

TECHNICAL NOTE

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Successional Behavior and Occurrence Matrix of Carrion-Associated Arthropods in the Urban Area of Medellín, Colombia*

ABSTRACT: Forensic entomology is a scientific tool applied to the study of insect or arthropod succession at the scene of a crime or that associated with an accident or natural death. Interpreting this succession provides information to determine minimum and maximum limits of the Postmortem Interval (PMI), that is, the time between death and the discovery of the body. This study was carried out during the rainy season, from 27 October to 12 December 2002 in an urban area of the city of Medellín, Colombia. Three domestic pigs were used as animal models. The results showed that both the aforementioned species as well as the physical characteristics of the carcass determined the indicator species of the postmortem interval in urban areas of the city. In total, 11,937 individuals were collected and identified, belonging to 12 orders, 29 families and 42 genera. Diptera were the most abundant order (90%) represented mainly by Calliphoridae (80%). Coleoptera were scarcely present, representing only 2.8% of the total collected. Based on information obtained of eggs and larvae reared to adult of this successional study, an occurrence matrix was elaborated for determining of the PMI. From the same study area and under the same conditions, a list of arthropods associated with carrion is presented in this manuscript.

KEYWORDS: forensic science, forensic entomology, Medellín, insect succession, postmortem interval, occurrence matrix

Forensic entomology is a scientific tool applied to the study of insect or arthropod succession at the scene of a crime or that associated with an accident or natural death. Information is thus provided for medical, legal and forensic investigations (1). Interpretation of this succession is mainly used to establish the minimum and maximum range of the postmortem interval (PMI), in other words, the time between death and the discovery of the body (2–7).

From the moment of death in animals and humans, different types of insects are attracted to the body by smells caused by bacterial decomposition. This putrefaction is strongly influenced by environmental conditions such as temperature, humidity, rainfall and sunlight (4). The colonization and insect succession pattern is given by the physical and chemical changes taking place in the body (2).

The analysis of insects and other arthropods as evidence in a criminal investigation is based on the concept of ecological succession (8–11). Invasive insects feeding on an exposed carcass arrive in a series of different stages. The first stage involves the removal of soft tissues from the carcass, which in turn, makes it more attractive for the subsequent phase of insects preferring dry skin or cartilage (1).

Carrion-feeding fauna, particularly the dipteran families Calliphoridae and/or Sarcophagidae are usually the first “witnesses”

to arrive at the scene of the crime. They come to the carcass to feed and lay their eggs or larvae. As decomposition progresses, they are followed replaced by Coleoptera (11).

Studies on arthropod succession on exposed carcasses in different environments are well documented in temperate regions by Payne (11), Anderson et al. (12), Arnaldos et al. (13), Anderson (14), Miralbé (15). In the tropics Hawaii, successional studies have been carried out in Hawaii by Early & Goff (16), Tullis & Goff (17), Shalaby et al. (18), Goff & Flynn (19), and Goff and Odom (20).

In the Neotropics, research has been carried out mainly in Sao Paulo, Brazil by Carvalho et al. (21,22) and in Costa Rica by Jirón & Solano (23), Jirón et al. (24), and Jirón (25). The following families were reported in the above studies: Calliphoridae, Sarcophagidae, Muscidae and Stratiomyidae, and in Panama by Dunn (26). Specifically for Colombia, Barreto et al. (27) reported Diptera and Coleoptera specimens found on human bodies in Cali and Wolff et al. (28) carried out the first studies in forensic entomology in a metropolitan area of Medellín.

The contribution of entomological indicators in resolving unsolved crimes may be a reliable method of estimating PMI as long as the insect succession and biological characteristics of the associated species are taken into account (29). Schoenly et al (29,30) introduced the concept of successional records in an “occurrence matrix”. This system registers the day of appearance of each species so that an analysis of presence or absence of a certain group of species allows PMI to be estimated with only one or two days’ margin of error.

The objective of this study was to know the succession of carrion insects and to create an “occurrence matrix” using information

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* This work was supported by Colciencias (Instituto Colombiano para el Desarrollo de la Ciencia y la Tecnología), project 1115-05-11503 and by the Universidad de Antioquia, Colombia.

Received 8 Dec. 2004; and in revised form 18 May and 9 Oct. 2004; accepted 10 Oct. 2004; published 2 Feb. 2005.

from insects found during the decomposition of an exposed animal model (*Sus scrofa* L) during the rainy season in an urban area of Medellín, Colombia.

This study (in the bio-climatic zone, Pre-montane Humid Forest) is part of a larger project being carried out in different climate zones in Colombia.

Materials and Methods

The project was carried out in a field within the campus of the Universidad de Antioquia in Medellín, at an altitude of 1.450 msnm. Average temperatures fluctuated between 18 and 24°C, average annual rainfall amounted to 1.409 mm (31). The corresponding bio-climatic zone is bh-P according to Holdridge (32).

The experiment lasted from 27 October to 12 December 2002, corresponding to the rainy season. Three domestic pigs (*Sus scrofa* L) weighing 10 kg each were used as animal models according to methods proposed by Shean et al. (33) and Ferllini (34). The pigs were killed by cardiac puncture at 09:00 in the study area.

At each sampling time, the physical characteristics of the body were noted as well as the insect activity. In addition to body weight, internal (rectal) and environmental temperatures were also recorded. Eggs and larvae were collected from the body by hand and adults with an entomological net. Samples were taken from the soil beneath the carcass up to a depth of 3 cm. Immature insects were fixed in 80% alcohol and the adults were killed with ethyl acetate. To ensure the correct identification of the different larvae colonising the body at different instances, some of the eggs and larvae were reared to adult under laboratory conditions. This also avoided problems in the identification of 1st and 2nd larval instars. All insect material was deposited in the Entomological Collection of the University of Antioquia in Medellín. Identification was carried out using taxonomic keys (35–37). Some species of the larvae were fixed on slides with Canada balsam (38).

Results

In total, 11,937 individuals were collected belonging to 12 orders, 29 families and 42 genera (Table 1). Diptera was the most abundant order with 10,756 individuals (90%), represented principally by the following families: Calliphoridae with 9,633 individuals (89.5%), Muscidae, 624 (5.8%), Piophilidae 308 (2.9%), and Sarcophagidae 165 (1.5%). Coleoptera showed a scarce presence with only 2.8% (341) and specimens were found at the end of the period of decomposition. The order was mainly represented by the family Dermestidae with 120 individuals (1.0%).

The species of Calliphoridae with major adult abundance were: *Phaenicia sericata* ($N = 2220$), *Cochliomyia macellaria* ($N = 696$), *Chrysomya albiceps* ($N = 680$), and *Chrysomya megacephala* ($N = 575$). The most representative species of Muscidae was *Ophyra capensis* ($N = 504$) and for Sarcophagidae, *Oxysarcodexia* sp. Adults of the latter genera were present throughout the succession in small numbers ($N = 146$).

Physical changes, internal body temperature and the reduction in biomass were used to define five different stages of decomposition: fresh, bloated, active decay, advanced decay and dry remains. This coincided with studies performed by Wolff et al. (28), Early & Goff (16) y Goff (39).

The data for first occurrence of a species, and the overall numbers collected were compared in the three pigs, there were no statistical differences and the data from all carcasses were pooled (Kruskall Wallis ANOVA, $P > 0.05$).

The following succession of carrion-associated insects, used in the succession table, was found for the rainy season from October to December.

Fresh Stage

The fresh stage began at the moment of death and ended when the body began to swell (13). In this study the stage lasted 1 day (Table 1). Morphological changes and internal temperature variation were minimal during this period (Figs. 1, 2). *Phaenicia sericata* was the first species to lay eggs in the nasal orifices, 45 min after death (9:45 am). The eggs rapidly hatched (approximately 6 h later, after oviposition Tables 1, 2), probably due to high environmental temperature (36°C).

Bloated Stage (1)

Began from the second day of the study until day 6 (Tables 1, 2). Body weight and internal temperature increased slightly (Figs. 1, 2). First and second larval instars of Calliphoridae were observed. The larvae were later identified as *C. macellaria* and *C. megacephala* (Table 2).

Dorymerys sp., *Pheidole* sp., and *Solenopsis* sp. ants were preying actively on dipteran eggs and larvae. Individuals of the genus *Diplonevra* (Phoridae) were attracted to the bodies to feed but were not observed ovipositing.

Active Decay (1)

This stage lasted from day 7 to day 12 (Table 1). Larval penetration of the abdomen was observed. This caused the body to deflate, thus marking the transition between the end of the bloated stage and the beginning of active decay (16).

The internal temperatures of the bodies were slightly higher than the environmental temperature (Fig. 1). Initially, the body weight decreased sharply as a result of the larval activity, but by the end of the stage it had begun to stabilize (Fig. 2).

From day 8 onwards, third larval instars of *C. albiceps* were collected and the lowest densities of *P. sericata* and *C. macellaria* were recorded; the majority of larvae stopped feeding and left the carcasses in order to pupate. *Chrysomya albiceps* and *C. megacephala* began to dominate the collected material.

Coleopteran adults were first observed during the active stage (Table 1) although in low numbers. Each genus occupied a special niche in the decomposition process: *Dermestes* and *Trox* were often observed feeding on the ventral surface of the bodies; *Edhopus* (Staphilinidae) were observed preying on dipteran larvae. With regard to the behaviour of the certain dipterans, *Ophyra capensis* (Muscidae) perched on the bodies in order to oviposit, while *Sarcophaga* and *Ravinia* (Sarcophagidae) behaved in the same way to lay of larvae although in smaller numbers; and *Fannia canicularis* was only observed feeding.

Advanced decay

It took place from approximately day 13 to day 22 (Table 1). The internal temperature continued to be slightly higher than environmental temperature (Fig. 1). By this stage, the majority of internal tissue had been removed, fluid had been lost and only a mucilaginous substance was observed. Tullis and Goff (17) described this

TABLE 1—Entomological succession for different stages of decomposition A = Adult; I = immature; E = egg.

Order	Family	Genus	Species	States												
				Fresh (0–1 days)		Floated (2–6 days)			Decay (7–12 days)		Advanced (13–22 days)		Dry (23–36 days)			
				A	F	A	F	I	A	I	A	I	A	I		
Diptera	Calliphoridae	<i>Chrysomya</i>	<i>megacephala albiceps</i>			×		×	×	×	×	×				
			<i>Cochilomyia macellaria</i>			×		×	×	×	×	×	×	×		
		<i>Comsomiops</i> sp					×		×					×		
		<i>Phaenicia sericata</i>	×	×	×		×	×	×	×	×	×	×	×		
	Cloropidae	<i>Liohrppelats</i>	sp								×		×			
	Fannidae	<i>Fannia</i>	<i>canicularis</i>			×			×		×		×			
	Muscidae	<i>Musca</i>	<i>domestica</i>			×			×		×		×			
		<i>Ophyra</i>	<i>Capensis</i>			×		×	×	×	×	×	×	×		
		<i>Synthesiomyia</i>	sp			×			×				×			
	Otitidae	<i>Euxesta</i>	sp			×			×		×		×			
	Phoridae	<i>Diplonevra</i>	sp			×			×		×					
	Piophilidae	<i>Piophila</i>	<i>foveolata</i>			×		×	×	×	×	×	×	×		
	Sarcophagidae	Not identified											×			
		<i>Oxysarcodexia</i>	sp			×			×		×		×			
		<i>Ravinia</i>	sp								×					
		<i>Sarcophaga</i>	sp					×		×						
		<i>Synthesiomyia</i>	sp											×		
		Not identified											×			
Coleoptera	Anthicidae	Not identified											×			
	Cleridae	<i>Necrobia</i>	<i>rufipes</i>			×			×		×		×			
	Coccinellidae	Not identified									×					
	Dermestidae	<i>Dermestes</i>	sp			×		×	×	×	×		×	×		
	Elateridae	<i>Conoderus</i>	sp					×					×			
	Histeridae	<i>Hister</i>	<i>abbreviatus</i>						×				×			
	Scarabaeidae	<i>Copris</i>	sp											×		
		<i>Onthophagus</i>	sp							×						
		<i>Trox</i>	sp							×		×	×	×		
	Staphylinidae	<i>Copris</i>	sp							×						
		<i>Didnous</i>	sp							×		×				
		<i>Edaphus</i>	sp			×			×		×					
		<i>Ontholestes</i>	sp							×		×				
<i>Staphylinus</i>		sp							×		×					
Tenebrionidae	Not identified												×			
Acarina	Not identified													×		
Aracnidae	Lycenidae	Not identified			×				×							
	Pisauridae	Not identified					×									
	Araneidae	Not identified					×									
Diplopoda	Not identified	Not identified									×				×	
Diptyoptera	Blaberidae	<i>Hyporichnada</i>	sp									×				
		<i>Ischnoptera</i>	sp			×				×						
	Blattellidae	<i>Xestoblata</i>	sp							×		×				
Hemiptera	Reduviidae	Not identified			×											
Hymenoptera	Apidae	<i>Eulema</i>	sp								×					
	Colletidae	<i>Colletes</i>	sp			×										
	Formicidae	<i>Camponotus</i>	sp			×									×	
		<i>Dorymyrmes</i>	sp		×	×				×		×		×	×	
		<i>Odontomachus</i>	sp			×						×		×	×	
		<i>Pheidole</i>	sp		×	×				×		×		×	×	
		<i>Solenopsis</i>	sp		×	×				×		×		×	×	
		<i>Tranopelta</i>	sp			×				×		×		×		
	Vespidae	Not identified													×	
		<i>Polystes</i>	sp			×				×					×	
	Isopoda	Scyphacidae	<i>Scyphacella</i>	sp			×			×		×		×		
Lepidoptera	Lycenidae	<i>Hemiagros</i>	sp			×										
Miriapoda	Not identified	Not identified				×										
Thysanoptera	Not identified	Not identified														

viscous material as by-products of decomposition or BOD. The high liquid component of the decomposing carcass is characteristic of the beginning of advanced decay.

The number of adult dipterans observed was notably low. Individuals belonging to the following genera were observed: *Euxesta* (Otitidae), *Diplonevra* (Phoridae) and *Liohrppaltes* (Cloropidae) (Table 1). Furthermore, a large number of *Ophyra capensis*

larvae were seen feeding on the BOD. Ants of the genera *Dorymyrmes*, *Solenopsis*, and *Pheidole* continued to be the main predators of dipteran larvae.

Larvae of *Piophila foveolata* (1st instar) were seen for the first time on day 18 (Table 2) sharing the substrate with *O. capensis*. Skin rupture and exposure of bones such as the skull became generalized in the pigs. During days 20 to 22 skin humidity and BOD decreased.

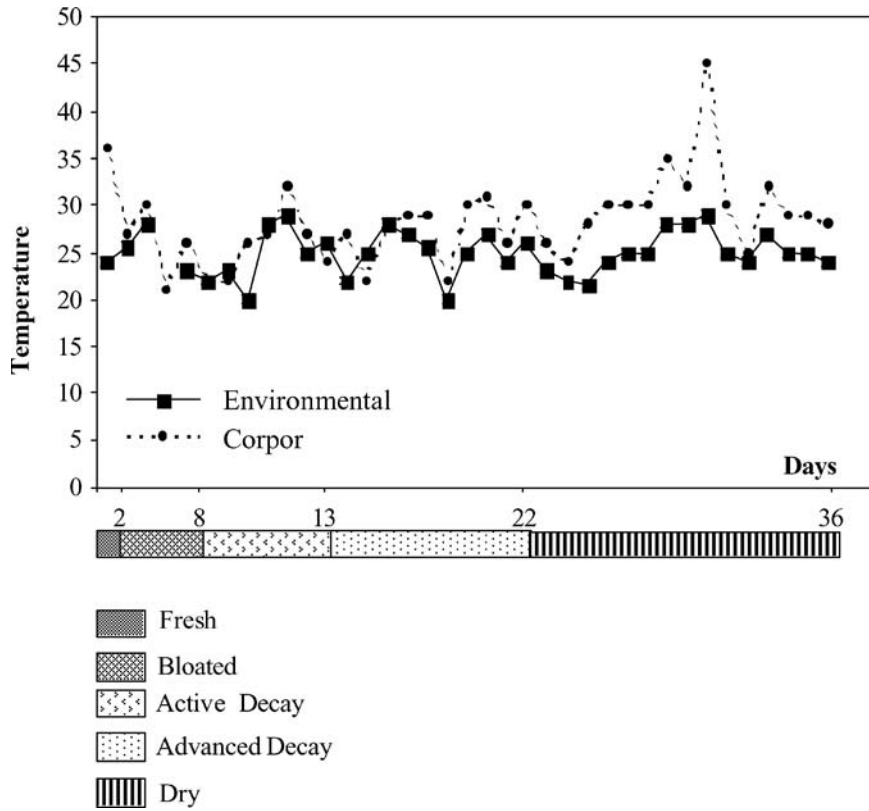


FIG. 1—Daily temperature variations related with decomposition phase. These curves, were obtained with the average atmospherical and rectal temperatures in the three pigs, taken once a day, during the whole period of sampling and at the same hour.

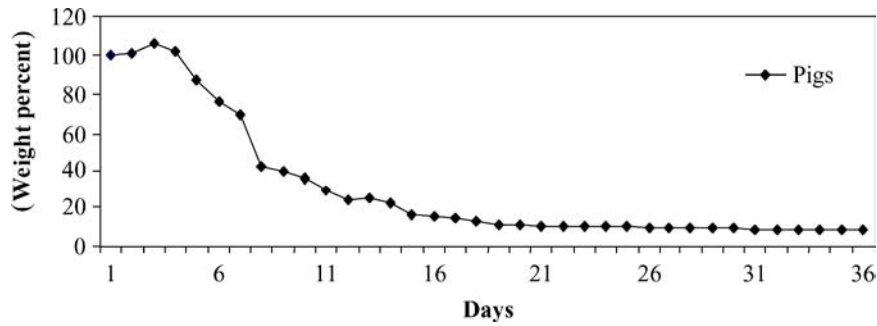


FIG. 2—Rate of biomass removal indicated as percent of weight. This figure was obtained with the average body weight of the three pigs during every period of sampling and at the same hour of the day.

TABLE 2—Matrix occurrence of immature stadia of Diptera and Coleoptera Species L1 = 1st instar diptera; L = larva coleoptera; 1 = presence; 0 = absence.

Genus	Species	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36		
<i>Phaenicia</i>	<i>sericata</i>	0	L1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
<i>Cochliomyia</i>	<i>macellaria</i>	0	0	L1	1	1	1	1	1	1	1	1	1	1	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>Chrysomya</i>	<i>megacephala</i>	0	0	0	L1	0	1	0	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>Chrysomya</i>	<i>albiceps</i>	0	0	0	0	0	0	L1	0	1	1	1	1	0	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>Ophyra</i>	<i>capensis</i>	0	0	0	0	0	0	0	0	0	0	0	L1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	
<i>Piophil</i>	<i>foveolata</i>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	L1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	0	0
<i>Trox</i>	sp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	L	1	1	1	1	0	0	1	1	1	1	1	1	1	1	0	0
<i>Dermestes</i>	sp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	L	1	1	1	1	1	0	1	1	1	

Dry remains

The transition between advanced decay and dry remains took place gradually (days 23–36). Adults and larvae of *O. capensis* previously dominating the advanced stage decreased considerably. *P. foveolata* became the indicator species feeding on the remaining mucilaginous material left under the skin and on the ground. Larvae of *Dermestes* sp and *Trox* sp, frequently observed at this stage, preferred to feed on skin, bone and cartilage, thus performing an important role in the skeletonisation process.

A list of species collected in Medellín is also given (Appendix).

Discussion

This study sought to provide new alternatives to complement existing methods of accurately estimating the postmortem interval (PMI). Similarly, we sought to improve the model database used to compare taxa collected on human remains. Therefore, occurrence matrix was drawn up for the species of Diptera (*P. sericata*, *C. macellaria*, *C. megacephala*, *C. albiceps*, *O. capensis*, and *P. foveolata*) and Coleoptera (*Dermestes* sp and *Trox* sp) totally dependent on the carcass for the development of their life cycle (Table 2). Together with the physical characteristics of the body, indicator species were established for each stage of decomposition in the urban area of Medellín during the rainy season (Table 1).

From the succession table obtained in this study, we can deduce that if only eggs were observed on a human body, found in similar micro-environmental conditions to those of the study, then the body will have a PMI of less than seven hours. Similarly, if the samples found are 3rd instar larvae of *P. sericata*, *C. macellaria* and 1st instar larvae of *C. megacephala*, then the estimated PMI would be between two and three days. However, if the latter samples were 3rd instar, then PMI would be calculated at three to seven days but not more since after day 8, it is very likely that 3rd instar larvae of *C. albiceps* would be collected. The PMI could be calculated even more accurately if the stage of decomposition of the body was also taken into account.

A determining step in drawing up the succession (Table 1) was describing the insect succession over time. This process is closely related to the stage of decomposition of the body, the micro-climatic conditions and the geographic region. As Wolff et al. (28) has been shown, the differences in the species composition of the first species of the colonization process in order to reach a body depend on the characteristics of each region and its season.

In this study, the first colonisations, or primary species for the urban area of Medellín during the rainy season, were *P. sericata* and *C. macellaria*. The ovipositing adults showed a preference for natural orifices (mouth, nose and anus) as well as the darkest and most humid body parts, which was also observed by Norris (40) and Tullis & Goff (17). *Phaenicia sericata* was the first species to oviposit and were most abundant for the first six days. *C. macellaria* was present from day 2 to day 15, *C. megacephala* from day 3 to day 22, *C. albiceps* from day 6 to day 18.

C. megacephala and *C. albiceps* are exotic species (41,42) and are considerably more abundant than native species. *C. albiceps* was seen to be feeding in large groups on the surface, whereas *C. megacephala* fed on internal tissues. These species are known to prey on one another and this physical separation could avoid that *C. megacephala* was not preyed upon by *C. albiceps* and allowed their presence throughout the advanced stage of decomposition. These facts together with their great propensity for dispersion could affect normal succession patterns and also the indicator species for PMI in this region.

Adults and larvae of *O. capensis* (Muscidae), and *P. foveolata* (Piophilidae) dominated the carcass at the advanced stage of decomposition. The mucilaginous material (BOD) provides a suitable substrate for the development of these dipterans. A similar situation has been reported for human remains found in waterlogged conditions (13).

Larvae of Piophilidae were collected from day 18 onwards, marking the transition between active and advanced decay. In other regions, these species have arrived at very different times—from 26 days to six months after death (43–45,12).

During the last stage of body decomposition, *Dermestes* sp and *Trox* sp (Coleoptera) arrived and started the cleaning process of the bones, as was described previously (1,16). However, the number of former species was very inferior to the number of species of the Diptera. This difference is probably due to the specific urban characteristics of the habitat where the study was done.

The general pattern of succession showed that as decay advanced, Diptera were followed by Coleoptera, coinciding with the study by Wolff et al. (28) carried out in the same city, with the same experimental model but during the dry season in the first half of the year. However, differences were found with regard to the number of species, whereas in this study species were collected belonging to 12 orders and 42 genera (Table 1), in the previous study, only 7 orders and 31 genera were found.

Ants played a determining role in the time taken for decomposition. The process was slowed down by the removal of large quantities of dipteran eggs. *Solenopsis* sp was mainly responsible for this. Similar results were reported elsewhere (12,16,46,47) whereas Anderson et al. (12) suggested that ants do not have an impact on the process.

This study reflects the importance in determining and interpreting succession patterns of indicator species in each bio-climatic zone of forensic importance. In the tropics, given that ecological conditions are so different within the same geographic region, it is impossible to extrapolate results and even less feasible to make comparisons with temperate countries.

Acknowledgements

This study received financial support, in part, from Colciencias (Instituto Colombiano para el Desarrollo de la Ciencia y la Tecnología), project 1115-05-11503 and from the Universidad de Antioquia, Colombia and from Drs Jeffrey Wells and Carl Lowenberger for the revision of manuscript.

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