

Analysis of the Successional Patterns of Insects on Carrion in Southwest Virginia

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ABSTRACT Studies of carrion-insect succession on domestic pig, *Sus scrofa* L., were conducted in the spring and summer of 2001 and 2002 in Blacksburg, VA, to identify and analyze the successional patterns of the taxa of forensic importance in southwest Virginia. Forty-seven insect taxa were collected in the spring. These were represented by 11 families (Diptera: Calliphoridae, Sarcophagidae, Muscidae, Sepsidae, Piophilidae; Coleoptera: Staphylinidae, Silphidae, Cleridae, Trogidae, Dermestidae, Histeridae). In the summer, 33 taxa were collected that were represented by all of the families collected in the spring, except Trogidae. The most common flies collected were the calliphorids: *Phormia regina* (Meigen) and *Phaenicia coeruleiviridis* (Macquart). The most common beetles were *Creophilus maxillosus* L. (Staphylinidae), *Oiceoptoma noveboracense* Forster, *Necrophila americana* L., *Necrodes surinamensis* (F.) (Silphidae), *Euspilotus assimilis* (Paykull), and *Hister abbreviatus* F. (Histeridae). Occurrence matrices were constructed for the successional patterns of insect taxa during 21 sampling intervals in the spring and 8 intervals in the summer studies. Jackknife estimates (mean \pm 95% confidence limits) of overall Jaccard similarity in insect taxa among sampling intervals in the occurrence matrices were 0.213 ± 0.081 (spring 2001), 0.194 ± 0.043 (summer 2001), 0.257 ± 0.068 (spring 2002), and 0.274 ± 0.172 (summer 2002). Permutation analyses of the occurrence matrices showed that the patterns of succession of insect taxa were similar between spring 2001 and 2002 ($P = 0.001$) and between summer 2001 and 2002 ($P = 0.007$). The successional patterns seem to be typical for the seasonal periods and provide data on baseline fauna for estimating postmortem interval in cases of human death. This study is the first of its kind for southwest Virginia.

KEY WORDS forensic entomology, postmortem interval, successional pattern, occurrence matrix, Jaccard coefficient

MEDICOCRIMINAL OR MEDICOLEGAL entomology is the area of forensic entomology that focuses on the use of arthropod evidence in criminal investigations, particularly those involving wrongful or unexplained death (Hall 1990, 2001). Data collected by forensic entomologists on the arthropod populations associated with a deceased individual can be used to determine the place where death occurred and to estimate the postmortem interval (PMI), or the time that elapsed since death (Hall 2001).

Information on the development and succession of arthropod species (mainly insects) is used in two primary ways to estimate the PMI of human remains. In cases where death was recent, the PMI can be estimated by analyzing the degree of development of early-arrival species that colonize the corpse (Goff 1993, Wells and Lamotte 2001). Data on the development of forensically important species derived from controlled studies (e.g., Kamal 1958, Greenberg and Szyska 1984, Greenberg 1991, Anderson 2000, Byrd and Allen 2001) are used to estimate the degree of

development of similar species collected on a corpse. The PMI of a corpse also can be estimated from the successional patterns of carrion-arthropods. Because insects arrive at a corpse in a predictable manner specific to the location and environmental conditions under which the remains are found (Payne 1965), the time of death can be estimated using succession data when insect development data are unavailable. In such cases, the composition of taxa found on a corpse at the time of discovery (corpse fauna) are usually compared with the composition of the arthropod assemblage at a given period of time derived from an animal model (baseline fauna) to estimate the PMI (Schoenly et al. 1996).

The two approaches for estimating PMI (i.e., based on arthropod development or successional patterns) are complementary, although each may be better suited for use during different stages of carcass decomposition. The use of development data are most applicable in the early phases of decomposition when the immature stages of the first colonizers are present. This approach enables investigators to establish a minimum PMI, because flies generally do not deposit their

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eggs on live individuals. Succession data are most valuable for estimating PMI in the later stages of decomposition when the earliest colonizers are no longer present (Goff and Flynn 1991).

Although the decomposing corpse represents a dynamic microecosystem that attracts different taxa as it changes (Goff 1993), the pattern of visitation on the corpse is somewhat predictable (Rodriguez and Bass 1983, Anderson 2001). The first and most significant insects to arrive at and oviposit on a corpse are typically species of necrophilous flies in the families Calliphoridae and Sarcophagidae (Watson and Carlton 2003). Predators of fly maggots, including beetles in the families Silphidae, Staphylinidae, and Histeridae, arrive later to feed on the maggots (Goff 1993). Late arrivers, such as beetle species in the family Dermestidae, invade a corpse in the dry stages of decomposition after early colonizing taxa have already left the remains (Rodriguez and Bass 1983, Watson and Carlton 2003). While succession of insects generally follows a similar pattern at the family level, there seems to be variation at the genus and species levels among locations (e.g., Reed 1958, Early and Goff 1986, Shean et al. 1993, Anderson and VanLaerhoven 1996).

The identification, occurrence, and successional pattern of forensically important taxa in specific regions are useful in estimating PMI (Anderson 2001). Studies on the successional patterns of arthropods (based on arthropod visitation) have been conducted throughout the world in climactically different areas using several different animal models (e.g., Bornemissza 1957, Reed 1958, Payne 1965, Rodriguez and Bass 1983, Early and Goff 1986, Anderson and VanLaerhoven 1996, Watson and Carlton 2003). However, there are still many regions for which no baseline successional data are available on forensically important arthropods. The purpose of this investigation was to collect data on carrion-insect succession (baseline fauna) for Montgomery County and nearby regions in southwest Virginia, which could be used for estimating PMI in cases of human death. The pattern of decomposition of the domestic pig (*Sus scrofa* L.) closely approximates that of a human corpse, and as such, the pig is generally the most acceptable animal for use as a human model in succession studies (Catts and Goff 1992). This study, therefore, describes the successional patterns of carrion insects on pig carcasses and examines the seasonal changes in these patterns through the analysis of occurrence matrices.

Materials and Methods

Study Site. Studies were conducted in the spring (late-April to June) and summer (late-June to July) of 2001 and 2002 at Kentland Farm (37°11' N, 80°25' W, altitude 608 m), a research facility of Virginia Polytechnic Institute and State University located in Montgomery County in southwest Virginia. Climate in the area is temperate, with average temperatures in the spring and summer of 15.0 and 20.7°C, respectively. The average annual rainfall is 102 cm, and the average

annual snowfall is 71 cm (National Weather Service, www.nws.noaa.gov).

Animals and Cages. We used commercially cross-bred pig carcasses obtained from the Swine Center at Virginia Tech in the successional studies conducted during the two periods (spring and summer) each year. Two pigs were used in each study. The animals in the spring studies were larger (41–45 kg) than those in the summer studies (23–27 kg). Each pig was killed by electrical shock early in the morning just after dawn. The carcasses were transported immediately to the test site at Kentland farm ≈13 km from the Swine Center. We examined the carcasses carefully for signs of fly oviposition before each was placed inside a test cage.

Each pig carcass was placed inside a wire cage with its legs pointing toward the hinged door on the front of the cage (Fig. 1). Each cage measured 92 by 92 by 153 cm and consisted of a metal frame constructed with 2.5-cm steel-welded tubing, which was enclosed with 1.27-cm mesh hardware cloth. The bottoms of cages were open so that the carcasses were in full contact with the ground. The cages also were staked to the ground to prevent disturbance of the carcasses by scavengers.

In each study, the two cages with pig carcasses were placed ≈92 m apart at the interface of open pasture and a thickly wooded area. Because decomposition and colonization of a carcass by insects is affected by the placement of the carcass (Shean et al. 1993), the cages were positioned so that both carcasses received direct sunlight until mid-day, although one carcass usually was slightly shaded during the afternoon.

Sampling Protocol. A typical sample included adult insects obtained from ≈10 aerial net sweeps above and around the carcass and ≈100 fly eggs or maggots collected using forceps directly off the carcass, when they were present. Pitfall traps consisting of a plastic cup filled with soapy water also were placed 8 cm from the head (two traps) and abdomen (two traps) of each carcass to collect crawling insects. We also recorded data on maggot mass temperatures (using a meat thermometer) at the time of sampling. Data on ambient temperature and rainfall at Kentland Farm were obtained online (<http://www.vaes.vt.edu/colleges/kentland/weather/>).

Sampling intensity in spring and summer studies was determined mainly by the rate of decomposition of the carcasses. In the spring, we sampled twice daily for the first 10 d, and thereafter, daily for 11 d; in the summer, we sampled twice daily for the first 3 d and daily for the next 5 d. Sampling ceased when the carcasses were in advanced decay.

Adult samples were placed immediately in individual 4.5-liter Hefty Onezip plastic bags (Pactiv, Lake Forest, IL) containing a piece of paper towel saturated with ethyl acetate. When they were collected, ≈50 maggots or fly eggs were placed directly into glass vials filled with KAA fixative (10 ml kerosene, 80–100 ml 95% ethanol, and 20 ml glacial acetic acid) (Catts and Haskell 1990), which was replaced with 80% ethyl alcohol within 24 h. The remaining group of 50 mag-

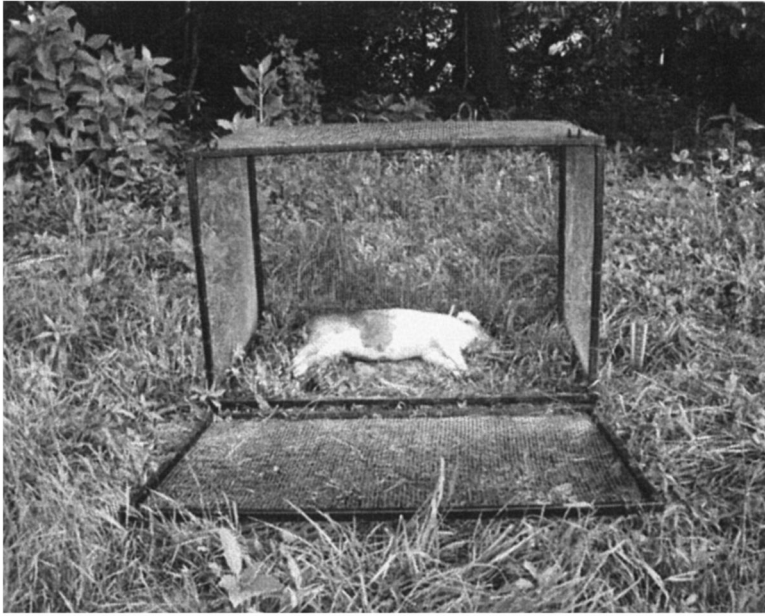


Fig. 1. Open-bottom cage used to isolate pig carcasses during the study. The hinged door at the front of the cage allowed easy access to the carcass.

gots or eggs was reared to the adult stage in 500-ml MicroGourmet SOLO cups (Solo Cup, Urbana, IL). We placed the maggots in pouches made with aluminum foil containing ground pork (Catts and Haskell 1990) and a wet paper towel to keep the insects and meat moist. Each pouch was placed in a rearing cup containing ≈ 7 cm of Hoffman vermiculite (A.H. Hoffman, Lancaster, NY) in which migrating maggots could pupate. A large hole was cut in the lid of each cup to provide aeration, and a piece of mesh cloth was placed under the lid to prevent maggots from escaping. We placed the rearing cups with maggots in an incubator set at 27°C. Adults that emerged were killed after a few days by placing the rearing cups in a freezer for 30 min. All collected and reared adults were pinned, labeled, and identified. Specimens also were sent off to taxonomic specialists for verification and vouchers were deposited in the Virginia Museum of Natural History at Virginia Tech.

Data and Statistical Analysis. We combined the data collected from the two pigs in each study to develop a diagram of succession of insect taxa and an occurrence matrix. Therefore, a total of four occurrence matrices were developed (for the spring and summer of 2001 and 2002). In an occurrence matrix, 1 and 0 represent the occurrence and absence, respectively, of a taxon within a sampling interval. For each of the occurrence matrices, we assessed the variability (mean \pm SD) in the number of taxa among sampling intervals and in the number of occurrences of each taxon during the study period. We also derived Jaccard similarity coefficients and developed diagrams to describe temporal changes in the between-sample similarities in composition of insects during the study

period (Schoenly 1992). The Jaccard metric ranges from 0 to 1, representing complete dissimilarity between sampling intervals for any taxon to a perfect match between sampling intervals (Schoenly 1992, Krebs 1999).

The Jaccard similarity diagrams were used to identify periods of taxonomic stasis (i.e., periods of little or no change in the composition of taxa) during succession based on both strict and loose definitions of this phenomenon. By the strict definition, taxonomic stasis occurred when the mean pairwise similarities for two or more consecutive sample-pairs were equal. By the loose definition, taxonomic stasis occurred when the confidence intervals for two or more consecutive pairwise similarities overlapped.

We used the between-sample, or pairwise, Jaccard similarities for each occurrence matrix to calculate an overall or mean similarity (S_{gmean}) of insect taxa throughout the entire study period (i.e., among all sampling intervals). The precision of S_{gmean} (i.e., $S_{gmean} \pm 95\%$ confidence limit [CL]) was determined using the Jackknife procedure (Meyer et al. 1986, Manly 1997, Krebs 1999). With the Jackknife procedure, we omitted each of the n sampling intervals in an occurrence matrix one-at-a-time, with replacement, and recomputed the overall similarity of taxa (S_{-i}) for the new occurrence matrix with $(n - 1)$ sampling intervals. The Jackknife estimate or pseudo-value of overall similarity (PV_i) for the occurrence matrix, with the i th sampling interval discarded, was calculated as

$$PV_i = nS_{gmean} - (n - 1)S_{-i} \quad i = 1 \dots n \quad [1]$$

where n is the number of sampling intervals in the original occurrence matrix, S_{gmean} is the overall sim-

ilarity for the original occurrence matrix with n sampling intervals, and S_{-i} is the partial estimate of S_{gmean} when the i th sampling interval is discarded. Finally, the PV_i values ($i = 1 \dots n$) were used to compute the Jackknife estimate of S_{gmean} as

$$\hat{S}_{gmean} = \frac{1}{n} \sum_{i=1}^n PV_i \quad [2]$$

along with its SE and 95% confidence intervals.

Overall Jaccard similarity indices for two occurrence matrices with similar numbers of sampling intervals and taxa can be equal, and yet the successional patterns of taxa in the matrices could differ. Therefore, S_{gmean} cannot be used with certainty to measure the degree of similarity between the successional patterns of species in two occurrence matrices of similar size. To determine the degree of similarity in species occurrence for corresponding periods in the two years (i.e., spring of 2001 and 2002 and summer of 2001 and 2002) and hence the degree of consistency in the successional patterns of insects for those periods, we applied a permutation approach based on Mantel's test, which has been used to compare distance or similarity matrices (Mantel 1967, Dietz 1983, Cheverud et al. 1989, Manly 1997). As is typical in this type of analysis (e.g., Cheverud et al. 1989, Manly 1997), we tested the null hypothesis of no similarity between the patterns of succession in the occurrence matrices for similar study periods between years.

We observed that in a few cases, a species was present in only one sampling interval during a study period in 1 yr but was absent during all sampling intervals in the corresponding period of the other year. These species were excluded in developing the respective occurrence matrices so that the matrices would be of similar size. For each of the adjusted occurrence matrices, we derived a Jaccard similarity matrix in which the value of the elements, i,j represents the degree of similarity of taxa between the i th and j th sampling intervals. The matrix is square ($n \times n$) and symmetric because the similarity of taxa between sampling intervals i and j is the same as that between j and i , and the similarity of taxa within an interval with itself (i.e., $i = j$) is 1.0. We next calculated the Pearson correlation coefficient (K_{obs}) between the two similarity matrices. A permutation distribution of K values was developed by carrying out successive correlations after corresponding rows and columns of one of the similarity matrices were randomly permuted simultaneously. That is, if for example, rows one and four of the similarity matrix were exchanged, columns one and four were also exchanged. This type of randomization had the effect of maintaining symmetry in the similarity matrix (Mantel 1967, Cheverud et al. 1989, Manly 1997). In all cases, we carried out 999 permutations to create a permutation distribution of 1,000 K -values (i.e., 999 permutation values plus K_{obs}), which has been shown to be sufficient for testing significance at the 5% level (Cheverud et al. 1989, Manly 1997). Statistical significance (P) of the observed correlation coefficient, K_{obs} , under the null

hypothesis of no similarity between the successional patterns of insect species in the two similarity matrices was determined by its position in the distribution of 1,000 K values. That is, the P value is the proportion of K values $\geq K_{obs}$, with a low value ($P < 0.05$) indicating that the successional patterns of taxa in the two similarity matrices are similar. All of the analyses on the occurrence and similarity matrices were carried out using MATLAB 6.5 (The MathWorks, Natick, MA) with the Image and Signal Processing toolboxes.

Results

Diagrams of the succession of insect taxa on decomposing pig carcasses during the spring and summer of 2001 and 2002 are shown in Figs. 2–4. Forty-seven insect taxa were observed in the spring, and 33 taxa were observed in the summer during the 2 yr. The earliest visitors to the pig carcasses in the spring and summer of both years were dipterans in the families Calliphoridae, Sarcophagidae, and Muscidae.

Approximately 60% of all flies collected were calliphorids, but this family was only represented by eight species. Of these, *Phormia regina* (Meigen) (Calliphoridae) was the most abundant in the spring, but was equally abundant with *Phaenicia coeruleiviridis* (Macquart) (Calliphoridae) in the summer. Both *Calliphora vicina* Robineau-Desvoidy (Calliphoridae) and *Calliphora vomitoria* L. (Calliphoridae) were collected in the spring studies, but not in the summer. *Cochliomyia macellaria* (F.) (Calliphoridae) was only collected during the summer. Although sarcophagids comprised a small proportion (< 5%) of all flies collected on the carcass, this family was represented by 11 species. The most abundant species in the spring was *Helicobia rapax* (Walker); the most abundant in both spring and summer was *Sarcophaga utilis* Aldrich.

Piophilidae was represented by only two species: *Stearibia nigriceps* (Meigen) and *Prochyliza xanthostoma* Walker. *Meroptilus minutus* (Wiedemann) was the most abundant species of Sepsidae in both spring and summer. Of the muscids that were identified, *Musca domestica* L. and *Hydrotaea leucostoma* Wiedemann were the most common. Dominant beetle species in spring and summer of both years included *Creophilus maxillosus* L. (Staphylinidae), *Oiceoptoma noveboracense* Forster, *Necrodes surinamensis* (F.), *Necrophila americana* L. (Silphidae), *Euspilotus assimilis* (Paykull), and *Hister abbreviatus* F. (Histeridae).

Mean pairwise Jaccard similarities for the successional patterns of insect taxa in the occurrence matrices (after the removal of rare species) are presented in Fig. 5. Jackknife estimates of overall Jaccard similarities and the precision of these estimates ($\hat{S}_{gmean} \pm 95\% \text{ CL}$) for each of the successional patterns were 0.213 ± 0.081 (spring 2001), 0.257 ± 0.068 (spring 2002), 0.194 ± 0.043 (summer 2001), and 0.274 ± 0.172 (summer 2002). Permutation analyses detected no differences between the similarity matrices for the spring of 2001 and the spring of 2002 ($K_{obs} = 0.60$; $P = 0.001$) and between those for the summer

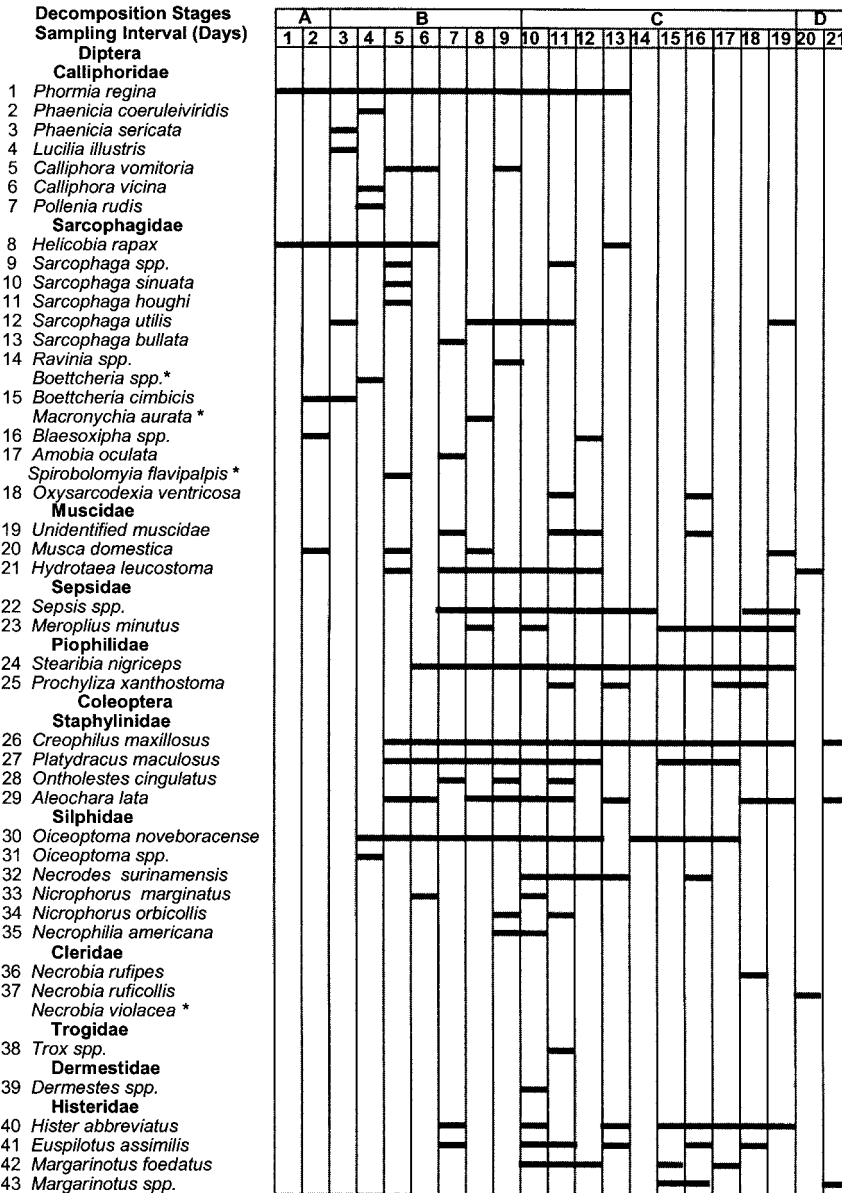


Fig. 2. Succession diagram for insect taxa on pig carcasses during 21 sampling intervals in the spring of 2001 in Blacksburg, VA. Stages of decomposition are shown and are (A) fresh, (B) bloat, (C) active decay, and (D) advanced decay and dry. *Unnumbered (rare) taxa that were observed only in one sampling interval in the spring of 1 yr but not during any sampling interval in the corresponding spring study. These taxa were excluded in developing the occurrence matrix for the spring of 2001.

of 2001 and the summer of 2002 ($K_{obs} = 0.53$; $P = 0.007$).

Based on our strict definition, none of the successional patterns in Fig. 5 seem to contain periods of taxonomic stasis. However, when we applied the definition loosely to consider pairwise similarities to be equal when their confidence intervals overlapped, the successional patterns in Fig. 5 contained several periods of taxonomic stasis, e.g., between sampling

intervals 1–4, 11–13, and 15–19 in the spring of 2001 (Fig. 5A) and between sampling intervals 3 and 6 in the summer of 2001 (Fig. 5B).

The number of taxa present during each sampling interval and the number of occurrences of each taxon during each of the study periods are presented graphically in Figs. 6 and 7, respectively. The mean number of taxa was 9.0 ± 4.6 and 11.2 ± 4.8 per interval for the spring of 2001 and 2002, respectively, and 9.3 ± 1.8 and

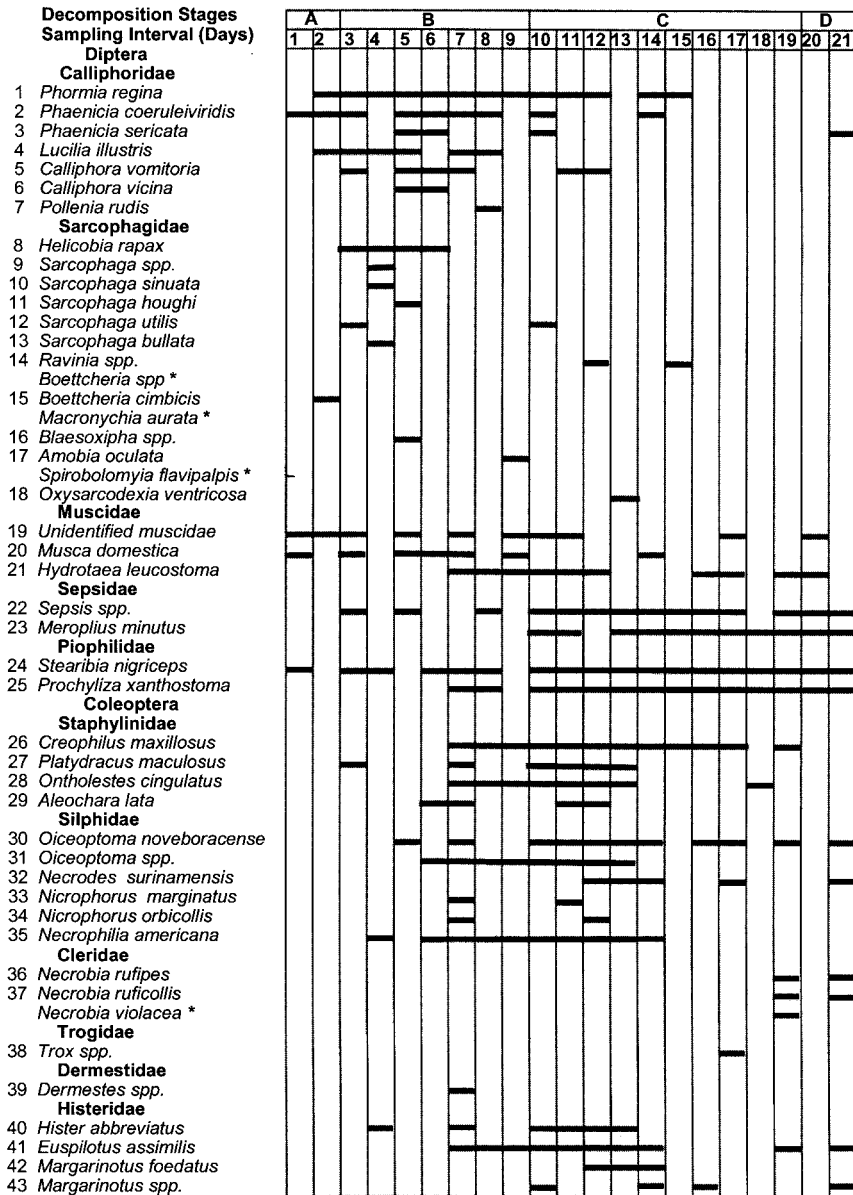


Fig. 3. Succession diagram for insect taxa on pig carcasses during 21 sampling intervals in the spring of 2002 in Blacksburg, VA. Stages of decomposition are shown and are (A) fresh, (B) bloat, (C) active decay, and (D) advanced decay and dry. *Unnumbered (rare) taxa that were observed only in one sampling interval in the spring of 1 yr but not during any sampling interval in the corresponding spring study. These taxa were excluded in developing the occurrence matrix for the spring of 2002.

11.6 ± 4.3 per interval for the summer of 2001 and 2002, respectively. With respect to the number occurrences, 12 taxa occurred more frequently in the spring of 2001 than in 2002, but 19 taxa occurred more frequently in the spring of 2002. The remaining taxa occurred with equal frequency in both years (Fig. 7, A and C). Likewise, five taxa occurred more frequently in the summer of 2001 than in the summer of 2002, but 18 taxa occurred more frequently in the summer of 2002

(Fig. 7, B and D). In addition, the number of occurrences of individual taxa was generally higher in 2002. The mean number of occurrences was 4.4 ± 4.2 and 5.5 ± 4.5 per taxon in the spring of 2001 and 2002, respectively, and 2.6 ± 1.2 and 3.2 ± 1.6 per taxon in the summer of 2001 and 2002, respectively.

Mean (range) ambient daytime temperatures for the spring and summer of 2001 and 2002 were 18.3 (1.8–28.0), 17.7 (5.5–28.3), 22.6 (13.2–29.1), and

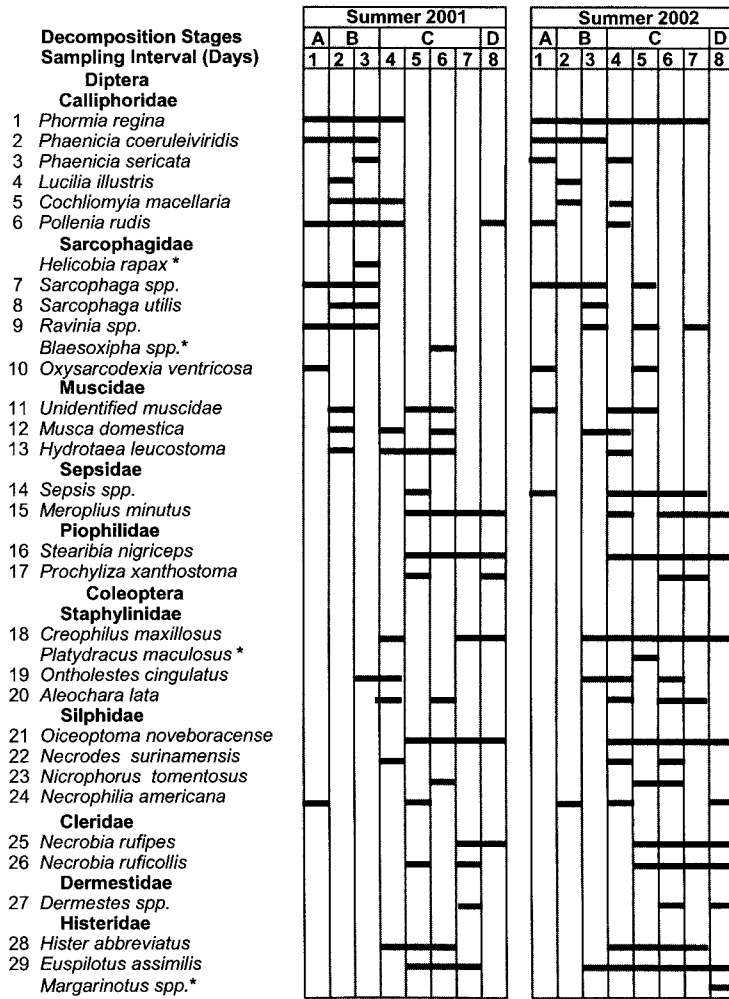


Fig. 4. Succession diagrams for insect taxa on pig carcasses during eight sampling intervals in the summer of 2001 and 2002 in Blacksburg, VA. Stages of decomposition are shown and are (A) fresh, (B) bloat, (C) active decay, and (D) advanced decay and dry. *Unnumbered (rare) taxa that were observed only in one sampling interval in the summer of 1 yr but not during any sampling interval in the corresponding summer study. These taxa were excluded in developing the occurrence matrices for the summer of 2001 and 2002.

24.5°C (15.9–31.7°C), respectively. Mean maggot mass temperatures for the same periods were 32.0, 36.0, 37.0, and 40.0°C, respectively. Total rainfall during the study periods was 8.2, 6.7, 2.3, and 4.2 cm for the spring of 2001 and 2002 and the summer of 2001 and 2002, respectively.

Discussion

Arthropods that visit a corpse can be classified as necrophagous species (i.e., those that feed on the corpse); predators and parasites of necrophagous species; omnivorous species (e.g., wasps, ants, and some beetles); or adventive species, such as spiders and centipedes, that exploit the corpse as habitat. This study reports on the necrophagous and predatory/parasitic insect species observed on decomposing pig

carcasses in southwest Virginia during the first 21 d in the spring and 8 d in the summer after placement in the field (Figs. 2–4).

Taxonomic composition in the spring study was greater than in the summer studies (47 versus 33 taxa). The number of taxa reported in similar studies on vertebrate carcasses was between 54 and 522. These included studies that reported 217 species in Tennessee (Reed 1958), 522 species in South Carolina (Payne 1965), ≈101 species in Hawaii (Richards and Goff 1997), and 54 species in Brazil (Carvalho et al. 2000). The large range of taxa found in these studies can be attributed to differences in climate, sampling frequency, number of animal models that were used, and the reporting of arthropods other than insects. These and other factors are known to affect insect successional patterns on carrion (Anderson 2001).

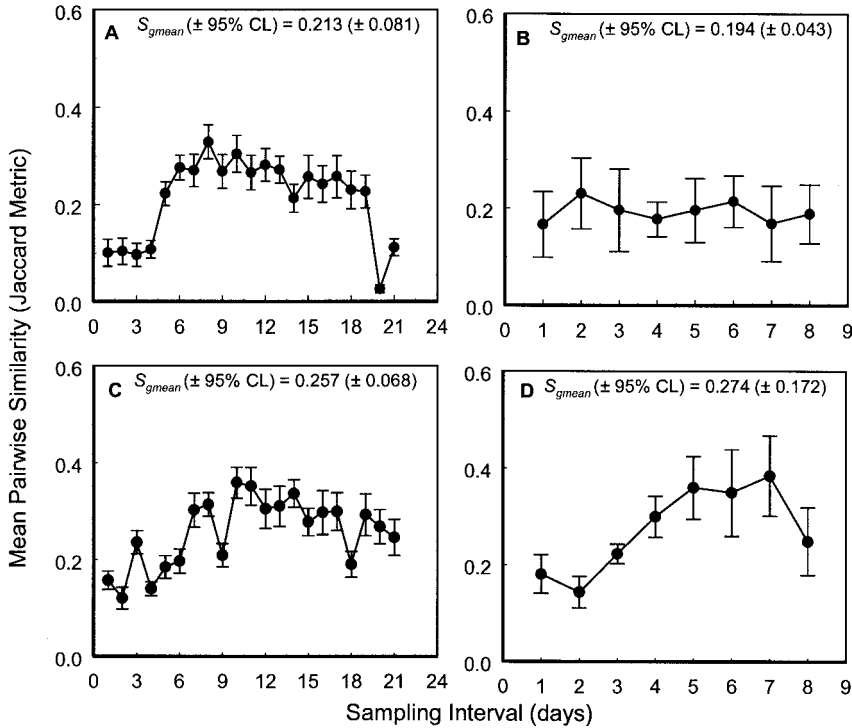


Fig. 5. Jaccard similarities for the occurrence patterns of insect taxa on pig carcasses in Blacksburg, VA. Mean and SE (vertical bars) of the Jaccard coefficient are shown for the similarity between each interval and all other intervals. The plots are spring 2001 (A), summer 2001 (B), spring 2002 (C), and summer 2002 (D). Overall Jaccard coefficient in insect faunal succession ($\hat{S}_{gmean} \pm 95\% CL$) for each period is given. Observed correlation (K_{obs}) and significance value for the successional patterns of insects in the spring (A and C) are 0.57 and $P = 0.001$, respectively; K_{obs} and the significance value for the summer patterns (B and D) are 0.53 and $P = 0.007$, respectively.

Not surprisingly, the earliest visitors to the carcasses in all of the studies were dipterans in the families Calliphoridae, Sarcophagidae, and Muscidae (Hall 2001). *P. regina* was the dominant calliphorid species in the spring but was co-dominant with *P. coerulei-*

viridis in the summer. However, the presence of *P. regina* in the summer was somewhat surprising given that this species tends to favor cool weather conditions (Byrd and Castner 2001). Although *C. vicina* favors urban habitats, whereas *C. vomitoria* is common in

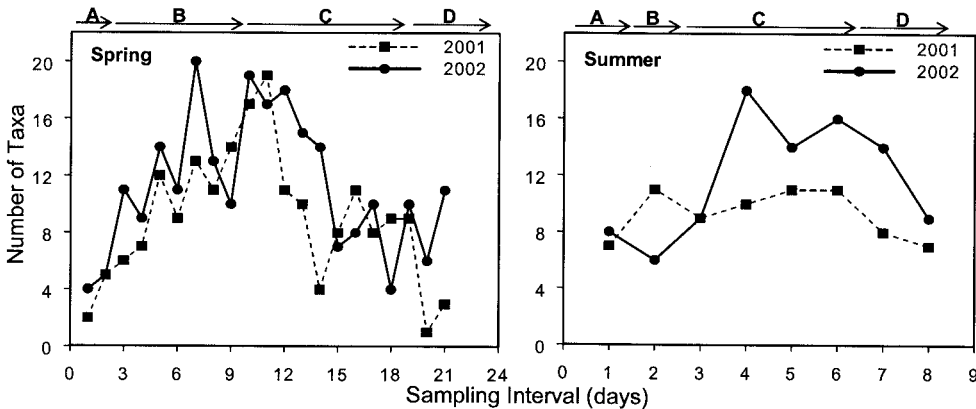


Fig. 6. Number of taxa during each sampling interval during the succession of insects on pig carcasses in the spring and summer of 2001 and 2002 in Blacksburg, VA. Graphs were developed from succession diagrams after eliminating rare taxa. Stages of decomposition are shown above top panel and are (A) fresh, (B) bloat, (C) active decay, and (d) advanced decay and dry.

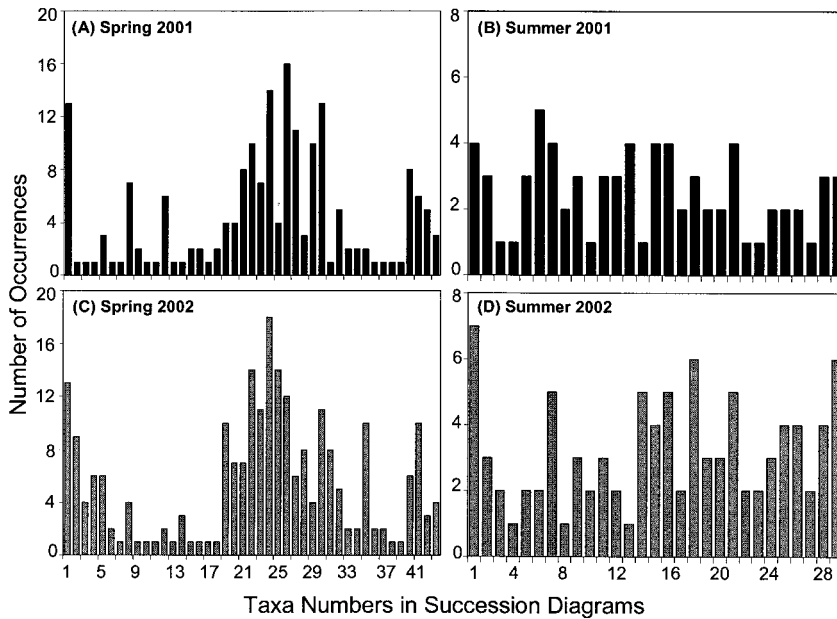


Fig. 7. Number of occurrences for each taxon during the succession of insects on pig carcasses in the spring and summer of 2001 and 2002 in Blacksburg, VA. Graphs were developed from succession diagrams after eliminating rare taxa. Each number on the x-axis represents the number assigned to a taxon in the succession diagram.

wooded rural and suburban areas, their biology and preference for shaded areas are similar, and thus, they were expected to occur together (Byrd and Castner 2001). The presence of *C. macellaria* only during the summer was expected because this species is known to prefer warm humid climates (Byrd and Castner 2001).

The two piophilid species (*S. nigriceps* and *P. xanthostoma*) were collected as adults, but not as larvae. Most species of Piophilidae are scavengers; adults often frequent decaying carcasses in the early stages of decay, but do not colonize until the carcass is in advanced decay, the preferred stage for egg deposition and larval development (McAlpine 1977).

Hall (2001) analyzed succession data on arthropods on human cadavers from Rodriguez and Bass (1983) and showed that beetles are present mainly during the bloat and decay stages of decomposition. In our study on pig carcasses, beetles appeared as early as the third day in the spring studies and on the first day in the summer studies. However, the majority of beetles were observed during the mid- to late-successional periods (Figs. 2–4), which corresponded to the bloat and decay stages of decomposition.

Knoxville, TN, is probably the closest in location and climate to southwest Virginia where another study of carrion-arthropod succession on an animal model was conducted (Reed 1958). In that study, Reed (1958) reported on 217 insect species, many of which were also observed in our study, despite the fewer number of species. Several factors might account for the difference between the two studies. Reed (1958) used a greater number of carcasses (45 dog carcasses versus 8 pig carcasses in this study) and sampled in

nine different areas (versus one in this study) throughout an entire year in contrast to two seasons during this study. In addition, Reed (1958) reported incidental, omnivorous, and adventive species, as well as necrophagous and predators of necrophagous species.

Rodriguez and Bass (1983) noted that there is a direct correlation between the rate of decay and the succession of insect families and species arriving at a cadaver. In general, the succession patterns in our study were similar to those described by Reed (1958) and Rodriguez and Bass (1983), with minor differences in the time at which certain species begin frequenting the carrion. These differences might be explained by the different rates of decomposition exhibited by dogs (Reed 1958), humans (Rodriguez and Bass 1983), and pigs. In general, the domestic pig (~23 kg carcass) is the most accepted human model in succession studies because it approximates the pattern of human decomposition more closely than other animal models (Catts and Goff 1992). As such, it has been shown that the patterns of arthropod succession do not differ significantly between pig carcasses with moderate differences in size (Hewadikaram and Goff 1991). However, one study showed that extreme size differences can greatly affect the rate of decomposition (Kuusela and Hanski 1982). Although the pigs used in the spring studies were larger than those in the summer studies, the pigs within each season were of approximately the same size. This allowed us to make statistical comparisons between the succession patterns of insect taxa within seasons. We did not make statistical comparisons of the succession patterns of

insect taxa between spring and summer studies, so differences in carcass size between seasons are not relevant to this study.

An occurrence matrix was developed for each of the succession diagrams in Figs. 2–4. We made the size (number of taxa and number of sampling intervals) of the occurrence matrices for corresponding study periods equal by eliminating “rare” taxa from the successional diagrams. Rare taxa were considered to be those that appeared in one sampling interval during the study period in 1 yr but were absent during the corresponding period in the other year. As such, three dipterans, *Boettcheria* Parker, *Macronychia aurata* Coquillett, and *Spirobolomyia flavipalpis* Aldrich (Sarcophagidae), and one beetle, *Necrobia violacea* L. (Cleridae), were eliminated from the spring observations. Similarly, two dipterans, *H. rapax* (Walker) and *Blaesoxipha* Loew (Sarcophagidae), and two beetles, *Platydacus maculosus* Gravenhorst (Staphylinidae) and *Margarinotus foedatus* LeConte (Histeridae), were eliminated from the summer observations. It is likely that these species naturally occur less frequently in southwest Virginia than other species in the same families.

The values of jackknife estimates in overall similarity were low (Fig. 5) but are within the range of values (0.20–0.50) that have been derived from data on the successional patterns of carrion-arthropods in other studies (e.g., Schoenly 1992). The low values are indicative of the rapid turnover in species composition, which is to be expected for carrion arthropods. As is common for successional patterns of carrion arthropods (see Schoenly 1992), pairwise similarities in taxa are low at the start of decomposition (fresh stage), but increase to a maximum during mid-decomposition (bloating and active decay), and decrease toward the end of decomposition (advanced decay and dry stages). The attractiveness of a carcass to specific taxa changes during decomposition and this results in a typical horseshoe-shaped arch pattern of succession (Schoenly 1992). During the bloat and active decay stages of decomposition, the carcass is more attractive to a wider range of taxa than in the fresh and dry stages (Hall 2001, Watson and Carlton 2003). Therefore, there is usually greater taxonomic richness in the mid-successional period, which results in higher pairwise similarities in taxa between samples and the horseshoe-shaped arch pattern. This pattern is evident in Fig. 5, although it is much better defined in Fig. 6 for each of the study periods.

We found some evidence of taxonomic stasis in the successional patterns for the studies in 2001 (Fig. 5, A and B). In general, only periods in which pairwise similarities are equal for more than two consecutive sampling were considered taxonomic stasis. Therefore, although there are instances in the spring (Fig. 5C) and summer of 2002 (Fig. 5D) where pairwise similarities were equal (by our loose definition) for two consecutive sampling intervals, we did not consider these as periods of taxonomic stasis. The lack of evidence for taxonomic stasis in the spring and summer of 2002 was likely because of the greater

variability in the successional patterns during these periods of the study compared with similar periods in 2001. From the data presented graphically in Fig. 6, we saw that the mean number of taxa among sampling intervals in the spring was greater in 2002 than in 2001 (11.2 ± 4.8 versus 9.0 ± 4.6 taxa per interval). Similarly, the number of species among sampling intervals in the summer studies was greater in 2002 than in 2001 (11.6 ± 4.3 versus 9.3 ± 1.8 taxa per interval). The number of occurrences of each taxon during the study periods was also greater in 2002 compared with the corresponding periods in 2001 (Fig. 7). The greater variability in the successional patterns of insect taxa in 2002 was likely because of consistently higher temperatures during those study periods compared with similar periods in 2001.

Despite differences in the number of instances of taxonomic stasis, numbers of taxa occurring in each sampling interval, and number of occurrences of these taxa, the overlapping confidence intervals in overall Jaccard similarities for corresponding periods suggest that the patterns of insect succession between these periods might be similar. This was confirmed by the permutation analysis, which detected no differences between similarity matrices developed from the occurrences matrices for the spring of 2001 and 2002 and for the summer of 2001 and 2002. As such, the successional patterns of insects in the corresponding periods were similar. This suggests that the successional patterns of the insects that we observed during the spring and summer studies are typical for the respective periods and that variations in these patterns from year to year should be minimal.

Estimation of PMI from succession data requires knowledge of local carrion fauna and their successional patterns at different times of the year (Anderson 2001, Wells and Lamotte 2001). Our data show that such patterns can remain similar from year to year and thus can be used to provide the basis for PMI estimates. The successional patterns of insect species can differ at different seasonal periods of investigation even at the same site. These differences highlight the need for further studies of carrion-arthropod succession at different times of the year and in different regions. This study represents the first step toward developing a knowledge base of the successional patterns of carrion-arthropods for the southwest Virginia area.

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