

CASE REPORT

José R. Pujol-Luz,¹ Ph.D.; Helder Marques,² M.Sc.; Alexandre Ururahy-Rodrigues,³ M.Sc.; José Albertino Rafael,³ Ph.D.; Fernando H.A. Santana,¹ B.Sc.; Luciano C. Arantes,⁴ M.Sc.; and Reginaldo Constantino,¹ Ph.D.

A Forensic Entomology Case from the Amazon Rain Forest of Brazil

ABSTRACT: The first case of application of forensic entomology in the Brazilian Amazonia is described. The corpses of 26 men were found in the rainforest in Rondonia State, Brazil. Fly larvae collected on the bodies during autopsy were identified as *Paralucilia fulvinota* (Diptera, Calliphoridae). No data or specimens were collected at the crime scene. At the laboratory, the larvae developed into pupae in 58 h and into adults in 110.5 h. The total development time for *P. fulvinota* was measured in field experiments inside the forest. The age of the larvae when collected from the bodies was estimated as the difference between the time required for them to become adults and the total development time for this species. The estimated age of the maggots and the minimum postmortem interval was 5.7 days.

KEYWORDS: forensic science, forensic entomology, postmortem interval, Amazon rainforest, *Paralucilia fulvinota*

A cadaver constitutes a dynamic system that supports a rich community of arthropods, which is affected by several local factors. The development rate of necrophagous arthropods can be a valuable tool to estimate the postmortem interval (PMI) (1). However, this estimation depends on information about the composition and dynamics of the local communities of necrophagous arthropods.

Amazonia is the world's largest and most diverse tropical forest region, covering more than 6 million km² (2). The highly diverse and poorly studied insect fauna, combined with the constant high temperature and humidity, the great distances, and poor local infrastructure, creates special difficulties in the application of forensic entomology in this region.

In this paper, we describe the first case of estimation of PMI based on necrophagous insects in the Brazilian Amazonia.

Case Description

On 19 April 2004, the corpses of 26 men were found in the forest in an Indian reservation (Parque Indígena Aripuanã), in Rondonia State, Brazil (Fig. 1). This is a remote region within

¹Departamento de Zoologia, Universidade de Brasília, 70910-900 Brasília, DF, Brazil.

²Setor Técnico-Científico do Departamento de Polícia Federal de Rondônia, Av. Lauro Sodré 2905, 78900-050 Porto Velho, RO, Brazil.

³Instituto Nacional de Pesquisas da Amazônia, Caixa Postal 478, 69011-970 Manaus, AM, Brazil.

⁴Instituto de Criminalística, SAISO Complexo da Polícia Civil, 70610-200 Brasília, DF, Brazil.

Financial support: Brazilian Ministry of Justice and Brazilian National Research Council (CNPq 520176/03-0).

Received 23 Oct. 2005; and in revised form 4 Mar. 2006; accepted 26 Mar. 2006; published 31 Aug. 2006.

dense rainforest. These men were identified as diamond miners, who had been involved in altercations with the Cinta Larga Indians. The bodies were found in different positions, 23 of them close together and another three isolated, at a distance of 1000 m from the main group. The cadavers were dressed in T-shirts, shorts, or pants, and were in different conditions of exposure to sunlight and moisture. The decomposition stage varied from saponified to skeletonized (Fig. 2). Postmortem damages such as bites and perforations were present, presumably caused by insects and other animals. The autopsy indicated that most deaths were caused by craneo-cephalic traumatism. Arrow heads, clubs, and spears were present at the crime scene.

No forensic pathologist or entomologist was present at the crime scene, no insect was collected, and temperature and humidity data were not recorded. The corpses were removed and transported by helicopter to the city of Porto Velho, located c. 500 km from the crime scene. On 20 April 2004, in the course of the autopsy, 320 fly larvae were collected from five of these corpses. The larvae were immediately refrigerated and sent to the University of Brasília to estimate the PMI, arriving at the laboratory on 21 April 2004.

Only 57 larvae arrived alive at the laboratory. The dead ones were preserved in 70% ethanol for identification using the available literature (3). All larvae were third instar (L3) of a single fly species. The living larvae were kept in a BOD incubator at 28°C and 80% relative humidity. Among these, 41 pupated after 58 h and 10 emerged as adults in another 110:30h. The rest died of unknown causes. The total time to emergence as an adult was, therefore, of c. seven days after arrival of the larvae.

With the help of the available identification keys (4, 5), the 10 adults were identified as *Paralucilia fulvinota* (Bigot, 1877) (Diptera: Calliphoridae; Fig. 3). This identification was later confirmed by an expert taxonomist (Dr. Rubens P. de Mello). Unfortunately,

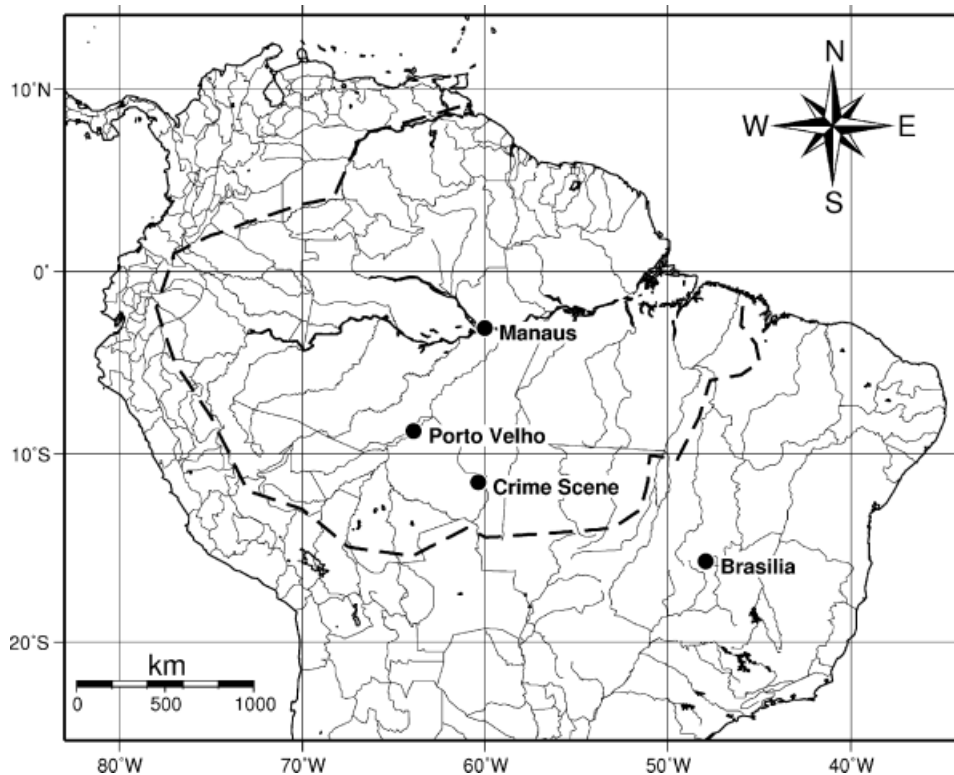


FIG. 1—Location of the crime scene and other localities mentioned in this paper. The dashed line indicates the approximate limits of the Amazon forest.

there are no published data on the development time of this species. However, the 4th author (J. A. R.) had estimates of development times of *P. fulvinota* from recent field experiments conducted in the rainforest near Manaus, Amazonas State, Brazil.

The experiments in Manaus were conducted inside the forest, using dead pigs. Development times were measured by placing eggs in Petri dishes with pork, which were covered with a fine mesh and kept in a cage in the forest. The eggs and larvae were examined periodically under a stereomicroscope. During this experiment, air temperature varied between 23.4°C and 28.8°C, with an average of 26.0°C, and the relative humidity ranged from 72% to 99%, with an average of 90%. The average development times

were as follows: oviposition to eclosion: 18:03 h; L1: 30:30 h; L2:16 h; L3: 118 h; and pupa: 122:30 h, with a total of 305 h.

The minimum PMI was estimated as the age of the larvae present on the corpses. We estimated their age at the time they were collected as the difference between the total development time (obtained from the Manaus experiment) and the time necessary for them to develop into adults. Minimum PMI = 305–168:30 h = 136:30 h = 5.7 days (Fig. 4). We considered that development stopped during the period they were refrigerated (about 1 day). Therefore, the most probable day of oviposition was April 15th.

The maximum PMI could not be estimated from entomological evidence, because *P. fulvinota* was the only species collected and, based on the experiments conducted in Manaus with pig carcasses, it colonizes corpses at all stages of decay.



FIG. 2—Original position of some of the dead bodies found in the Aripuanã Indian Reservation, Rondonia, Brazil. Large numbers of maggots are visible as white dots on and near the corpses.



FIG. 3—Adult of *Parulicilia fulvinota* obtained from the maggots collected on the corpses from Rondonia, Brazil.

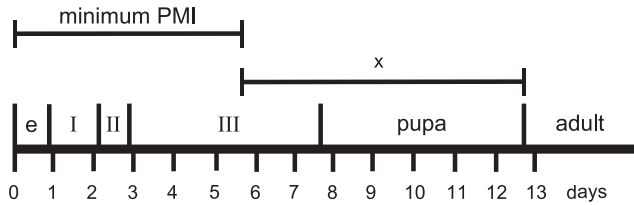


FIG. 4—Time scale showing the duration of each instar of *Paraluclia fulvina* and estimation of postmortem interval. I, 1st instar (L1); II, 2nd instar (L2); III, 3rd instar (L3); x, time of incubation at the laboratory for the larvae collected at the corpses to transform into adults.

Discussion

Estimation of the PMI is the main contribution of forensic entomology in death investigation. The minimum PMI can be determined by the age of the immature insects present at the time of corpse discovery (6). Their age can be estimated by several different methods.

The age of the maggots can be estimated by comparing their size with known growth curves (7). However, this was not a good option in this case due to several reasons: (a) the use of this method requires proper preservation of the maggots as soon as they are collected; (b) initially, we did not know how many species were present or their identification; (c) our first priority was to obtain adults for identification; and (d) we had no access to previous relevant information from Amazonia, and the laboratory where this case was studied is located in central Brazil (Fig. 1), a region with completely different fauna and climate. Without the identification, the preserved larvae would be useless.

Because temperature affects the velocity of development of the larvae, estimation of PMI is often based on cumulative degree-hours or degree-days. We did not use this method due to the absence of measurements of air and corpse temperatures at the crime scene. The Airbase at Porto Velho, Rondonia, provided meteorological records for this period, but it is located 500 km from the crime scene and the microclimate inside the forest is certainly different, with lower temperatures and higher humidity.

However, both daily and annual variation of temperature and humidity inside a tropical rain forest are very small when compared with regions with temperate climate. The daily average temperatures vary only 2–3°C along the year, and the daily variation is less than 10°C (8). The absolute minimum temperature is rarely below 20°C and never below 10°C, which is above the developmental minimum for fly larvae. In April, the absolute historical minimum reported for Rondonia (Porto Velho) was 13°C, and the average minimum was 22°C. Under these conditions, local microclimate and heat generated by the decomposition process and maggot activity are more important for larval development than air temperature.

Given all these limitations, our estimate of the minimum PMI was based solely on the development time of *P. fulvina*. The maximum PMI could not be estimated based on entomological evidence. This case illustrates the difficulties in the application of forensic entomology in Amazonia and the main differences from temperate regions of the Northern Hemisphere, where these methods have been developed: (a) very little knowledge about which species of necrophagous insects can be found in corpses and their biology; (b) great distances and poor local infrastructure; and (c) constantly high temperature and humidity, resulting in rapid decay.

Acknowledgments

The authors thank the Department of Federal Police at Rondonia for sending material and information. We are also grateful to the Brazilian Air Force at Porto Velho for sending meteorological information, to Dr. Rubens Pinto de Mello (FIOCRUZ, Rio de Janeiro) for helping with the identification of *Paraluclia fulvina*, and to Dr. Regina Macedo (UnB) for reviewing the manuscript.

References

- Megnin JP. La faune des cadavres: application de l'entomologie a la medecine legale. Encyclopedie Scientifique des Aides-Memoires. Paris: Masson et Gauthiers- Villars, 1894.
- Silva JMC, Rylands AB, Fonseca GAB. The fate of the Amazonian areas of endemism. *Conserv Biol* 2005;19(3):689–94.
- Guimarães JH, Papavero N. Myiasis in man and animals in the neotropical region—bibliographic database. São Paulo: FAPESP and Editora Plêiade, 1999.
- Dear JP. A revision of world Chrysomyini (Diptera: Calliphoridae). *Rev Brasil Zool* 1985;3(3):109–69.
- Mello RP. Chave para a identificação das formas adultas das espécies da família Calliphoridae (Diptera, Brachycera, Cyclorhapha) encontradas no Brasil. *Entomol Vec* 2003;10(2):255–68.
- Catts EP. Analyzing entomologica data. In: Catts EP, Haskell NH, editors. *Entomology and death, a procedural guide*. Clemson: Joyce's Print Shop, 1990:124–37.
- Wells JD, Lamotte LR. Estimating the postmortem interval. In: Byrd JH, Castner JL, editors. *Forensic entomology*. Boca Raton: CRC Press, 2001:261–85.
- Instituto Nacional de Meteorologia [Brazilian National Institute of Meteorology] URL http://www.inmet.gov.br/climatologia/combo_climatologia_1.html. Accessed 28 September 2005.

Additional information and reprint requests:

José R. Pujol-Luz, Ph.D.
 Depto de Zoologia
 Universidade de Brasília
 70910-900 Brasília, DF
 Brazil
 E-mail: jrpujol@unb.br