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Article

Performance of sampling methods of Coleoptera associated with carcasses

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Abstract

We examined the performance of three different coleopterofauna sampling methods associated with pig carcasses in an area of Caatinga. In order to capture these insects we used different traps including trays, pitfall, and modified Shannon, during the dry and rainy seasons. A total of 4 851 individuals were collected, belonging to 88 species. Active collection using trays was the most abundant (4 474), followed by pitfall traps (328), and modified Shannon (49). Observed high complementarity values (ranging from 0,67 to 0,89), among the three types of collection methods, demonstrate the importance of using the three methods in conjunction to obtain sampled richness. The richness estimators Chao1 and ACE have confirmed the efficiency of the assembly, which was responsible for fathering more than 70% of the richness estimated for both seasons.

Additional keywords: beetles, forensic entomology, postmortem interval, richness estimators.

Introduction

The insects have different alimentary habits and can be found in various environments, including crime scenes. This fact is supported by forensic entomology: study of insects and other arthropods associated with crime scenes and dead bodies (Catts and Goff 1992). The objectives of this science is in determining how, when and, in particular, where the death occurred with information taken from insects found on or near corpses (Keh 1985).

Beetles are important insects for forensic entomology, they are: necrophagous, feeding on tissues, and thus directly accelerate decomposition rates; predators feeding on larvae, pupae and adults, and omnivorous feeding on body and associated fauna. In either case, the beetles' actions may affect decomposition rates. Moreover, all ecology categories can be informative for thanatology (Santos *et al.* 2013, Santos 2014).

In a study, New (2001) lists 18 of the most used collection methods in surveys of coleopterofauna. According to the authors, certain methods increase the sampling range of a particular assembly, while excludes species not associated with the provided resource. Three different methods are the most used in faunal surveys of forensic entomology: 1) active collecting (trays), 2) pitfalls, and 3) modified Shannon (Mise *et al.* 2007, Rosa *et al.* 2011, Silva and Santos 2012, Mayer and Vasconcelos 2013, Santos *et al.* 2014a). This study reports the performance of three different sampling methods on Coleoptera associated with carcasses, using complementarity index and richness estimators.

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Material and Methods

The study was carried out at the private conservation unit Fazenda Almas, in São José dos Cordeiros, state of Paraíba, Brazil (lat 7° 28' 19" S, long 36° 53' 40" W). The reserve covers 3 505 ha (600-720 m.a.s.l.) and the vegetation varies from open to dense arboreal Caatinga, with a strong deciduous characteristic during the dry season. During the dry season (October 2010) and rainy season (February 2011), two pig carcasses with about 15 kg in weight were used in both seasons. The animals were killed with a single gunshot to the head. Each carcass was exposed in an iron cage (3x10 cm mesh opening) to prevent interference from vertebrate scavengers.

Underneath the cage a tray trap was buried at ground level containing sawdust in order to actively collect Coleoptera that take shelter under the carcasses. Four pitfall traps were placed around the cage, 1 m away from it for passive capture. Each cage was also covered by a modified Shannon trap with a collector tube containing 70% alcohol at the apex. Coleoptera were collected daily from the traps until the end of the carcasses decomposition (15 days). Details of the procedures adopted are described in Alves *et al.* (2014) and Santos *et al.* (2014a). Three sampling methods were evaluated as complementarity pairs, using the Marczewski-Steinhaus index (Magurran 2011).

The Chao1 and ACE nonparametric estimators were used to estimate species richness during each season and sampling method, using species abundance data for calculations with the software EstimateS (Colwell 2013). The data were subjected to 1 000 randomizations without replacement (Walther and Moore 2005). The software also constructed an observed species (S_{obs}) curve, equivalent to the species accumulation curve. This data was included in all analyzes as the baseline for comparing the performance of the estimators.

The material used was incorporated into the "Coleção Entomológica do Departamento de Sistemática e Ecologia" at "Universidade Federal da Paraíba" (DSE/UFPB). The study was authorized by the Ethic Commission for the Usage of Animals of the same institution (CEUA/UFPB).

Results and Discussion

A total of 4 851 individuals belonging to 19 families and 88 species were collected. Among the sampling methods, the active sampling in trays (4 474) was the most abundant, followed by pitfalls (328) and modified Shannon traps (49) (Table 1).

Table 1. Coleoptera species associated with pig carcasses and their respective sampling methods in an area of Caatinga (adapted from Santos *et al.* 2014a).

Ecological category	Family	Species	Sampling method			Total
			Active	Pitfall	Shannon	
Necrophagous	Cleridae	*Necrobia rufipes* De Geer, 1775	107	1	12	120
	Dermestidae	*Dermestes maculatus* De Geer, 1774	328		2	330
		Dermestes haemorrhoidalis Küster, 1852	3			3
	Trogidae	*Omorgus suberosus* Fabricius, 1775	62	13		75
Predator/Parasite	Carabidae	*Loxandrus* sp.	16	49		65
		Carabidae spp. (6)	6	4		10
	Histeridae	*Eremosaprinus* sp.	4			4
		Euspilotus azureus (Sahlberg, 1823)	186	2		188
		Euspilotus sp.	441	20		461
		Hister punctifer Paykull, 1811	80	2		82
		Hololepta reichii Marseul, 1853	6			6
		Omalodes foveola Erichson, 1834	63	2	2	67
		Phelister sp.	101	13		114
		Xerosaprinus diptychus (Marseul, 1855)	290	41		331
		Histeridae spp. (5)	11			11

Table 1. Cont. Coleoptera species associated with pig carcasses and their respective sampling methods in an area of Caatinga (adapted from Santos *et al.* 2014a).

Ecological category	Family	Species	Sampling method			Total
			Active	Pitfall	Shannon	
Predator/Parasite	Hydrophilidae	Hydrophilinae sp.	3			3
	Staphylinidae	*Aleochara bonariensis* Lynch, 1884	100	2		102
		Atheta iheringi Bernhauer, 1908	1 631	53	1	1 685
		Acylophorus sp.	1			1
		Belonuchus spp. (2)	50			50
		Heterothops spp. (2)	15			15
		Philonthus figulus Erichson, 1840	19	2	1	22
		Philonthus spp. (2)	295	8	1	304
		Xenopygus analis (Erichson, 1840)	2			2
		Staphylinidae spp. (2)	3			3
Omnivorous	Nitidulidae	*Stelidota geminata* (Say, 1825)	356	13	25	394
		Nitidulidae sp.		1		1
	Scarabaeidae	*Ataenius* sp.	3	1		4
		Ateuchus carbonarius (Harold, 1868)	119	10		129
		Canthidium manni Arrow, 1913	43	4		47
		Canthon sp.		1		1
		Coprophanaeus pertyi (Olsoufieff, 1924)	1	1		2
		Deltochillum verruciferum Felsche, 1911	4	9		13
		Dichotomius geminatus (Arrow, 1913)	20	5		25
		Dichotomius nisus (Olivier, 1789)	9			9
		Ontherus digitatus Harold, 1868	1			1
		Onthophagus hirculus Mannerhein, 1829	19	3		22
		Trichillum sp.	27			27
		Uroxys sp.	10	1		11
		Aphodiinae spp. (2)	3	5		8
		Melolonthinae sp.			1	1
	Tenebrionidae	Tenebrionidae spp. (7)	13	24		37
Incidental	Anobiidae	Anobiidae sp.	6	1		7
	Bostrichidae	Bostrichidae sp.	3			3
	Chrysomelidae	Chrysomelidae spp. (8)	1	16	1	18
	Curculionidae	Curculionidae spp. (10)	7	11	3	21
	Elateridae	Elateridae spp. (2)		8		8
	Erotylidae	Erotylidae sp.	4			4
	Melyridae	Melyridae sp.		1		1
	Mordellidae	Mordellidae sp.	1	1		2
	Ptiliidae	Ptiliidae sp.	1			1
Total			4 474	328	49	4 851

The greater species abundance and diversity in the tray traps over other methods are because this trap looks like a big baited pitfall trap, simulating the ground below the carcass. In this environment, larvae of Diptera establish themselves while feeding on decaying tissues and then pupate. These events attract a lot of predator beetles as Histeridae and Staphyliniadae, which can easily be actively collected.

Despite the low relative abundance, the pitfalls traps obtained a high species richness (53). This data was observed because of many incidental species as Chrysomelidae and Curculionidae that were collected through in traps. The modified Shannon was inefficient for beetle sampling with low species richness and high relative abundance of only two species: *Stelidota geminata* (Say, 1825) (Nitidulidae) (51,0%) and *Necrobia rufipes* De Geer, 1775 (Cleridae) (24,5%). However, Mise *et al.* (2007) collected 2 016 individuals and 64 species, most of them Staphylinidae, with a similar trap. The disparity of these data is probably due to differences in the other parts of the methodology adopted by the authors.

The use of different capture methods is very important to obtain a considerable inventory of insect diversity of an area. Because alternative capture methods may result in very different species assemblages (Sørensen *et al.* 2002). As shown in Table 1, few species were sampled by the three collection methods.

Therefore, the purpose of comparing sample methods is to highlight their differences, emphasizing that they are complementary to each other. Species with crepuscular or nocturnal habits are more easily collected in passive traps (Hernández 2007). In this study, 75,4% of *Loxandrus* sp. (Carabidae) and 69,2% of *Deltochillum verruciferum* Felsche, 1911 (Scarabaeidae) were sampled by pitfall traps. On the other hand, necrophagous species are better collected through active sampling on the carcasses or below them in tray traps (Mayer and Vasconcelos 2013). In this study, 99,4% of *Dermestes maculatus* De Geer, 1774 (Dermestidae) and 89,2% of *N. rufipes* (Cleridae) were collected using this method. These differences were confirmed through the Marczewski-Steinhaus complementarity index analysis between collection methods (Table 2).

In the complementarity index, sampling methods are evaluated in pairs, and values range from 0 to 1, respectively representing, no complementarity and total complementarity, *i.e.* the bigger the difference in assembly between pairs of analyzed methods, more complementary they are to each other (Magurran 2011). The observed high values varied between 0,671 and 0,891, demonstrating how these collection methods are complementary to each other in order to obtain the sampled richness.

The importance of multiple collection methods is also confirmed for other arthropods. Sørensen *et al.* (2002) employed six sampling methods to inventory and estimate spider diversity. The complementary values obtained indicated that among all 15 possible pairs, only two of the values were below 0,5.

Zanetti *et al.* (2016) evaluated two collection methods of Coleoptera in carcasses, active collection and pitfall traps, through a method that combined the number of beetles for each family/sampling unit/sampling method. A large discrepancy was found for all families, with some of them being more abundant with active collection, while others were more abundant with pitfall collection, which highlighted the need to use all methods.

The advantage of using various collection methods is even stronger when we compare the observed and estimated richness of associated species. The estimators Chao1 and ACE indicate that collection methods used together respectively account for 84,5% and 73,5% of the estimated diversity during the dry season, and 80,3% and 74,4% during the rainy season (Table 3). The species accumulation curve did not reach the asymptote and richness curves estimated by Chao1 and ACE were outside the confidence interval in both seasons (Figure 1 a, b). However, collection methods used ensured over 70% of the richness estimated by both estimators, demonstrating the efficiency of the paired methods.

A higher efficiency was confirmed for active collection in trays of the collection methods analyzed. Chao1 and ACE showed 75,6% and 78,3%, respectively, of the sampled coleopterofauna estimated richness where observed richness was closest to the estimated richness (Table 3). The use of species richness estimators is especially important in tropical invertebrate inventories, in which the observed richness hardly reaches an asymptote, even after intense sampling (Gotelli and Colwell 2001). The species accumulation curves did not reach an asymptote for any of the methods. Also, the richness curves estimated by Chao1 and ACE were outside the confidence interval of the species accumulation curves (Figure 1 c, e).

Table 2. Marczewski-Steinhaus complementarity index of sampling methods of Coleoptera species associated with pig carcasses in an area of Caatinga.

Sampling method	Active	Pitfalls	Shannon
Active	0	0,671	0,881
Pitfalls		0	0,891
Shannon			0

Table 3. Estimated (Chao1 and ACE) and observed richness (S_{obs}) among seasons and sampling methods of Coleoptera species associated with pig carcasses in an area of Caatinga.

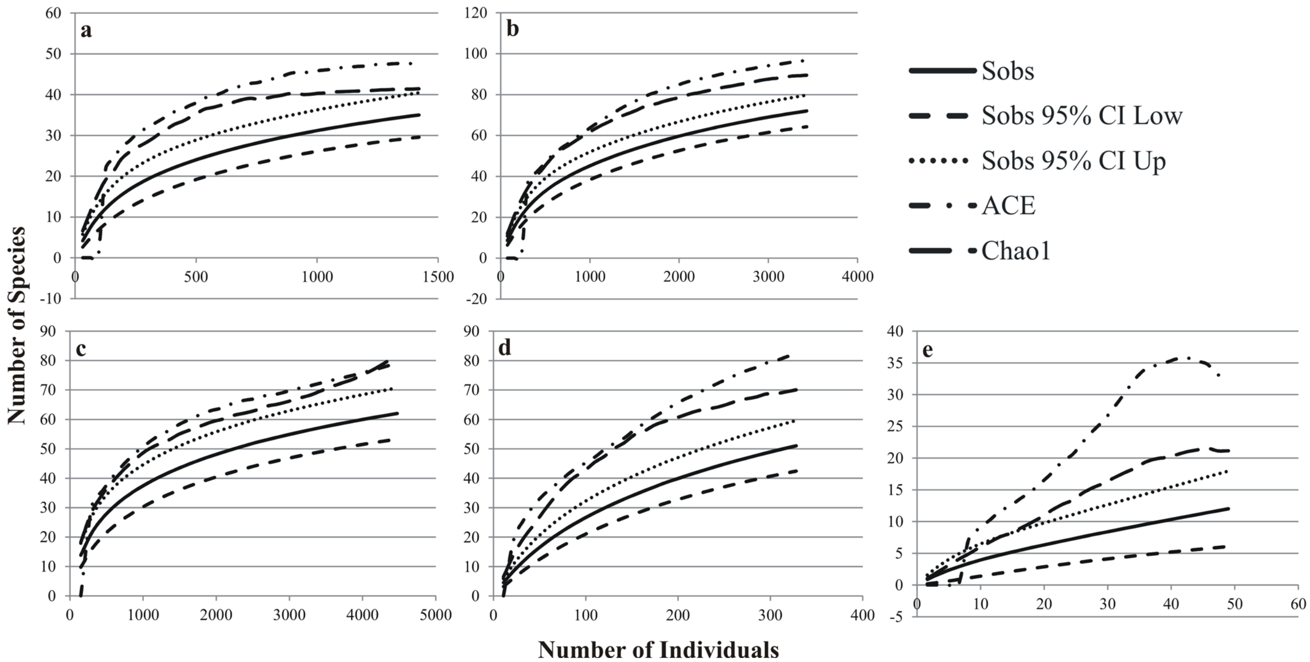
Richness estimator	Season		Sampling method		
	Dry	Rainy	Active	Pitfalls	Shannon
Chao1	41,42	89,67	82,00	70,03	21,4
ACE	47,62	96,77	79,14	82,38	32,00
S_{obs}	35	72	62	51	12

In general, we observed that the ACE estimator overestimates richness compared to Chao1. Both estimations are based on abundance data and value rare species, however in the Chao1 estimation rare species are those represented by only one individual, while in the ACE estimation, rare species are those represented by up to 10 individuals. Thus, there is a greater chance that in the same number of individuals there are more rare species according to ACE than according to Chao1, which influences the estimated values (Walther and Moore 2005, Magurran 2011).

Chao1 was the richness estimator with the best performance between seasons and traps in the studied assembly, except active collection (Table 3, Figure 1). However, according to the taxon studied conclusions can be different. Foggo *et al.* (2003) studied marine macro-invertebrates and also concluded that Chao1 is the best choice for accurately estimating diversity. On the other hand, Dias and Bonaldo (2012), studying spider diversity, concluded that ACE was a better estimator, due to its performance curve with a greater tendency towards stabilization. In extremely abundant groups and/or groups difficult to sample the whole community, such as termites (Ernesto 2013), ants (Longino *et al.* 2002, King and Porter 2005) and ground beetles (Brose 2002), estimators that are based on incidence, such as ICE and Chao2, obtain the best results.

The results also indicate that the collection methods may be improved to try to approximate the observed richness to the estimated richness. The number of pitfall traps could be extended to eight, covering thus a larger area surrounding the carcass. Following the same principle, the modified Shannon trap could have a larger base, extending beyond the area of the tray. However, the most efficient combination of possible methods, able to indicate how to better represents the species composition, still requires future studies.

A sampling methodology for criminal investigations should focus on active collecting on and under the body due the facility and efficiency. However, inventory studies of coleopterofauna associated with carcasses must use different sampling methods. Furthermore, other insects, such Diptera and Hymenoptera, can be additionally collected (Alves *et al.* 2014, Santos *et al.* 2014b). Comparisons of insect fauna associated with decaying carcasses are quite difficult due to the fact that the studies conducted did not follow standardized methods (Amendt *et al.* 2007). Necrophilous beetle studies apply few different methodologies as well (Mise *et al.* 2007, Rosa *et al.* 2011, Santos *et al.* 2014a). Furthermore, sometimes different substrates are also used (Santos and Alves 2016, Santos and Santos 2016). As stressed by Pujol-Luz *et al.* (2008), forensic entomology needs to establish guidelines to ensure a minimum standardization of the procedures



Figuree 1. Performance of nonparametric richness estimators (Chao1 and ACE) and observed richness (S_{obs}) among the dry (a) and rainy (b) seasons; and the sampling methods, Active (c), Pitfalls (d) and Shannon (e), of Coleoptera species associated with pig carcasses in an area of Caatinga.

adopted for the sampling of entomological evidence, and thus promote a great establishment of this science.

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Literature Cited

Alves ACF, Santos WE, Farias RCAP, Creão-Duarte AJ. 2014b. Blowflies (Diptera, Calliphoridae) associated with pig carcasses in a Caatinga area, Northeastern Brazil. *Neotropical Entomology,* 43(2): 122-126.

Amendt J, Campobasso CP, Gaudry E, Reiter C, LeBlane HN, Hall MJR. 2007. Best practice in forensic entomology – standards and guidelines. *International Journal of Legal Medicine,* 121(2): 90-104.

Brose U. 2002. Estimating species richness of pitfall catches by non-parametric estimators. *Pedobiologia,* 46(2):101-107.

Catts EP, Goff ML. 1992. Forensic entomology in criminal investigations. *Annual Review of Entomology,* 37: 253-272.

Colwell RK. 2013. EstimateS: Statistical estimation of species richness and shared species from samples. Available in: http://viceroy.eeb.uconn.edu/EstimateS/. Accessed: 22 Nov 2015.

Dias SC, Bonaldo AB. 2012. Abundância relativa e riqueza de espécies de aranhas (Arachnida, Araneae) em clareiras originadas da exploração de petróleo na bacia do rio Urucu (Coari, Amazonas, Brasil). *Boletim do Museu Paraense Emílio Goeldi. Ciências Naturais,* 7(2): 123-152.

Ernesto MV. 2013. Térmitas de duas áreas de Floresta Atlântica Brasileira: Uma análise do desempenho de estimadores não paramétricos. [M.Sc. Dissertation]. João Pessoa: Universidade Federal da Paraíba. 71 p.

Foggo A, Attrill MJ, Frost MT, Rowden AA. 2003. Estimating marine species richness: an evaluation of six extrapolative techniques. *Marine Ecology Progress Series,* 248: 15-26.

Gotelli NJ, Colwell RK. 2001. Quantifying biodiversity: procedures and pitfalls in the measurement and comparison of species richness. *Ecology Letters,* 4(4): 379-391.

Hernández MIM. 2007. Besouros escarabeíneos (Coleoptera: Scarabaeidae) da Caatinga paraibana, Brasil. *Oecologia brasiliensis,* 11(3): 356-364.

Keh B. 1985. Scope and applications of forensic entomology. *Annual of Review Entomology,* 30: 137-154.

King JR, Porter SD. 2005. Evaluation of sampling methods and species richness estimators for ants in upland ecosystems in Florida. *Environmental Entomology,* 34(6): 1566-1578.

Longino JT, Coddington JA, Colwell RR. 2002. The ant fauna of a tropical rain forest: estimating species richness in three different ways. *Ecology,* 83(3): 689-702.

Magurran AE. 2011. Medindo a Diversidade Biológica. Curitiba: Universidade Federal do Paraná. 261 p.

Mayer ACG, Vasconcelos SD. 2013. Necrophagous beetles associated with carcasses in a semi-arid environment in Northeastern Brazil: implications for forensic entomology. *Forensic Science International,* 226(1-3): 41-45.

Mise KM, Almeida LM, Moura MO. 2007. Levantamento da fauna de Coleoptera que habita a carcaça de *Sus scrofa* L., em Curitiba, Paraná. *Revista Brasileira de Entomologiam,* 51(3): 358-368.

New TR. 2010. *Beetles in Conservation*. Hong Kong: Graphicraft Limited. 237 p.

Pujol-Luz JR, Arantes LC, Constantino R. 2008. Cem anos da entomologia forense no Brasil (1908-2008). *Revista Brasileira de Entomologia,* 52(4): 485-492.

Rosa TA, Babata MLY, Souza CM, Sousa D, Mello-Patiu CA, Vaz-de-Mello FZ, Mendes J. 2011. Arthropods associated with pig carrion in two vegetation profiles of Cerrado in the State of Minas Gerais, Brazil. *Revista Brasileira de Entomologia,* 55(3): 424-434.

Santos DS, Santos WE. 2016. Association of necrophilous beetles (Coleoptera) with rat carcasses in a semi-arid area, Northeastern Brazil. *Entomotropica,* 31(20): 167-171.

Santos WE, Alves ACF, Creão-Duarte AJ. 2014a. Beetles (Insecta, Coleoptera) associated with pig carcasses exposed in a Caatinga area, Northeastern Brazil. *Brazilian Journal of Biology,* 74(3): 649-655.

Santos WE. 2014. Papel dos besouros (Insecta, Coleoptera) na Entomologia Forense. *Revista Brasileira de Criminalística,* 3(2): 36-40.

Santos WE, Alves ACF, Farias RCAP, Creão-Duarte AJ. 2013. Ecological roles of Coleoptera associated with carcasses in Caatinga. *EntomoBrasilis,* 6(3): 248-250.

Santos WE, Carneiro LT, Alves ACF, Creão-Duarte AJ, Martins CF. 2014b. Stingless bees (Hymenoptera: Apidae: Meliponini) attracted to animal carcasses in the Brazilian dry forest and implications for forensic entomology. *Sociobiology,* 61(4): 490-493.

Santos WE, Alves ACF. 2016. Registro de besouros (Coleoptera) em carcaça bovina e rápida decomposição sob condições naturais em ambiente semiárido, nordeste do Brasil. *Entomotropica,* 31(14): 105-108.

Silva RC, Santos WE. 2012. Fauna de Coleoptera associada a carcaças de coelhos expostas em uma área urbana no Sul do Brasil. *EntomoBrasilis,* 5(3): 185-189.

Sørensen LL, Coddington JA, Scharff N. 2002. Inventorying and estimating subcanopy spider diversity using semi-quantitative sampling methods in an afromontane forest. *Environmental Entomology,* 31(2): 319-330.

Walther BA, Moore JL. 2005. The concepts of bias, precision and accuracy, and their use in testing the performance of species richness estimators, with a literature review of estimates performance. *Ecography,* 28(6): 815-829.

Zanetti NI, Camina R, Visciarelli EC, Centeno ND. 2016. Active search on carcasses versus pitfall traps: a comparison of sampling methods. *Neotropical Entomology,* 45(2): 221-226.