Study of Fly Species Composition in Human Decomposed Bodies A Prospective Study for 2 Years

Shashikanth Naik C.R.*, Lohith Kumar**, S. Venkata Raghava***, Abhishek Yadav****, Kulbhushan Prasad****

Abstract

Dead bodies are attracted by numerous species of arthropods, primarily flies, beetles, mites, isopods opiliones, nematodes and their larvae. These arthropods feed, live and breed in and on the dead bodies, depending on their biological preference and on the state of decomposition. At the very initial stages of the decay, just a few moments after the death, no odour is perceptible by a human being, but arthropods do smell it and locate their target in minutes following the death, in optimal environmental conditions of humidity, temperature, sun exposure, and corpse localization. Here in this study on 51 decomposed cases, we have collected eggs, larvae, pupae and reared till flies and these flies were identified and studied.

Keywords: Arthropods; Decomposition; Larvae; Pupae.

Introduction

Insects have existed on the earth for more than 250 million years; comparatively humans have existed for about 300,000 years. Such an enormous amount of time has allowed insects to attain wide diversity in both form and development. There are currently about 700,000 described species and it is estimated that there may be more than 10 million species of insects yet to be described [1].

Forensic entomology is the branch of forensic science in which information about insects is used to draw conclusions when investigating legal cases relating to both humans and wildlife, although on occasion the term may be expanded to include other arthropods. Insects can be used in the investigation of a crime scene both on land and in water [2].

Forensic entomology is not a new science. The first

Reprints Requests: Abhishek Yadav, Assistant Professor, Dept. of Forensic Medicine, AIIMS, New Delhi-110029. E-mail: drayad_in@yahoo.com recorded case of forensic entomology was in China in 1235AD [8]. Chinese lawyer and death investigator Song Ci in the medico-legal text book (Xiyuan jilu; one possible translation: "Collected writings on the washing away of wrongs") describes the case of a stabbing near a rice field. The day after the murder, the investigator told all workers to lay down their working tools (i.e., sickles) on the floor. Invisible traces of blood drew blow flies to a single sickle. So he confronted, the tool's owner confessed to his crime and "knocked his head on the floor". It took nearly eight hundred years until the next Chinese book on forensic entomology was published [3].

In last 20 years, forensic entomology has become more and more common in police investigations. In 1996, the American Board of Forensic Entomology, a certification Board for Forensic Entomologists was developed, similar to the Board for Certification available for Forensic Odontologists and Forensic Anthropologists [4].

Insect specimens, such as blow fly larvae (maggots) or adults, must be considered as physical evidence just as blood stains, fingerprints, hairs, fibres, or any other biological material [5].

Objectives of the Study

1. To identify the various species of insects infesting the dead body and to determine their life cycle

Authors Affiliation: *Assistant Professor, Dept of Forensic Medicine, St John's Medical College. **Assistant Professor, Dept of Forensic Medicine, Shivamogga institute of Medical Sciences. ***Associate Professor, Dept of Forensic Medicine, Bangalore Medical College & Research Institute. ****Assistant Professor, Dept of Forensic Medicine, AIIMS, Delhi.

and growth characteristics.

Material and Methods

Source of Data

Medico legal autopsies of decomposed bodies conducted at the Department of Forensic Medicine, Victoria hospital, Bangalore Medical College and Research Institute, Bangalore for a period of 24 months from Nov-2012 to Oct-2014 were examined to obtain insect samples.

Collection of Data

- 1. *Study design:* Cross sectional study
- a. Study period: Nov 2012- Oct 2014. (24 months)
- b. *Sample size:* 51 Medico legal autopsies of decomposed bodies

Inclusion Criteria

- a. All decomposed bodies with clear entomological evidence.
- b. Exhumed bodies with clear entomological evidence

Exclusion Criteria

- a. Bodies with early decomposition without entomological evidence.
- b. Body stored in cold storage after arrival to mortuary.
- c. Body preserved by other means during transportation.

Methodology

The decomposed bodies subjected to autopsy as per the request by the police department were examined for the clear entomological evidence. Only those bodies with the presence of any stage or stages of insects were utilized for the study. Background history of the decomposed body was filled in the proforma and the respective post mortem number was assigned for the individual cases. Samples of eggs and larvae were collected from different areas of the body and clothings. Fifty per cent of the samples collected from each body were preserved and remaining 50% of the sample was used for rearing. Beef liver served as food. The pupae were taken to the Department of Entomology, University of Agricultural Sciences, Bangalore along with the preserved samples in each case. Pupae were reared till adult emergence in the Entomology department.

For the identification of Calliphorid genera and species, the work of Senior White (1940) was used and the species were also got identified by Dr Meenakshi Bharti. The species of Sarcophagidae were identified using the work of Nandi (2002) and those of Muscidae by the work of van Embden (1965). All the identification was confirmed by Dr C. A. Viraktamath.

Photographs were taken using Leica M205 C microscope. Multiple images were taken at different depths and were combined using Combine ZM software.

Results

Out of 51 cases, 82.3% (42/51) cases were male and 17.7% (9/51) were female. Majority of the cases were in the age group of 31 to 50 yrs i.e. 56.8% (29/51) cases.

Out of 51 cases of decomposed bodies, *C. rufifacies* infested maximum number of cases i.e. 45% (23/51), followed by *C. megacephala* in 27.5% (14/51) cases, while mixed infestation was seen in 13.7% (7/51) cases, 11.7% (6/51) cases were infested by Sarcophagidae (*P. ruficornis*) and only one case (1.9%) was by *C. nigripes*.

In 7 cases of mixed infestation of flies 57.14% (4/7) had *C. rufifacies* and *C. megacephala* infestation, 14.28% (1/7) cases had *C. megacephala* and *P. ruficornis* infestation, 14.28% (1/7) cases had *C. rufifacies* and Muscidae infestation and 14.28% (1/7) cases had three species of flies infesting namely *C. rufifacies*, *C. megacephala* and *P. ruficornis*.

Among all the cases *C. rufifacies* infested 56.8% (29/51) cases, *C. megacephala* 39.2% (20/51) cases followed by Sarcophagidae (*P. ruficornis*), 15.6% (8/51) cases and 1.9% (1/51) cases each by *C. nigripes* and Muscidae.

Both *C. rufifacies* and *C.megacephala* infested more in bodies found outdoor i.e. 55.2% (16/29) and 60% (12/20) cases, respectively, followed by 44.8% (13/29) and 40% (8/20) cases found indoor, respectively. Sarcophagidae (*P. ruficornis*) dominated infesting 87.5% (7/8) indoor cases.

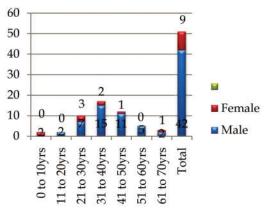
C. nigripes and Muscidae were found only in indoor cases.

As most of the bodies were in bloated stage,

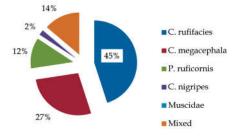
majority of the *C. rufifacies* species i.e. 57.1% (12/21) were in 3^{rd} instar larvae at the time of collection and majority of the *C. megacephala* species i.e. 44.4% (8/18) were in 2^{rd} instar larvae at the time of collection.

C. rufifacies is the dominating species and it was found infesting both male and female bodies 82.7% (24/29) cases and 17.2% (5/29) cases, respectively, followed by *C. megacephala* 85% (17/20) cases and 15% (3/20) cases, and Sarcophagidae (*P. ruficornis*) infesting 87.5% (7/8) and 12.5% (1/8) cases, respectively.

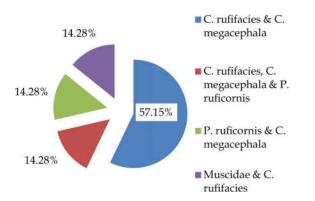
C. nigripes and Muscidae infestation were observed only in 1 male body each, of the total cases.



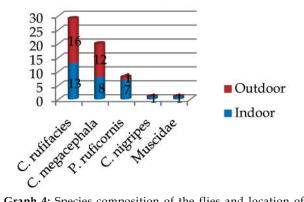
Graph 1: Age and sex - wise distribution of the cases



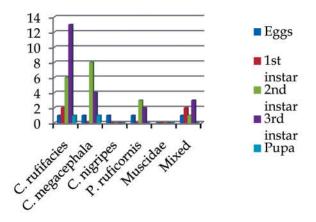
Graph 2: Species composition of flies infesting bodies



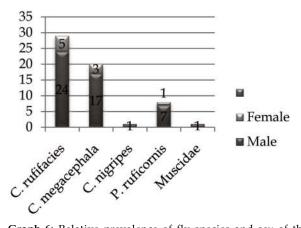
Graph 3: Species composition of flies in mixed infestation of bodies



Graph 4: Species composition of the flies and location of body



Graph 5: Distribution of fly species based on their stages of



Graph 6: Relative prevalence of fly species and sex of the development study subjects

Discussion

Morphology and and preparation of identification key

The larvae and adults of each species of flies identified were carefully observed under stereobionocular microscope and its morphological features were examined and compared to develop an identification key for species found in this study. Both larval and adult stages could be differentiated by the following set of characters.

Larvae

- a. Mandibular hooks
- b. Processes of the body
- c. Caudal spiracles
- d. Body colouration

Adults

- a. Total body length
- b. Body colour
- c. Colour of prothoracic spiracle
- d. Stripes on the mesothorax
- e. Colour of the male genital capsule
- f. Distance between compound eyes in male
- g. Relative size of the facets of compound eyes in upper and lower region of compound eyes

Based on these characters a dichotomous key for the identification of the families and species of the flies reared from human cadavers were prepared.

Identification Features of Larvae and Adult Flies

Larvae

- a. Mandibular hooks: Only one hook was found in Muscidae larvae compared to two mouth hooks in both the larvae of Calliphoridae and Sarcophagidae (Photo 4A and 4B).
- b. Processes of the body: Larvae of Muscidae, Sarcophagidae and *Chrysomya nigripes* and *Chrysomya megacephala* were smooth without body processes. Larvae of *Chrysomya rufifacies* were having series of short processes on the body (Photo 1, 2, 3).
- c. Caudal spiracles: Larvae of Calliphoridae (all the three species) had caudal spiracles in deep cavity whereas the caudal spiracles of Muscidae and Sarcophagidae were completely exposed (Photo 5 and 6).
- d. Body colouration: The larvae of *Parasarcophaga ruficornis* had dorsal sclerites of the body dark brown. In the remaining species of both Calliphoridae and Muscidae larvae were creamy white (Photo 1, 2, 3)

Adult flies

a. Total body length: Length of adult flies is given for each species.

Calliphoridae- Chrysomya megacephala 8-10 mm

Chrysomya rufifacies 7 mm

Chrysomya nigripes 5-6 mm

Sarcophagidae- Parasarcophaga ruficornis 11-14 mm

Muscidae – 7 mm.

- b. Body colour: All the three species of Calliphoridae were metallic blue or green in colour. Species of Sarcophagidae and Muscidae were non metallic grey or black with stripes on mesothorax (Photo 9)
- c. Colour of prothoracic spiracle: Both *Chrysomya nigripes* and *Chrysomya rufifacies* have white prothoracic spiracle (Photo 8A, 8C and 10A, 10C), *Chrysomya megacephala*, Muscidae and Sarcophagidae have brown prothoracic spiracles (Photo 7A and 11A).
- d. Dorsal stripes on mesothorax: Species of Calliphoridae have black transverse stripes and short longitudinal stripes only in the anterior half. *Parasarcophaga* (Sarcophagidae) have three longitudinal stripes extending entire length of mesothorax. Muscidae has four longitudinal stripes extending entire length of mesothorax (Photo 9A and 9B).
- e. Colour of male genital capsule: It is black in colour in the three species of Calliphoridae and one species of Muscidae. It is reddish brown in *Parasarcophaga* (Sarcophagidae) (Photo 11A).
- f. Distance between compound eyes in male flies: In case of Muscidae, Sarcophagidae and *Chrysomya nigripes* (Calliphoridae) compound eyes are well separated (Photo 10B and 11B). In case of *Chrysomya rufifacies* and *Chrysomya megacephala* eyes are close together (Photo 7B and 8B).
- g. Relative size of facets of upper and lower portion of compound eyes in male flies: The facets of compound eyes are more or less of same size or gradually reduced in size downwards in Muscidae, Sarcophagidae, *Chrysomya rufifacies* and *Chrysomya nigripes*. In case of *Chrysomya megacephala* facets in the upper 2/3rd of the portion were larger compared to the lower 1/3rd portion and the transition is abrupt (Photo 7B).

Key to larvae found in human cadaver

1.	With one mouth hook (Photo 4B)	Muscidae
-	With two mouth hooks (Photo 4A)	
2.	Larvae with caudal spiracles exposed (Photo 5)	Sarcophagidae
-	Larva with caudal spiracles concealed in deep cavity (Photo 6) 	
3.	Larva with short projecting processes on body (Photo 2B and 2C)	
	Chrysomya	<i>rufifacies</i> (Macquart)
-	Larva smooth without projecting processes on body (Photo 1)	

Key to adult flies reared from human cadaver

1.	Flies metallic blue or green (Photo 7A & 7B; Photo 8A & 8C)	
-	Flies non metallic (Photo 9A & 9B) 4	
2.	Flies not more than 6 mm in length, eyes widely separated both in male and female (Photo 10) <i>Chrysomya nigripes</i> Aubertin	
-	Flies more than 6 mm in length, eyes close together in male, widely separated in female (Photo 7 & 8)	
3.	Compound eyes of males with large facets in upper two thirds and smaller ones in lower one third (Photo 7B), prothoracic spiracle brown in both sexes (Photo 7A & 7C)	
-	Compound eyes of both sexes with uniformly sized facets of eye (Photo 8B & 8D): prothoracic spiracle white in both sexes (Photo 8A & 8C).	
	Chrysomya rufifacies (Macquart)	
4.	Thorax with three black stripes (Photo 9A); large flies measuring more than 11 mm; male with genital capsule reddish brown (Photo 11A)	
	Sarcophagidae	
-	Thorax with four black stripes (Photo 9B); smaller flies not measuring not more than 8 mm; male with genital capsule dark brownMuscidae	



Photo 1: Larvae of *Chrysomya megacephala* (Fabricius). 1st Instar (A), 2nd Instar (B), 3rd Instar (C) larvae, lateral view.



Photo 2: Larvae of *Chrysomya rufifacies* (Macquart). 1st Instar (A), 2nd Instar (B), 3rd Instar (C) larvae, lateral view Please note the projecting processes (B and C)

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Photo 3: Larvae ot *Parasarcophaga ruficornis* (Fabricius). 1st Instar (A), 2nd Instar (B), 3rd Instar (C) larvae, lateral view. Note dark brown dorsal abdominal sclerites (C).

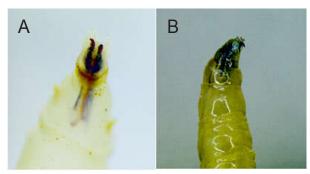


Photo 4: Larvae of Sarcophagidae and Muscidae. A - Sarcophagid larvae, showing two mandibular hooks; B - Muscid larvae, showing one mandibular hook



Photo 5: Caudal spiracles of *Parasarcophaga ruficornis* (Fabricius) in shallow depression.



Photo 6: Caudal spiracles of *Chrysomya megacephala* (Fabricius) in deep cavity

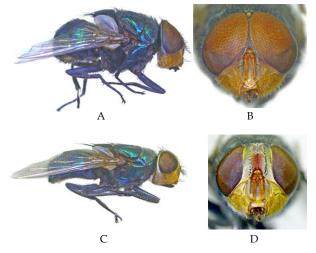


Photo 7: Adult flies of *Chrysomya megacephala* (Fabricius). A - Male, lateral view; B - Male face, front view showing eyes close together and large sized facets on upper region and smaller sized facets on lower region; C – Female, lateral view; D - Female face, front view showing widely separated eyes with uniformly sized facets.

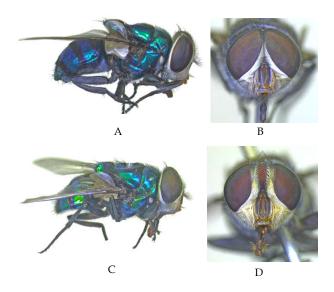


Photo 8: Adult flies of *Chrysomya rufifacies* (Macquart). A - Male, lateral view; B – Male face, front view showing eyes close together with similar sized facets; C – Female, lateral view; D – Female face, front view showing widely separated eyes.



Photo 9: Sarcophagid and Muscid flies. A – Sarcophagidae, Parasarcophaga ruficornis (Fabricius),

A – Sarcophagidae, *Parasarcophaga ruficornis* (Fabricius), dorsal view, showing three black stripes on mesothorax; B – Muscidae, dorsal view, showing four black stripes on mesothorax

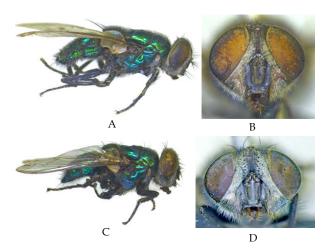
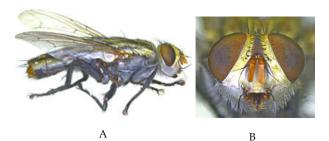


Photo 10: Adult flies of *Chrysomya nigripes* Aubertin. A - Male, lateral view; B - Male face, front view showing eyes much closer to each other compared to female (see D); C -Female, lateral view; D - Female face, front view.



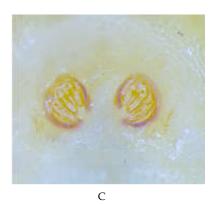


Photo 11: Adult fly and larval caudal spiracles of *Parasarcophaga ruficornis* (Fabricius).

A - Male, lateral view; B - Face, front view; C - Caudal spiracles of larva

Conclusion

Among the five species of flies recorded on human cadaver in this study, *Chrysomya megacephala* (Fabricus) and *Chrysomya rufifacies* (Macquart) are the most common and dominant species infesting throughout the year. *Chrysomya nigripes* Aubertin and species of Muscidae were the least common.

Parasarcophaga ruficornis (Fabricius) was prevalent mostly indoors.

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