a decomposing corpse.

Arthropod fauna in or on carrion was classified into several ecological categories by Catts and Goff (1992). They includes necrophages, omnivores, parasites and predators of the necrophagous species and incidentals or adventurers.

Galloway *et al.* (1989), Mann *et al.* (1990); So and Dudgeon (1990); Shean *et al.* (1993) confirmed that carrion decomposition rate and arthropod succession are influenced by many factors which include temperature, humidity, rainfall and abundance of insects. Of these environmental factors, temperature has been shown to be of great importance. Tantawi *et al.* (1996), proposed four different decomposition stages of carrion; fresh, bloated, decay and dry, which involves to series of activities and changes in carrion.

The fresh stage also known as the initial decay is the first stage in carrion decomposition. It involves microorganisms already present in the corpse, though not all microorganisms are involved in the decomposition process, as some die almost immediately after the death of their host. The bloated stage also called putrefaction stage is the second stage. This usually begins 24hours after death. Amidst strong odour and decay, the third stage, the decay stage also known as the black putrefaction stage begins. This stage is divided into 2 stages, the active decay and post or advanced decay. These stages terminate in an almost dry carcass and in the fifth stage; slow dry decay completes the process leaving only bones and sometimes hardened skin. A 40kg pig resembles a human body in its fat decomposition, cover of hair and ability to attract insects. These factors made pigs the next best thing to human when it comes to understanding the process of decay of the human body. Arthropods that usually infect animals should be considered on the events of death. Gonzales *et al.* (1954) noted that lice will remain alive on a corpse for 3 to 6 days and that if all lice are dead, then death of the person must have taken place more than six days previous to discovery of corpse. This concept has merit and could include head and crab lice (*Pthirus pubis*). The use of ectoparasite such as follicle mites (*Demodex* sp), itch mite (*sarcoptes* sp) as benchmarks are yet to be authenticated. Payne and King (1991) consider dipteran and coleopteran to be of prime importance comprising about 65% of total fauna.

On the death of an organism, the first thing that happens is that the body temperature starts to fall. Before the temperature in the body core drops, a temperature gradient must be established from the outside to the core. After this gradient has become established, the body temperature will drop at a theoretically predictably rate. After the onset of putrefaction, the body temperature

will increase again, due to the metabolic activity of the bacteria and other decomposing organisms.

The objectives of this work include; (1) Determining and establishing arthropod successional pattern and their larva developmental stages on carcasses of representatives of vertebrate across the vertebrate classes; (2) Comparing the arthropod fauna and degradation pattern of representative of vertebrates across the vertebrate classes; and (3) Comparing the succession pattern of arthropod fauna of exposed carrion of animals across the vertebrate classes.

Materials and Methods

Field studies of carrion decomposition and associated fauna of buried representative species across the vertebrate classes were conducted.

The experimental site measured approximately 10m² by 10m². Daily ambient temperature was recorded with a minimum and maximum thermometer. The mean temperature during the experiment was recorded at 31.91°C. In each experiment, three replicates were used for each class across the vertebrate group. Three replicates of each class were exposed while three other replicates were buried. The following species were representatives for each class.

Class Osteichthyes – *Parachanna obscura* (Snakehead fish) weighing a mean of 0.11kg.

Class Amphibia – *Bufo temporalis* (toad) weighing a mean of 0.025 kg.

Class Reptilia – *Agama agama* (Agama lizard) weighing a mean of 0.037kg.

Class Aves – *Gallus gallus domestica* (domestic chicken) weighing a mean of 4.51kg.

Class Mammalia – *Sus scrofa* (domestic pig) weighing a mean of 11.75kg.

These animals were killed by gassing with chloroform to avoid external bleeding and to maintain their bodies intact. Immediately after killing, some of the carcasses were buried and some exposed. The buried carcasses were buried individually in holes about 30cm deep and placed on their left side.

Ultimate care was the order of the day as samples were collected in an orderly manner and samples of individuals of each class were noted. Sites of carcasses were monitored and daily collections were made for the exposed carrions. Flying insects were captured with a standard sweep net. Egg samples were collected and placed in 70% ethanol. The various larva instar and pupa of flies were collected with a pair of forceps. Small fragile insects were collected with an artist paint brush moistened with alcohol. Samples of the soil beneath the carcasses were collected with putting trowel in search for post feeding larva and puparia of flies and other soil arthropods. These soil arthropods were extracted from the soil by processing the soil samples with a Berlese – Tullgren funnel.

Samples of larva collected were reared to adult stage in the laboratory for species identification and to monitor the animal time of pupation and exclusion. Pupae were also collected and put in an insect cage floored with dampened sawdust.

Other arthropods were collected and killed with chloroform. The samples were sorted and preserved using appropriate methods. The hard bodies arthropods like beetles and ants were preserved in 70% ethanol. The data collected were analysed using the Two-Way Analysis of Variance.

Results

A total of 23 arthropod species were found on the carrions. They spread over 19 families, 8 orders and 4 classes of the phylum arthropoda (Tables 1& 2).

Arthropod activities began on the carrions the first day with the dipteran flies arriving just hours after the animals were killed. The flies were mostly attracted to the fish while the pig was second in line.

Ants already present on the soil were attracted to the site as well. They were the next set of insects at the sits and were present from day 1. They came in large numbers and remained till exhaustion of resources.

The decomposition stages of the animals (Fig 1) slightly varied in the latter stage but the fresh decay was just for one day. However, the fish, frog and lizard had almost the sample duration for bloated, decay and dry stages of decomposition. The difference was seen in the pig whose decay stage extended to the 9th day.

Feeding activities of the dipteran flies and their larvae broke open the body and ended the bloated stage. The decay stage of all the carrions was marked by increased species richness of arthropod. A total of 531 pre-adult cyclorrhaphous flies were collected from the carrions.

Using the two-ANOVA, it was observed that there were significant differences between the larvae collected from the various animals.

The number of larvae collected from the pig and chicken was significantly higher than the larvae collected from the others: fish, frog and lizard. From the calculated value and tabulated value, there was a significant difference ($4.013 > 2.76$). The carrion of pig had the highest number.

Eggs and larvae were more abundant in the natural body opening like the eyes, anus, nose and mouth. They were abundant at the underside of the animals.

Some larvae collected were bred in the laboratory and percentage emergence record (Fig 2).

TABLE 1: ADULT ARTHROPOD FAUNA VISITING CARRION.

CLASS	ORDER	FAMILY	%	SPECIES	TOTAL		
Insecta	Diptera	Calliphoridae	34.5	<i>Chrysomia</i> sp	90		
				<i>Lucilia serricata</i>	59		
				<i>Calliphora</i> sp	52		
		Sarcophagidae		<i>Sarcophaga carnaria</i>	47		
		Muscidae		<i>Musca domestica</i>	11		
		Drosophilidae		<i>Drosophila</i> sp	46		
	Coleoptera	Dermentidae		15.2	<i>Dermestes maculatus</i>	42	
		Stapylinidae			<i>Quedius fuliginosus</i>	26	
		Scarabidae			<i>Trox scaber</i>	07	
					<i>Lyginus villosus</i>	10	
					<i>Hister cadaverinus</i>	35	
		Silphidae			<i>Necrophorus</i> sp	05	
		Cleridae			<i>Necrobia rufipes</i>	20	
		Hymenoptera		Formicidae	29.5	<i>Pheidole</i> sp	246
						<i>Formica</i> sp	95
				Vespoidae	0.5		05
		Orthoptera		Gryloblattidae	1.5		15
		Dictyoptera		Blattidae	0.5	<i>Blatta</i> sp	05
Arachnida	Acaridae	17.8	<i>Acarus siro</i>	130			
				Araneae	Lycosidae	42	
Chilopoda		Scutigeromorpha	0.2		02		
Diplopoda		Juliforma	0.3		03		

TABLE 2: MEAN NUMBER OF ARTHROPOD FAUNA COLLECTED VISITING THE VARIOUS CARRION

SPECIES	FISH	FROG	LIZARD	CHICKEN	PIG	TOTAL
Dipterans						
<i>Chrysomya sp</i>	9	11	15	25	30	90
<i>Lucilia serricata</i>	4	5	9	20	21	59
<i>Calliphora sp</i>	7	6	5	15	19	51
<i>Sarcophaga carnaria</i>	2	6	7	8	10	31
<i>Musca domestica</i>	5	6	10	11	15	47
<i>Fannia canicularis</i>	-	-	3	3	5	11
<i>Drosophila sp</i>	7	6	7	11	15	46
Coleopterans						
<i>Dermestes maculatus</i>	3	4	6	10	15	38
<i>Quedius fuliginosus</i>	-	-	3	11	12	26
<i>Trox scaber</i>	-	-	-	2	5	7
<i>Lygirus villosus</i>	-	-	-	3	7	10
<i>Hister sp</i>	-	-	-	17	18	15
<i>Necrobia rufipes</i>	-	-	-	5	19	24
<i>Necrophorus sp</i>	-	-	-	2	4	16
Hymenoptera						
<i>Pheidole sp</i>	25	30	32	45	60	192
<i>Formica sp</i>	15	12	13	25	30	95
Dictyoptera						
<i>Blatta sp</i>	-	-	-	3	3	6
Arachnida						
<i>Acarus siro</i>	15	16	20	34	45	130
<i>Aranae</i>	5	5	6	10	16	42

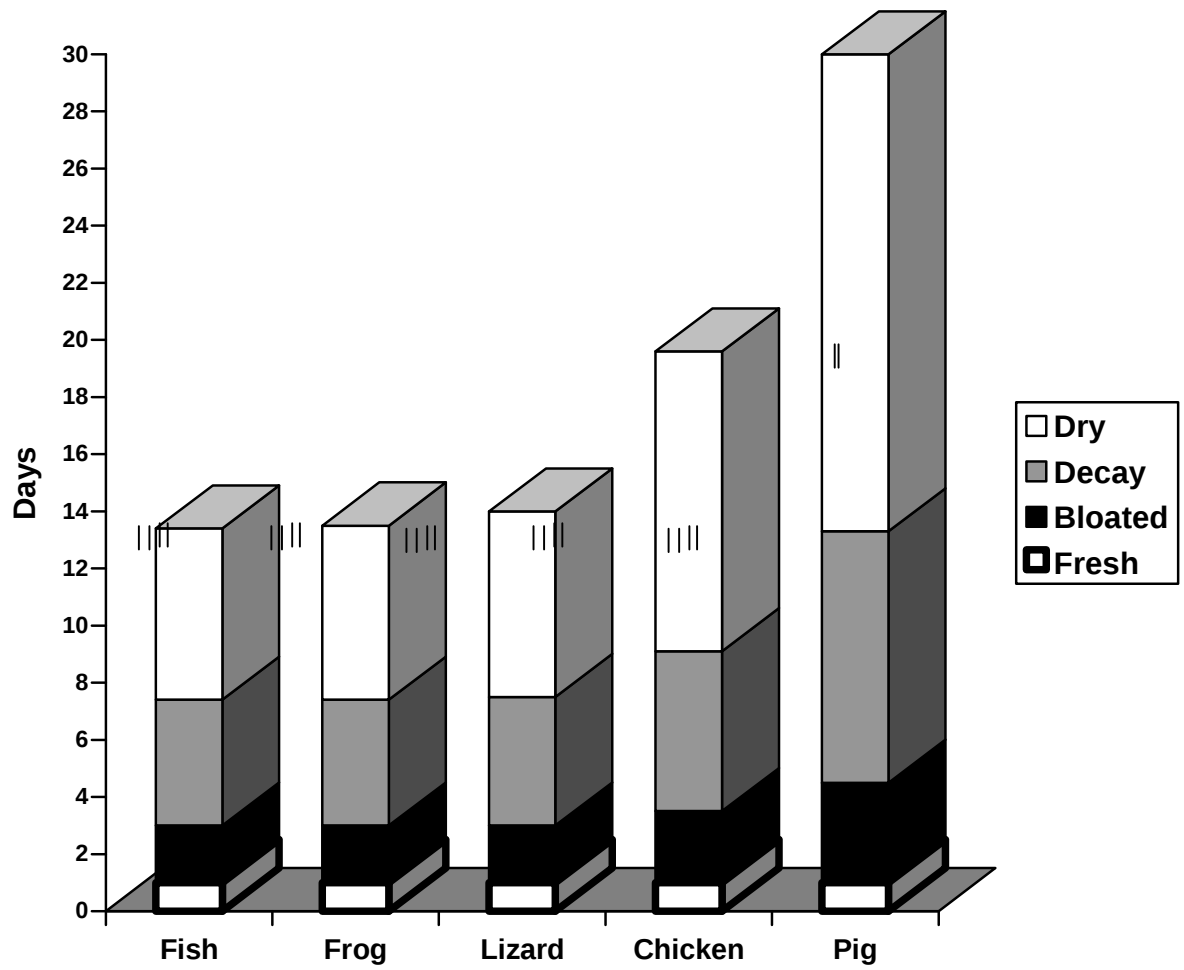


Fig 1: Duration of decomposition stages in the buried vertebrate classes.

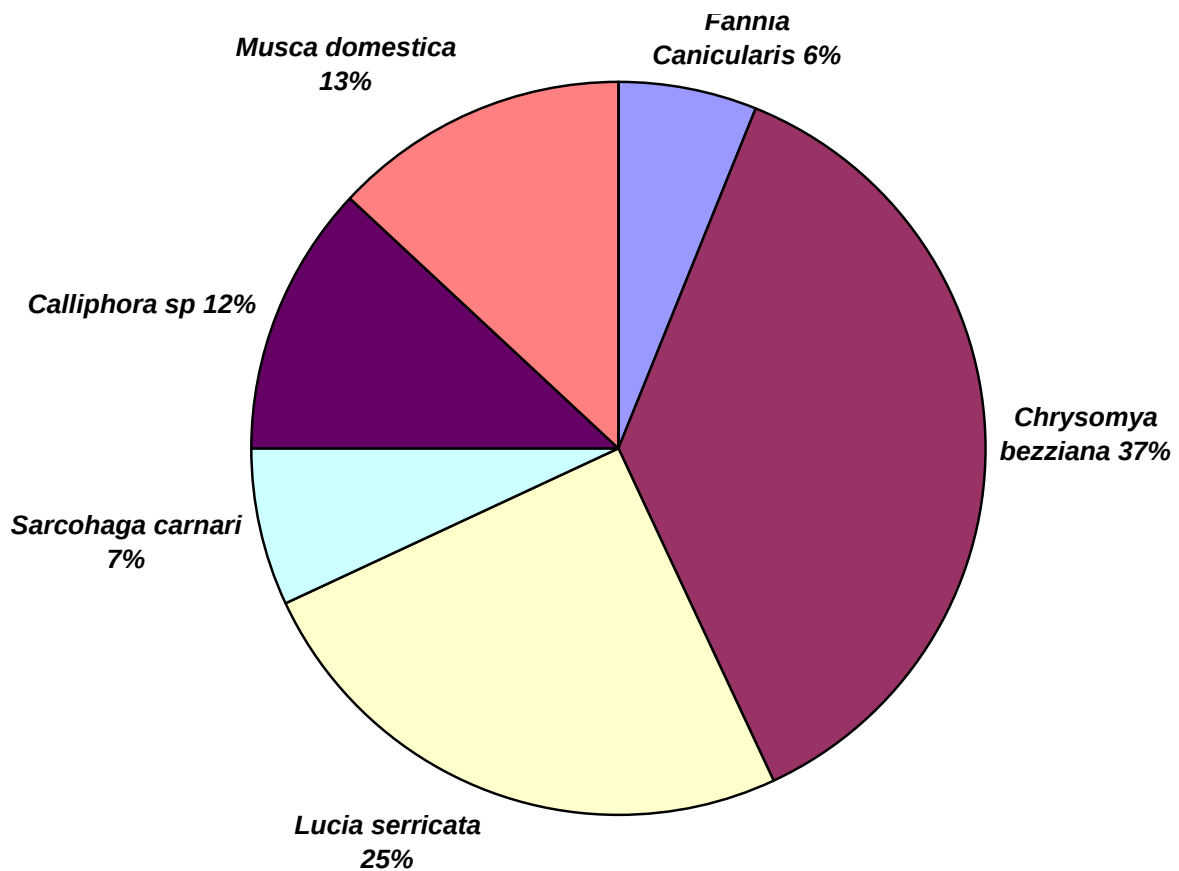


Fig 2: Pie chart showing percentage emergence of pre-adult cyclorrhaphous flies

Chrysomya bezziana has the highest 37%, *Lucilia serricata* 25% then *Musca domestica* 13% and *Calliphora sp* 12%, *Sarcophaga carnaria* and *Fannia canicularis* were the least abundant species of 7% and 6% respectively. There was significant difference between the calculated value and tabulated value ($4.0113 > 2.76$) using ANOVA.

Their abundance implies that *Chrysomya bezziana*, *Lucilia serricata* primarily breed on carrion while the least abundant *Musca domestica* and *Fannia canicularis* would breed on carrion as an alternative.

Calliphorid flies were the most abundant and they were present from day 1 attaining their peak during the decay stages of the various carrions. Their number declined at the onset of the dry stages of decomposition of the cyclorrhaphous flies visiting the carrions. *C. bezziana* was the most abundant. *L. serricata* was the next in abundance also attaining its peak during the decay stage. *Calliphora sp* was next followed by *M. domestica* and *S. carnaria* which like the others attained their peak at the decay stage. The larvae were observed moving away from the carrions sluggishly and burrowing (about 2 – 5cm) into the ground to pupate.

The larvae of *Piohilidae casei* (Chesse skippers) were observed on the pigs carrion. The larvae jumped for considerable heights while samples, were being collected.

The abundance of the flies on each of the carrion progressively increased and decreased after decay stages. (Fig 3-7)

Necrobia rufipes was the first coleopteran observed and they persisted from the post decay stage and attained their peak at the dry stage. They were only present on the pig and chicken. The appearance of *Necrobia rufipes* on the pig was later than in the chicken because its post decay

stage was also late. *Dermestes maculatus* was the next coleopteran appearing at the active decay of the carrion was found to be the most abundant beetle. They were present on all the carrion. *N. rufipes* and *D. maculatus* were seen hiding in crevices of the bones. *Quesdius fuliginosis* was the highest in number attaining its peak from the post decay stages into the dry stage. They persisted from the 3rd day to 9th day. They carried along with them mites that were attached to their appendages and the ventral side. They were presented on the lizard, Chicken and pig. *N. rufipes*, *Trox scaber* (Scarabidae) and *Hister sp* (Histeridae) were present on only the chicken and pig. However, *Necrophorus sp* was only present on the pig making it the least abundant coleopteran.

Pheidole megacephala and *Formica sp* were the only ant species present. These were most numerous. They were present from Day 1 arriving only hours after the animals were killed. These ants came in very large numbers predated and also feeding on body fluid of the carrions till the dry stage. They were seen carrying eggs, larvae and some parts of the decaying animals.

Of the arachnids present, the mites (*Acarus siro*) were the most abundant (130) while the spiders (Aranae) were also present (42). Ticks present on the pig were observed leaving the pig as the bloated stage approached to find a suitable host.

Scarabids present (*Trox scaber* and *Lygirus villosus*) were observed only on the pig and they were low in abundance. They were the last coleoptern observed.

Some incidentals were also observed; Vespaids, Chilopods, Diplopods, Blattids and Orthopterans.

The flies were the most abundant species (34.5%). Next was the formicids (29.5%) and after these were the coleopterans (15.2%). The absence, presence and abundance of arthropod species largely depended on the abundance of their food source and carrion as their breeding sites.

Discussion

The succession of arthropod fauna observed on the carrions followed the same pattern across the vertebrate classes. The earliest visitors to the carrion were the calliphorid flies. This made them the primary colonizers of the carrions. They arrived on all the carrions but their presence was first noted on the fish. This could be because the decay action first started on the fish and blowflies readily pick up faint traces of the odour of decay. They can fly up to 20km from the birth place in search of suitable corpse to lay eggs. The secondary colonizers were the ants (formicidae) and flesh flies (Sarcophagidae). Right after these were the muscids and coleopterans, making them the tertiary colonizers.

The high abundance of the flies especially *Chrysomya bezzina* implied that they breed on this carrion. The highest number of pre-adult cyclorrhaphous flies was recorded on the pig. This is due to their bulky mass and hence more body fluid. According to Hall (2003) and Zumpt (1965) synanthropic flies breed on regions with more fluid. Next to the pig was the chicken and then the lizard. The abundance of the flies also depended on the stage of decomposition. The decay stage which was averaged between the 4th to 5th day was marked by increased species richness of arthropod. However, the coleopterans arrived mainly during the decay stage and peaks in richness during the dry stage. Flies reduced as the carrions dried out.

The abundance of eggs and larva at natural body opening was because blow flies select natural body opening for oviposition. They however, would preferentially oviposit on any wound present. Ants (*Pheidole sp* and *Formica sp*) were present from the first day till exhaustion of resources. According to Tantawei *et al.* (1996), ants feed actively during all the stages of decay. Desmestid, staphylinid and scarabid beetle larvae were observed on the carrions. This implies that these species breed on carrion. However, larvae of *Necrobia rufipes* (Cleridae) were not recovered. This observation is supported by Blackburn's (1990). *Dermestes maculatus* (hide beetles) was observed and recovered from all the carrions irrespective of the class. Their larvae were also observed on the bones of the animals. They feed on dry animals of high protein such as carrions, insect remains. This explains why they were observed across the classes. The highest abundance was on the pig (which was the richest in protein) and the larvae were observed in crevices of exposed bone.

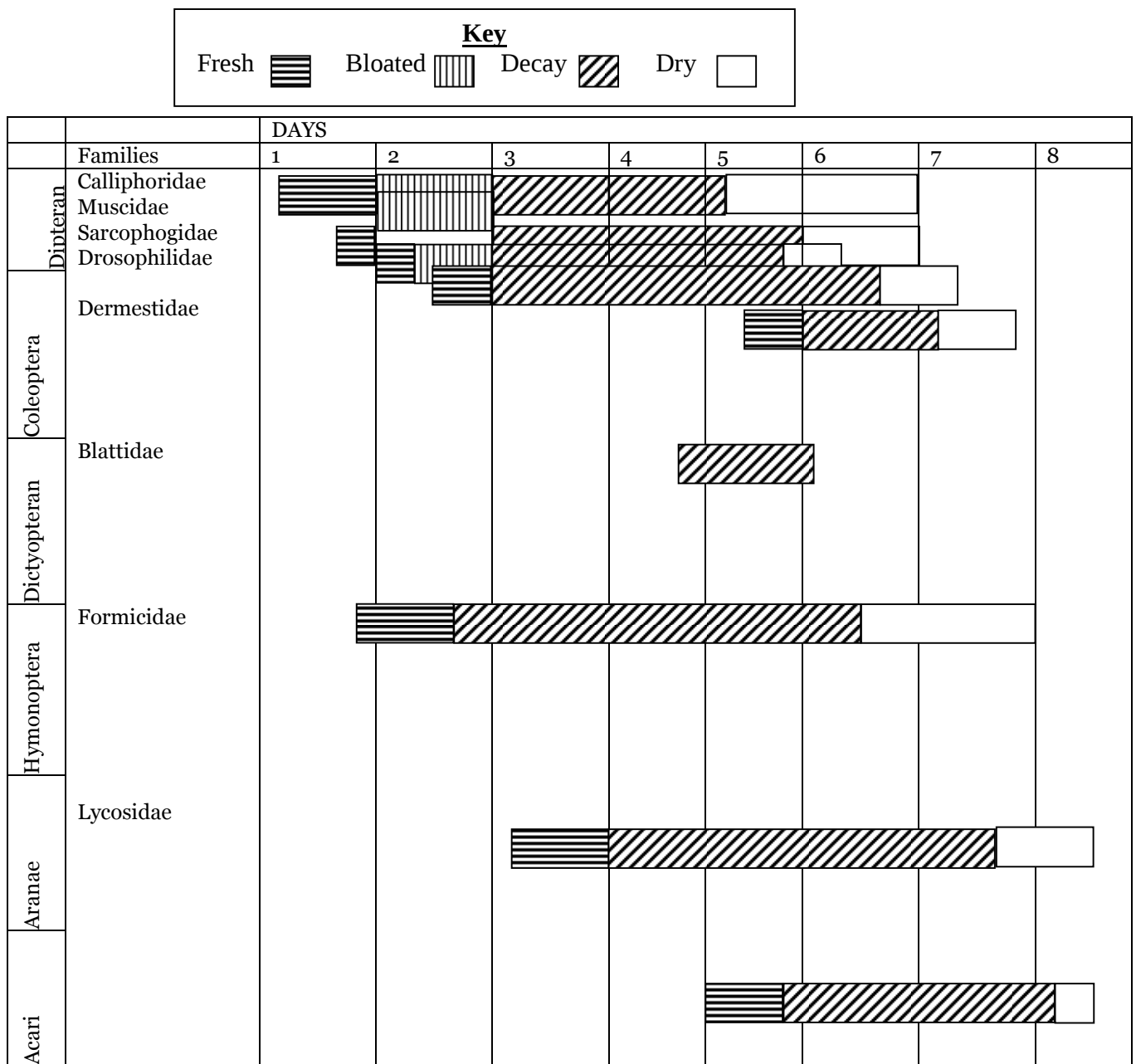


Fig 3: ARTHROPOD SUCCESSION ON EXPOSED FISH (*Pharachana obscura*) CARRION.

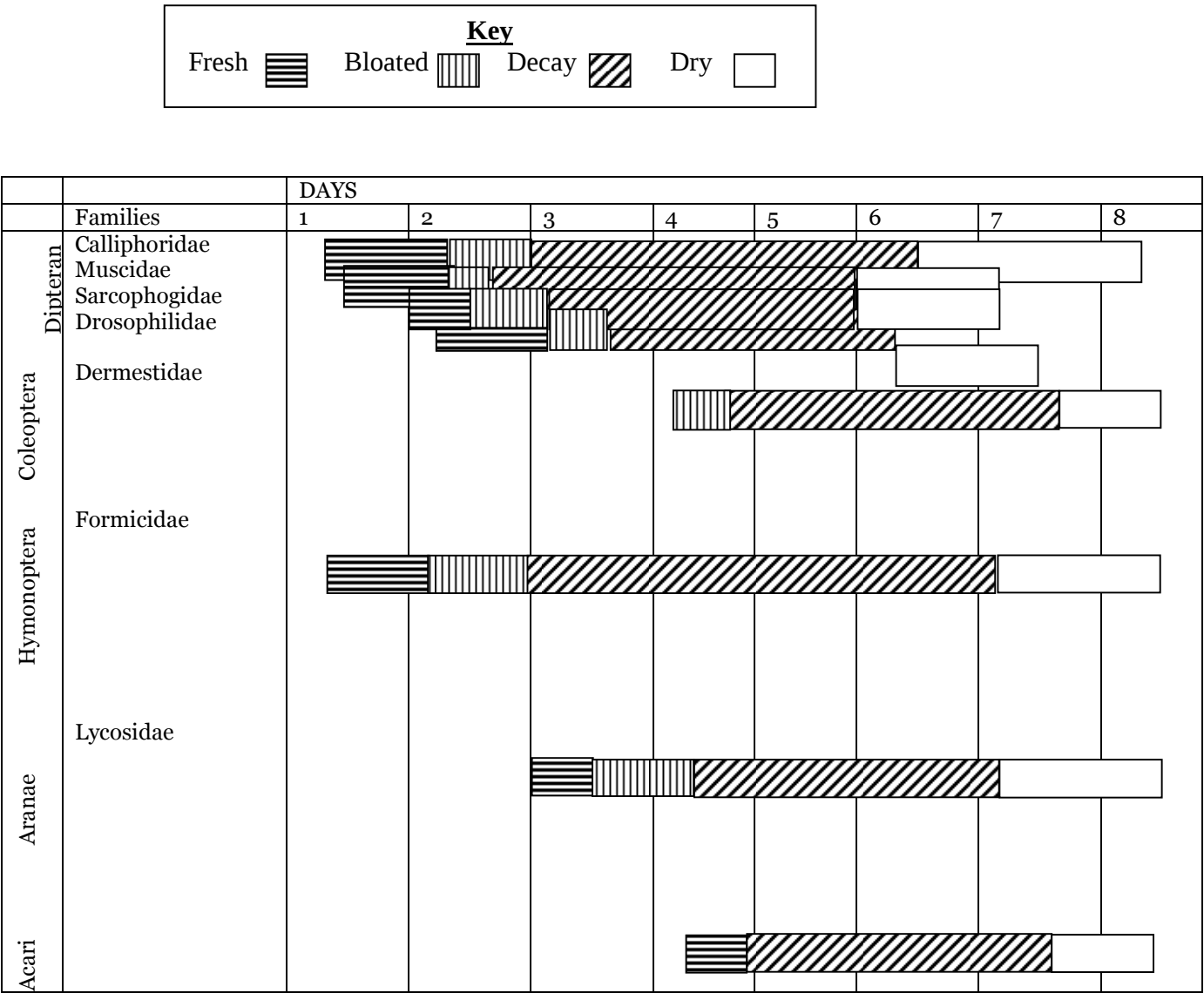


FIG 4: ARTHROPOD SUCCESSION ON EXPOSED FROG (*Bufo tempotalis*) CARRION

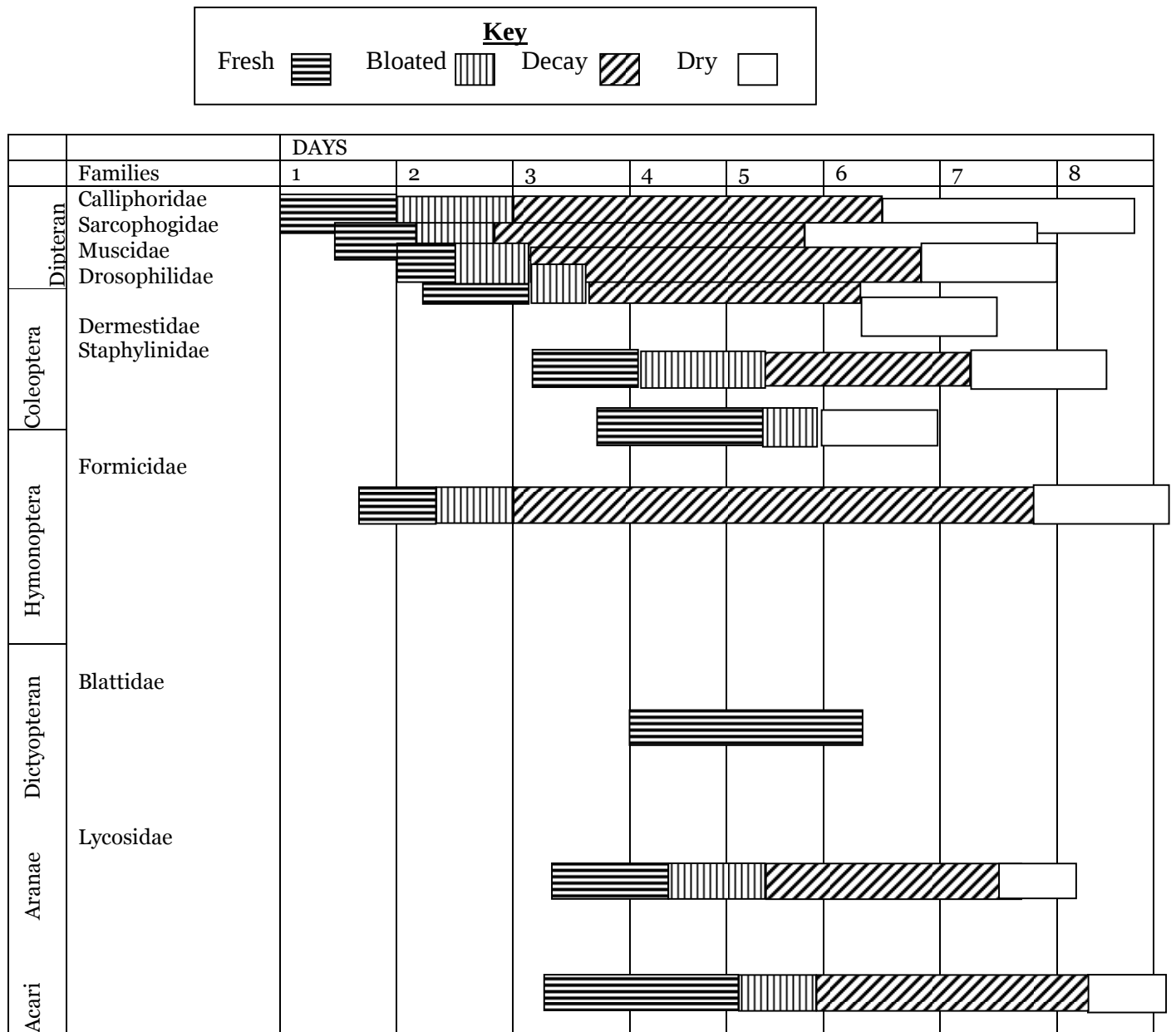


FIG 5: ARTHROPOD SUCCESSION ON EXPOSED LIZARD (*Agama agama*) CARRION

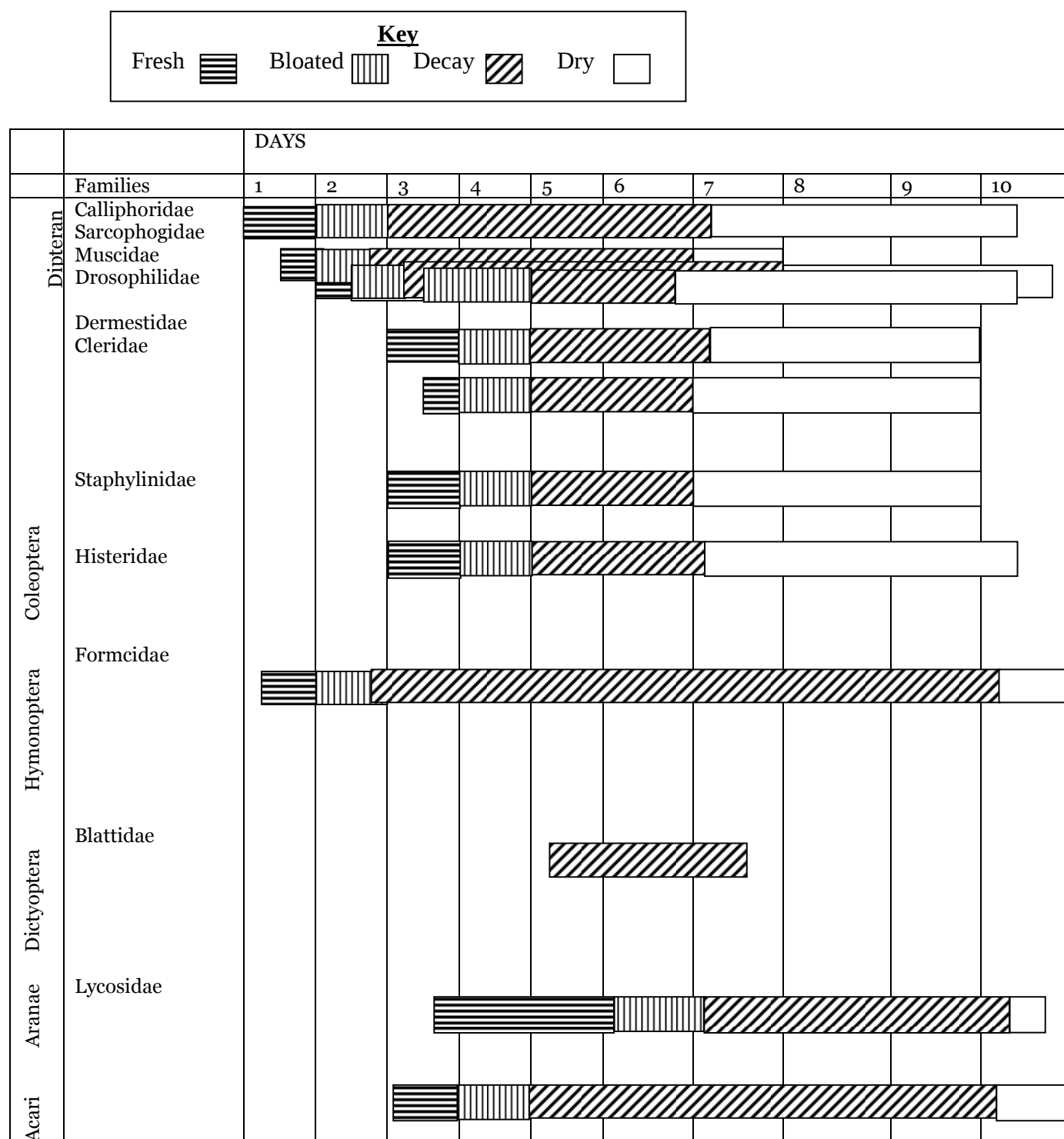


FIG 6: ARTHROPOD SUCCESSION ON EXPOSED CHICKEN (*Gallus gallus domestica*) CARRION

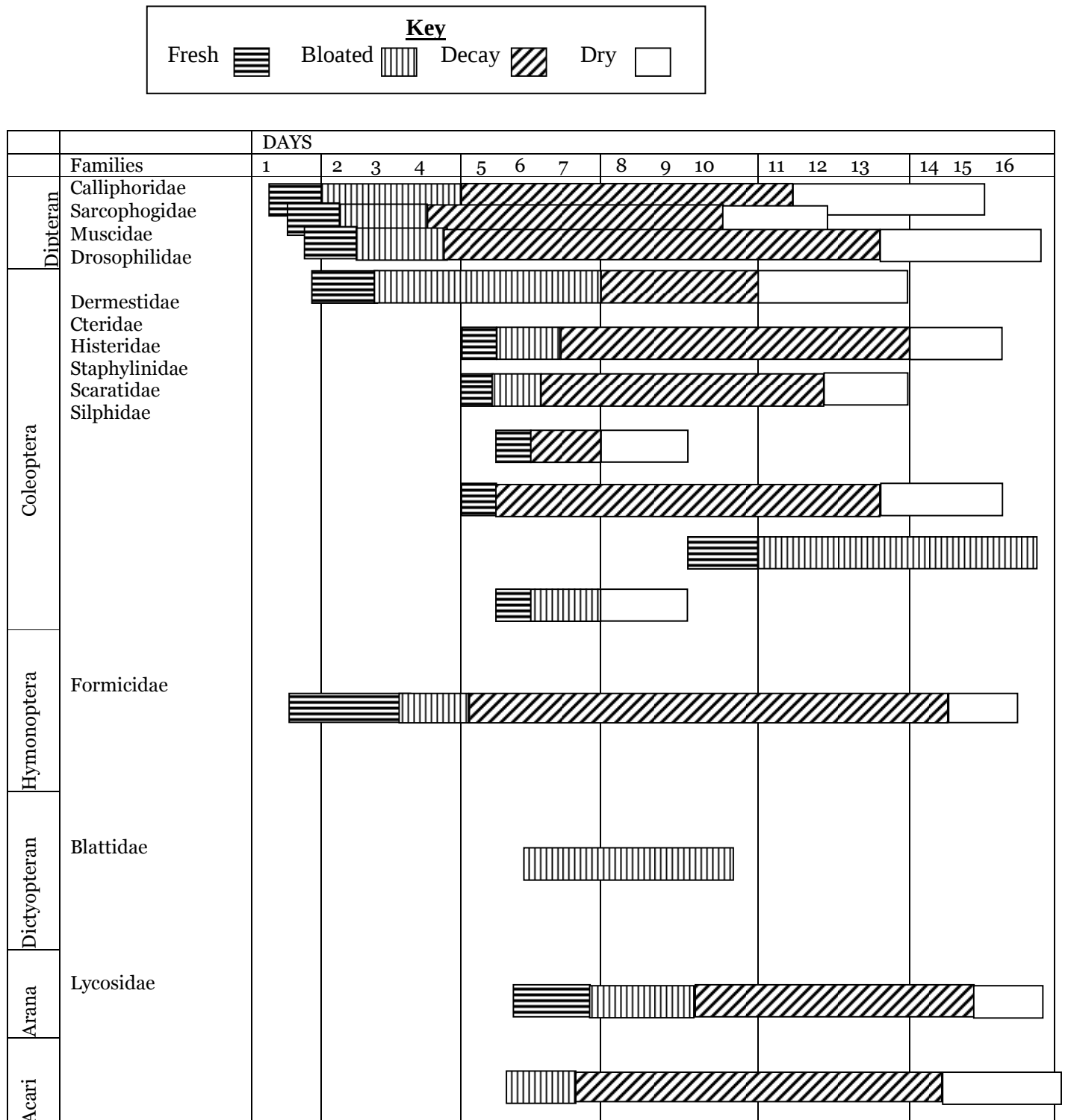


FIG 7: ARTHROPOD SUCCESSION ON EXPOSED PIG (*Sus scrofa*) CARRION

They are commonly used in museums to clean bone of their associated tissues. *Necrobia rufipes* (Red legged ham beetle) was the first coleopteran observed and was only present on the chicken and pig. Braack (1987) however believes that *N. rufipes* is not only necrovorous, but also predaceous on pre-adult of cyclorrhaphous flies. *Necrophorus* sp (Sexton beetles) was only observed on the pig and very few in number. It was observed that the first individuals of each sex to arrive at a carcass appear to fight later arrivals and turn them away. This however explains why they were found singly and least in abundance.

The presence of *Trox scaber* in the later stage and observed as the last coleopteran to invade the carcass is confirmed by Haaf (1954). Their appearance during the dry stage means they feed on dry animals carcass.

Mites present (*Acarus siro*) were seen at the underside of *Trox scabar* and attached to appendages of Rover beetles (*Quedius fuliginosus*). These mites actually live as commensals with these beetles. Mites were also present under the carrion across the classes.

Aranae (spiders) were predators of insects occurring on the carrions. No species was specific to the carrions as their numbers increased with increase of their preys and vice versa. They however have little or no importance in estimating the PMI.

The occurrence of the dipteran flies on all the carrions across the vertebrate classes denotes that these flies do not require any specific animals to breed on. However, their abundance depended on the masses and body fluid of the carrion.

Presence of coleopterans virtually depended on the abundance of the cyclorrhaphous pre-adult flies as they predated on them. The presence and abundance of some coleopteran larvae of *D. maculatus*, was because they breed on these carrions and some like *N. rufipes* only feed on them.

One important biological phenomenon that occurred on the carrion was the succession of arthropods that thrive on the different parts. For example, the beetle that specialize on bone will have to wait until bone is exposed. Predatory beetle had to wait until the blowflies arrived and lay their eggs.

In conclusion the theory behind estimating time of death or the post mortem interval (PMI) with the help of arthropod is very simple; arthropods arrive at the body soon after death and the knowledge of the biology and geographical distribution can help determine the time of death and whether the fauna collected is indigenous or foreign to the site where the body was found.

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