CENTRO DE INVESTIGACIÓN Y DE ESTUDIOS AVANZADOS DEL INSTITUTO POLITÉCNICO NACIONAL

UNIDAD IRAPUATO

"Arthropod diversity associated with decaying *Opuntia* spp. and *Citrus sinensis* fruits"

Tesis que presenta

B.E. Alejandra Castañeda González

Para obtener el grado de

Maestra en Ciencias

En la especialidad de

Biotecnología de Plantas

Directora de Tesis: Dra. Therese Ann Markow

Irapuato, Guanajuato Octubre, 2018

El presente trabajo se llevó a cabo en el Laboratorio de Genómica de Biodiversificación en la Unidad de Genómica Avanzada del CINVESTAV Unidad Irapuato, bajo la dirección de la Dra. Therese Ann Markow.

El presente trabajo se realizó con el financiamiento brindado por el Consejo Nacional de Ciencia y Tecnología (CONACYT) a través de la beca otorgada al becario 616061 y con el programa de Ciencia Básica (donativo CB-180385 otorgado a la Dra. Therese Ann Markow), con fondos de la UGA-LANGEBIO y de Sin Fronteras: Creativity without Borders.

A mi familia.

Agradecimientos

Agradezco enormemente a Teri por permitirme trabajar en su laboratorio. Gracias por apoyarme en todos los sentidos de mi vida académica. Gracias por ayudarme a mejorar mi manera de hablar, de escribir y de ver la biología. Por los viajes de colecta y a lugares desconocidos, por las largas pláticas, el hospedaje y las enseñanzas. También a cina por cuidarme.

También a mi comité, el Dr. Martin Heil y la Dra. June Simpson. Gracias por sus valiosas preguntas y por el tiempo que dedicaron para concluir este trabajo.

A CONACYT por la beca otorgada.

Al Dr. Edward Pfeiler quien amablemente me ayudó con los análisis de genética de poblaciones y me compartió su pasión por los insectos. Al Dr. Andrew Cline quien me ayudó con la identificación de las especies de nitidúlidos. Al Dr. José Luis Navarrete quien me ayudó a identificar algunas especies de estafilínidos y me ofreció una larga e interesante charla sobre escarabajos.

A mis compañeros de laboratorio. Gracias a todos por su valiosa amistad: Eunice, Fernán, Javi, Cynthia, Pablo Luis y obviamente a Néstor por sus agradables e interesantes pláticas. A Ale Pérez por compartirme su conocimiento de los “mites”. A todos los que me recibieron muy bien en el grupo, aunque me tocó despedirlos: Mariana, Kari, Ale Chávez y Daniel.

A mis amigos de generación, los “empáticos volátiles”, con quienes pasé las mejores horas de la maestría: Rogelio, Jaime, Aurora, Itzel, Anibal y Mane. Gracias por las fiestas, las horas de estudio, las pláticas y los viajes. A mis demás amigos y compañeros del CINVESTAV, incluyendo a los del CECIG (el consejo estudiantil).

Sobre todo, gracias a mi familia que, aunque no conocen mucho de lo que hago, siempre están dispuestos a escucharme y apoyarme. Gracias papá, Rogelio, porque sin ti no hubiera podido hacer lo que estoy haciendo ahora. También a ti mamá, Mayeli, por ser la mejor mamá de todas, por consentirme con tu comida, tus apapachos y tu manera de ser tan divertida. Gracias Dani por ser mi mejor amiga y la mejor hermana por siempre, obvio por escucharme cuando lo necesito y darme palabras de aliento. A mis hermanos Lorena y a Hugo por cuidarme desde que nací hasta el día de hoy y por hacerme tía de mis hermosas sobrinas Elisa y Silvana.

Contents

Table index

Chapter 1: Arthropod diversity associated with decaying *Opuntia* spp. and *Citrus sinensis* fruits

Chapter 2: Phoretic mites on Nitidulidae and Staphylinidae beetles on rotten fruits

Supplementary material

Figure index..

Chapter 1: Arthropod diversity associated with decaying *Opuntia* spp. and *Citrus sinensis* fruits

Chapter 2: Phoretic mites on Nitidulidae and Staphylinidae beetles on rotten fruits

Resumen

Artrópoda es el filo más exitoso y diverso conocido. Constituyen más del 80% de la diversidad del reino animal con 1.3 millones de especies descritas. Las estimaciones globales de biodiversidad son difíciles porque los artrópodos existen en todo el mundo en una amplia gama de ecosistemas y nichos. Las plantas en descomposición son nichos limitados para una variedad de organismos, incluidos los artrópodos, que los utilizan tanto para la alimentación como para la reproducción. Los artrópodos desempeñan un papel importante en la descomposición de los tejidos vegetales y, por lo tanto, en el reciclaje de nutrientes. A pesar de su importancia, solo hay un número limitado de estudios sobre la diversidad de artrópodos en plantas descompuestas. Sin embargo, las comunidades de artrópodos en frutos podridos están poco exploradas. En el presente trabajo estudié los artrópodos que utilizan frutas en descomposición y pregunté cómo el tipo de fruta y la ubicación geográfica afectan la diversidad. Para ello, colecté frutos de *Opuntia* spp. (tunas) y de naranjas Valencia (*Citrus sinensis*) de localidades en Guanajuato y Sonora en México. Descubrí que la ubicación geográfica, más que el tipo de fruta influyó en la composición de la comunidad de artrópodos. Las comunidades de artrópodos son muy diversas y la cantidad de especies esperadas es elevada. Coleoptera, Diptera, Hymenoptera y Acari fueron los órdenes más representativos, pero Nitidulidae, que es una familia de coleópteros distribuidos en todo el mundo, fue el taxón más diverso encontrado. Además, la composición de los artrópodos cambia durante la descomposición de la fruta: las especies saprófagas llegan primero mientras que los depredadores o parasitoides aparecen más tarde en los frutos. Además, exploré la diversidad genética de siete escarabajos nitidúlidos mediante el uso del gen mitocondrial CO1, descubriendo que la mayoría de ellos son genéticamente diversos. Del mismo modo, analicé la diferenciación geográfica en cuatro especies de nitidúlidos, de los cuales solo uno mostró diferenciación poblacional, posiblemente debido a una dispersión limitada o adaptación local. Las frutas en descomposición proporcionaron un modelo excelente para estudiar la diversidad de artrópodos asociados con la descomposición de las plantas y su potente importancia en el funcionamiento del ecosistema y el reciclaje de nutrientes.

Abstract

Arthropoda is the most successful and diverse phylum known. They constitute over 80% of the animal kingdom diversity with 1.3 million described species. Global biodiversity estimates are difficult because arthropods exist worldwide in a broad range of ecosystems and niches. Decaying plants are limited niches for a variety of organisms, including arthropods, which use them for both feeding and breeding. Arthropods play important roles in plant tissue decomposition and thus nutrient recycling. Despite their importance, there is only a limited number of studies on the arthropod diversity in necrotic plants. Nevertheless, arthropod communities in necrotic fruits are poorly explored. Here, I studied the arthropods that utilize decaying fruits and asked how the fruit type and geographic location affect the diversity. I sampled several rotten prickly pears from *Opuntia* spp. and from Valencia oranges (*Citrus sinensis*) from localities in Guanajuato and Sonora in Mexico. I found that geographic location, more than fruit type, influenced arthropod community composition. Arthropod communities are greatly diverse, and the number of expected species is elevated. Coleoptera, Diptera, Hymenoptera and Acari were the most representative orders, but Nitidulidae, which is a worldwide distributed coleopteran family, was the most diverse taxa found. Furthermore, arthropod composition changes during fruit decomposition: saprophagous species arrive first while predators or parasitoids appear later on the fruits. Additionally, I explored the genetic diversity of seven nitidulid beetles by using the mitochondrial CO1 gene, finding that most of them are genetically diverse. Likewise, I test for geographic differentiation in four nitidulid species, of which only one showed population differentiation, possibly because limited dispersion or local adaptation. Decaying fruits provided an excellent model to study the diversity of arthropods associated with plant decomposition and their potential importance in ecosystem functioning and nutrient recycling.

Chapter 1: Arthropod diversity associated with decaying *Opuntia* spp. and *Citrus sinensis* fruits

Introduction

Arthropod diversity

Arthropoda is the most successful and diverse animal phylum known. They appeared more than 500 million years ago and still evolving in all shapes and sizes. They constitute over 80% of the animal kingdom diversity with approximately 1.3 million described species (Zhang, 2013), while approximately 6.8 million total arthropods species are estimated to exist (Stork, Mcbroom, Gely, & Hamilton, 2015). Arthropods represent the dominant eukaryotic taxon on the planet and exhibit a wide variety of life styles. They play important roles in the regulation of ecosystems, including serving as predators, parasites, prey, pollinators and seed dispersers, disease vectors and decomposers (Kim, 1993). Decomposition of dead animals and plants is one of the most important processes for ecosystem maintenance (Swift et al. 1979) and nutrient recycling (Reichle, 1977). It is the process by which decomposer organisms break down complex organic molecules in smaller ones to utilize them as energy source. Once broken down, nutrients can be returned to the biosphere or utilized by other organisms (Reichle, 1977). Elimination of microarthropods from the litter layer reduces the rate of mass loss of organic material in semiarid ecosystems (Whitford & Parker, 1989). In addition, herbivorous insect frass increases the availability of nitrogen and carbon in the soils where the insects feed (Frost & Hunter, 2004).

The enormity of arthropod diversity renders it impossible to study at the global scale. Discrete systems are more manageable if we want to explore the diversity of a particular taxon and speculate about functional components in the community. Decaying plants,

because they are discrete units, provide highly tractable models to explore the diversity and ecology of arthropods.

Arthropods and plant decomposition

When a plant decays, the tissue initially is colonized by microbial decomposers, such as bacteria or fungi, that rapidly metabolize it, leaving a resource-rich microhabitat for many organisms, including arthropods, which consume, inhabit or breed in the rotting host. The decaying tissue eventually is completely decomposed, and nutrients return to the environment. Scavengers facilitate disintegration by exposing interior matter to the outside while decomposers, which more often are microorganisms, accelerate nutrient releasing by breaking down molecules into molecular level.

Despite their importance, few studies of arthropods associated with decaying plant tissues exist. Four well-characterized examples are the studies conducted in rotting cactus from the Sonoran Desert. Castrezana and Markow (2001) investigated the arthropod community inhabiting necroses of three columnar cacti species: cardón (*Pachycereus pringlei*), organ-pipe (*Stenocereus thurberi*), and senita (*Lophocereus schottii*) in central Sonora, Mexico. They found arthropods in two classes (Arachnida and Insecta), 10 orders, 23 families and 34 species, most of them belonging to the Diptera, Coleoptera and Acari. Cardón cacti had the most diverse arthropod community. Subsequently, Richmond et al. (*in revision*) replicated the Castrezana and Markow study, but in Baja California Sur, where the same species of cacti are found. Of the arthropod specimens they found in those columnar cacti, species diversity again was highest in cardón. In a smaller study, focused on the Coleoptera, Ferro et al. (2013) characterized the species in decaying fishhook barrel cacti (*Ferrocactus wislizeni*) in the Sonoran Desert. In 16 different cactus samples, they discovered 976 specimens, sorted into 11 families and 35 species. Recently, Delgado-Fernández et al. (2017) studied all animal interactions with cardón in Baja California, finding 27 different insect species and two arachnids, where 35% feed on necrotic cacti. These reports provide exceptional background concerning the wide variety of arthropods that can live in necrotic cactus arms and stems.

The most well-studied insect taxon in necrotic cactus niche is the genus *Drosophila*. Adult and larvae flies feed and breed in cactus to complete their life cycle. Four- species of cactophilic Drosophila endemic to the Sonoran Desert, *D. movajensis, D. pachea*, *D. arizonae*, and *D. nigrospiracula*, are each associated with a specific cactus host: organ-pipe (*Senocereus thurberi*), senita (*Lophocereus schottii*), cina (*Stenocereus alamosensis*), and cardón (*Pachycereus pringlei),* respectively (Fellows & Heed, 1972). The unusual *D. mettleri* has specialized on soil saturated by the putrid juice exuded from the necrosed cardón (Fogleman & Danielson, 2001). The above cacti produce several secondary compounds that are principally related to host specialization (Fogleman & Danielson, 2001; Ruiz & Heed, 1988). Senita contains high alkaloid levels, which are toxic for a variety of insects. *Drosophila pachea*, however, is adapted to feed and breed on the cacti (Fellows & Heed, 1972). Other specific arthropod taxa associated with necrotic columnar cacti also have been studied, but to a lesser degree. Several Coleopteran species, primarily of the family Histeridae (genus *Carcinops, Hololepta,* and *Iliotona*), are common colonizers of the necrotic cacti and may also exhibit some degree of host specialization (Ferro et al., 2013; Pfeiler & Markow, 2011; Reese & Swanson, 2017). Similarly, Staphylinidae and Tenebrionidae families have been found as common cacti arthropod fauna (Ferro et al., 2013; Mejía, 2016). Additionally, the pseudoscorpion *Dinocheirus arizonensis, a* predator of a variety of insects, including the cactophilic *Drosophila* also has been found in the necrotic cacti niche. This pseudoscorpion disperses by phoresy, using as principal disperser another cactus fly, *Odontoloxozus longicornis*. Both are commonly found in the Sonoran Desert in a special interaction (Pfeiler, Bitler, Castrezana, Matzkin, & Markow, 2009; Pfeiler & Markow, 2011).

Decaying fruit model

In angiosperms, the fruits have evolved to attract animals, which eat them and also disperse their seeds to new habits (Wright, 2015). When a fruit is not consumed by animals, it become decomposed by senescence, by yeast and bacteria pathogens, or by physical injury by herbivores which introduce new microorganisms.

Many factors could determine which species will occur in a decomposed fruit. The abundance and dominance of species in rotten environments can be influenced by temporal, spatial and biological dynamics. In an experiment conducted in pig carcass, the abundance and dominance of beetles from different trophic roles changed with season and decomposition stage (Zanetti, Visciarelli, & Centeno, 2015). Moreover, the ecologically similar pair species of fruit flies *D. melanogaster*/*D. simulans* and *D. immigrans*/*D. hydei* coexist because they arrive at different times during the decomposition of the fruit, giving rise to a pattern of colonization (Nunney, 1990). The occurrence of species also could be a function of their geographic location. The fungal communities of necrotic cladodes from Caribbean and Australia localities are very similar in composition, more than plant tissue type (Starmer, Lachance, & Phaff, 1987).

Rotten fruits offer a mixture of odorous compounds, products of metabolic activities of microorganisms that colonize them. Numerous arthropods are associated with decaying fruits, having been attracted mainly by the product of fermentation of those microorganisms (Wright, 2015). For example, *D. melanogaster*, showed stronger attraction, oviposition rate and larval development when tested with baker's yeast *Saccharomyces cerevisiae* over yeast-free fruit substrate (Becher et al., 2012). Furthermore, several natural yeast strains have been tested with organ-pipe cactus of the four geographically isolated subpopulations of *D. mojavensis*. Flies showed different patterns of preference, some of which were specific for their local hosts, revealing the importance of microorganisms in host use (Date, Crowley-Gall, Diefendorf, & Rollmann, 2017). Coleopterans also are highly attracted to necrotic fruits. Sap beetles of the worldwide-distributed family Nitidulidae (Cucujoidea) are commonly found in rotten fruits (Blackmer & Phelan, 1995; Mutinelli, Federico, Carlin, Montarsi, & Audisio, 2016). Similar to D*rosophila*, Blackmer and Phelan (1995) found that *Carpophilus lugubris* and *C. hemipterus* (Nitidulidae: Carpophilinae) are more attracted to the substrate when it is inoculated with yeast. In a study of *C. hemipterus*, there was no consistent preference for fig fruits, yeasts or the combination when was tested (Miller & Mrak, 1953) and still unclear is whether nitidulids have a preference for feeding on the fruit or primarily upon the yeast.

While the arthropod communities living in necrotic cactus tissue have been rather well-studied, the diversity of those utilizing decaying cactus fruits remain poorly investigated. Mutinelli et al. (2016) explored the arthropods associated with rotten citrus and kiwi fruits in orchards in a region of Italy. Only the oranges had insects wherein eight Nitidulidae, a couple of Staphylinidae species, both larvae and adults of *Drosophila* spp., *Musca domestica*, a Mycetophilidae and one Oniscoidea were present. Indeed, investigations of the arthropod communities in other decaying fruits, such as apples, peaches or pears, also are lacking. Rotting fruits are usually found on the ground and thus can rapidly return many nutrients to the soil. This make them especially attractive to a variety of arthropods because are they provide rich ephemeral, temporal and discrete microhabits. Moreover, the fruits of different plants differ chemically raising the question as to whether particular arthropods are specialized with respect to their fruit hosts, and if so, what is the basis of their specialization. The broad distributions of particular fruits allow us to ask how the host type as well as its the geographic location impact arthropod composition and species richness. Finally, necrotic fruits, because they are discrete units, can contain the full range of diversity occurring within them, allowing us to address questions about their ecology and biodiversity.

In my study, I compared the arthropods associated with two fruits similar in size: rotting prickly pears from *Opuntia* cactus species and fruits of the Valencia orange (*Citrus sinensis*). I asked the following questions:

- What is the diversity of arthropods associated with decaying *Opuntia* and *Citrus* fruits?
- What are the influences of the geographic location versus the fruit type in arthropod diversity?
- How does arthropod composition change during the decomposition process in decaying fruits?
- Is there evidence for geographic differentiation between populations of a given species?

Hypothesis

Necrotic fruits similar in size and from different localities are different in their arthropod communities.

Prediction

- Species richness and diversity will differ between necrotic fruit species.
- Arthropods in decaying fruits are changing over decomposition process.
- Arthropod species will exhibit some degree of geographic differentiation.

Main objective

My primary objective was to characterize the diversity of arthropods associated with two necrotic fruits: prickly pears (*Opuntia spp*.) and Valencia oranges (*Citrus sinensis*).

Specific aims

1. Estimate and compare the diversity of arthropods associated with necrotic *Opuntia* spp. and *Citrus sinensis* fruits from different localities.
2. Investigate the composition of arthropods in necrotic fruits during the decomposition process.
3. Explore whether geographic differentiation exists between populations of a given group of arthropods in the necrotic fruits.

Methods

Field collection

Sampling sites. Arthropods associated with necrotic fruits were collected from two different plant species: Valencia oranges (*Citrus sinensis*) and red prickly pears (*Opuntia ficus-indica, O. streptacantha, O. megacantha* and *O. robusta*) (Table S1). In September 2016, prickly pears were sampled in three localities in the state of Guanajuato: Irapuato, Guanajuato, and San Miguel de Allende (SMA) (N = 57). I collected again in September and October 2017 in Irapuato and SMA (N = 69). Rotten oranges were sampled from April to June and from October to December 2017 in Guanajuato and Irapuato (N = 38) and, ultimately, in August 2018 (N = 10) from private gardens where no pesticides are normally used. In Sonora, during the spring of 2017, October 2017 and March 2018, decaying oranges were collected at a free-pesticide orchard in Navojoa which is in the Sonoran Desert (N = 134) and prickly pears were sampled in October 2017 and August 2018 in Batacosa (N = 33). Finally, I had the opportunity to make some smaller collections of *Opuntia* fruits in Lagos de Moreno (N = 13) and oranges in Guadalajara (unknow number of samples) in the state of Jalisco (Figure 1). Specimens from Jalisco were counted in the general arthropod data-base but were too limited in number to consider in further comparative analyses. Collections thus were placed into four arthropod assemblages that were separated by state and fruit species, including only Guanajuato and Sonora states.

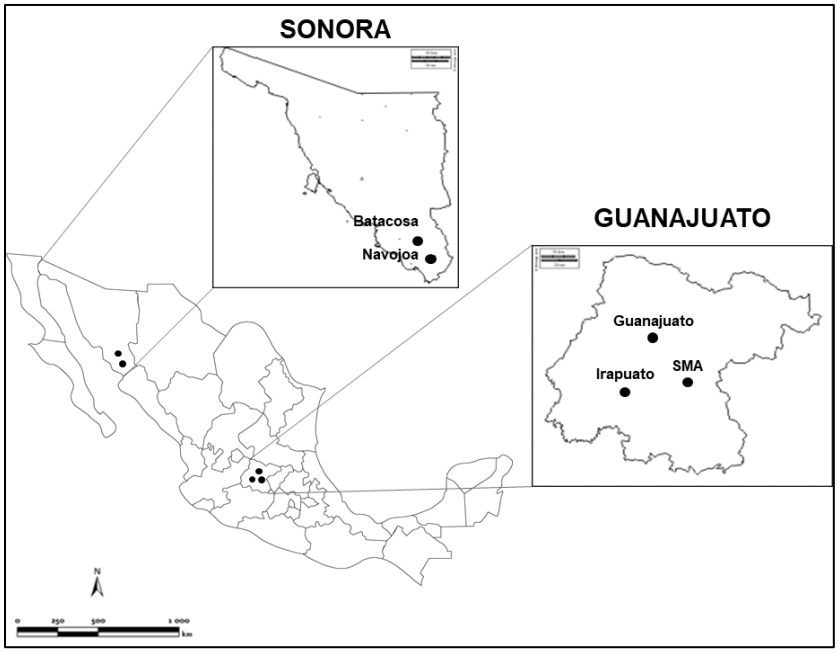


Figure 1. Sampling sites. Rotten prickly pears were collected in the state of Guanajuato (localities: Guanajuato, Irapuato, and San Miguel de Allende (SMA)) and Sonora (Batacosa). Oranges were collected in the states of Guanajuato (localities: Guanajuato and Irapuato) and Sonora (Navojoa).

Arthropod collection. Individual fruits from each locality were directly collected from the ground, saved in plastic containers and visually examined in the laboratory. Adult arthropods were collected from fruits both with forceps and an aspirator. All adults and some larvae were saved in vials with and 95% ethanol for DNA preservation at -20 °C. Specimens were sorted to species or morphospecies within a genus, family or order and counted (see below). Additionally, decomposed fruits were classified in five decomposition stages, based on the classification employed by carrion literature (Zanetti et al., 2015), a system that depends on texture, water content and color: fresh (F), early decay (E), active decay (Ac), advanced decay (Ad), and remains (R)).

Identification of arthropod species

Morphological identification. I first classified adult arthropod specimens using morphological keys (Anderson, 2011 in http://bugguide.net/node/view/15740; Borrow & White, 1970; Ewing & Cline, 2005; Navarrete-Heredia, Newton, Thayer, Ashe, & Chandler, 2002) and assigned them to morphospecies. I identified to family, subfamily and genus when was possible. I spent more effort in the identification of coleopteran specimens as they were the most abundant. Morphological identification of Nitidulidae beetles was aided by Dr. Andrew R. Cline from Plant Pest Diagnostics Center in California, USA. Identification of some Staphylinidae specimens was supported by Dr. José Luis Navarrete Heredia from University Center of Biological and Agricultural Sciences in University of Guadalajara, México.

Molecular identification. Genomic DNA from Nitidulidae and Staphylinidae species was extracted with the DNeasy™ Blood & Tissue Kit (QIAGEN® Inc., Valencia, CA) using the whole individual due to their small size. I complemented the morphological identification by sequencing the 658-base pair barcode region of the cytochrome oxidase subunit I (CO1) mDNA gene, known as the "barcode region". I amplified the CO1 gene using the LCO1490f (5'-GGTCAACAAATCATAAAGATATTGG-3') and HCO2198r (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') (Folmer, Black, Hoeh, Lutz, & Vrijenhoek, 1994) applying the following PCR conditions: one cycle of denaturation at 94 °C for 3 min, 35 cycles of 94 °C for 30 s of denaturation, 52 °C for 1 min of annealing, and 72 °C for 1 min of extension, and a final extension of 7 min at 72 °C. Annealing conditions varied for some species in a range of 52 to 45 °C. Amplicons were purified with the QIAquick™ PCR Purification Kit (QIAGEN® Inc., Valencia, CA). PCR products were sequenced by Sanger technology at the LANGEBIO core DNA sequencing facility.

Arthropod diversity analysis

Species diversity. I used the latest version of the software EstimateS (Colwell & Elsensohn, 2014) to calculate the arthropod species richness and diversity estimators to compare between the fruit type and localities assemblages (Table 1). Classic Chao formula was used to estimate species richness. I performed a rarefaction curve to compare species richness and extrapolated the data to 150 samples using 100 replicates with the bootstrap method included in EstimateS. A pairwise compositional analysis was done to compare among fruit type and locality using the Chao-Sorensen (abundance-based) and Chao-Jaccard (abundance-based) similarity indices (Chao, Chazdon, Colwell, & Shen, 2005). A value of 1 means complete similarity while 0 means there are totally different.

Table 1. Indices used to estimate species richness and species diversity.

Index	Formula	Description
Chao1 (for abundance data)	$S_{obs} + \frac{F1^2}{2 * F2}$	Use abundance data of singletons and doubletons individuals present in a sample to estimate species richness.
Chao2 (for replicated incidence data)	$S_{obs} + \frac{Q1^2}{2 * Q2}$	Use incidence data (present or not) of unique or doubles species in a set of samples to estimate species richness.
Abundance-based coverage estimator (ACE)	$Sabun + \frac{Srare}{Crare} + \frac{f1}{Crare}\,\gamma^2 rare$	$Srare = \sum_{k=1}^{10} fk$ is the number of rare species in a sample (each with 10 or fewer individuals). $Sabun = \sum_{k=11}^{Sobs} fk$ is the number of abundant species in a sample (each with more than 10 individuals). $nrare = \sum_{k=1}^{10} kfk$ it is the total number of individuals in the rare species. $Crare = 1 - \frac{f1}{nrare}$ it is the proportion of all individuals in rare species that are not singletons. $\gamma^2 rare =$ it is the coefficient of variation.
Incidence-based coverage estimator (ICE)	$Sfreq + \frac{Sinfreq}{Cinfreq} + \frac{Q1}{Cinfreq}\,\gamma^2 infreq$	$Sinfreq = \sum_{k=1}^{10} qk$ it is the number of infrequent species in a sample (each with 10 or fewer individuals).

		$Sfreq = \sum_{k=11}^{Sobs} qk$ is the number of frequent species in a sample (each with more than 10 individuals). $ninfreq = \sum_{k=1}^{10} kqk$ it is the total number of incidences in the infrequent species. $Cinfreq = 1 - \frac{q1}{ninfreq}$ it is the proportion of all incidences of infrequent species that are not unique. $\gamma^2 infreq =$ it is the coefficient of variation.
Rarefaction curve		A statistical interpolation method of rarefying a reference sample by drawing random subsets of samples (or individuals) in order to standardize the comparison of samples.
Shannon index (H')	$\sum_{i=1}^{S} pi \ln pi$	Quantifies the uncertainty in the species identity of an individual randomly chosen in the sample.
Simpson index (D)	$1 - \sum_{i=1}^{S} pi^2$	Measure the probability that two individuals randomly selected from a sample belong to the same species.
Shannon evenness (J)	$\frac{\sum_{i=1}^{S} pi \ln pi}{\ln S}$	Measure of how similar species are in their abundances. High values indicate that are more similar.
Exp Shannon	exp(H')	Effective number of species: are transformed values of diversity estimations that are given in equivalent number of species.
Chao-Sorensen abundance based	$\frac{2U_{abd}V_{abd}}{U_{abd} + V_{abd}}$	It is the probability that two individuals from different samples randomly chosen to belong to the same species. It accounts for unseen species based on abundance data.
Chao-Jaccard abundance based	$\frac{U_{abd}V_{abd}}{U_{abd} + V_{abd} - U_{abd}V_{abd}}$	It is the probability that two individuals from different samples randomly chosen to belong to the same species. It accounts for unseen species based on abundance data.

S = total number of species; **pi** = relative abundance; **F1** and **F2** = singletons and doubletons individuals in a sample, respectively; **Q1** and **Q2** = unique and doubles species occurring in one or two samples, respectively. **U** and **V** = relative abundances of shared species in sample 1 and 2, respectively. Information from Chao et al., 2005; Gotelli & Chao, 2013; Magurran, 2004.

To understand the factors that influence their differences and the arthropod role in the necrotic fruit system, I classified the arthropods into five guilds: saprophagous (including

detritivores and mycephagous), predators, parasites, parasitoids and omnivorous, based upon field observations as well as an extensive literature search. A guild means a group of species that use similar resources in similar ways (Root, 1967), without the taxonomic effect. I accounted for incidence (presence or absence) for each group and compared among them.

Population genetics analysis of Nitidulidae beetles

Molecular analysis. Nitidulidae beetles were the most abundant and diverse group in both types of decaying fruits, thus it was possible to perform some population genetic analyses for these beetles. I hypothesized that species from different populations will exhibit geographic isolation. To address this hypothesis, we used the mitochondrial DNA fragment cytochrome oxidase subunit I (CO1) of seven species, which was amplified previously for molecular identification. CO1 sequences were visualized and aligned with ClustalO integrated in Seaview version 4 (Gouy, Guindon, & Gascuel, 2010). Diversity indices measurements and test of neutrality Tajima's D (Tajima, 1989) and Fu's *Fs* (Fu, 1997) were performed in DnaSP version 6.12.01 (Rozas et al., 2017). To compare individuals between populations and infer their genetic relationships, haplotype networks were constructed in Popart version 1.7.2 (Leigh & Bryant, 2015) using the implemented statistical parsimony method TCS (Clement, Posada, & Crandall, 2000). Hierarchical analysis of molecular variance (AMOVA) was done in Arlequin 3.5.2.2. (Excoffier, L. and Lischer, 2010), using the method Jukes & Cantor, to test for population structure among Sonora and Guanajuato populations.

Table 2. Indices and analysis used in the genetic population analysis of selected arthropods.

Index	Formula	Description
Nucleotide diversity (π)	$\sum_{i=1;\ j=1}^{n} pipj\ \pi ij$	The average proportion of nucleotides that differ between any randomly sampled pair of sequences.
Haplotype diversity (h)	$\frac{N}{N-1}\ (1-\sum_{i}^{n} x_i^2)$	An estimation of how many alleles are present. Probability that two randomly chosen alleles differ.

Tajima's D	$\frac{\pi - \theta}{\sqrt{V(\pi - \theta)}}$	Comparison of two estimators of the mutation parameter theta (θ = 2N, effective population size N) to distinguish between a DNA sequence evolving randomly (neutrality) and one evolving under directional selection.
Fu's *Fs*	$\frac{\theta_{S>1} - \theta_{S1}}{\sqrt{V(\theta_i - \theta_e)}}$	Evaluates the probability of observing a random neutral sample with a number of alleles similar or smaller than the observed value given the observed number of pairwise differences. It is based on the fact that singletons play a special role for different population histories. Uses the estimator θ_S.
AMOVA		Analysis of Molecular Variance is a method to estimate population differentiation explained by different population levels.
Fixation index (F_{ST})		Measures variation of allele frequencies between populations to explain the degree of differentiation between subpopulations.

pi, **pj** = frequency of sequence i or j; **πij** = proportion of nucleotides that differ between the sequences i or j; **N** = sample size; **xi** = haplotype frequency; **Θ** (theta) = number of segregating sites; S_i = number of segregating sites that affect i individuals in the sample. Information from Hamilton, 2009; Hedrick, 1999.

Results

1. Arthropod species diversity of fruits and general differences between fruit type and locality.

A total of 11,769 arthropod specimens, sorted in 117 designated species, were collected from 398 decaying fruits in this project. In total 27 genera were identified, 29 families, 14 orders and 4 classes (Table S2). Of the 126 rotting prickly pears from Guanajuato, 710 individuals were sorted into 61 species. In addition, 48 species from 3,335 individuals were found in 48 decaying oranges. From Sonora, 134 oranges were collected, and 7,316 specimens classified into 52 species were found. While, 77 prickly pears were sampled, 366 specimens were sorted in 23 species. From a small sampling effort, 8 species were found in 13 *Opuntia* fruits and 4 from rotten oranges in Jalisco (Table 3).

Table 3. Number taxa found in each biological organization level by fruit type and geographic location, number of specimens and number of samples.

Organization level	Prickly pears			Oranges			Total
	Guanajuato	Sonora	Jalisco	Guanajuato	Sonora	Jalisco	
Class	3	2	2	3	4	1	4
Order	12	7	4	9	11	3	14
Family	17	12	3	16	19	4	29
Genus	19	11	3	16	20	2	27
Species	61	23	8	48	52	4	117
Number of specimens	710	366	16	3335	7316	26	11769
Number of samples	126	77	13	48	134	n.d.	398

n.d. = no data

Four arthropod classes were found: Insecta, Arachnida, Collembola and Crustacea. Insects were the most abundant and varied in necrotic fruits, as expected, since they are the most diverse within the Arthropoda. Of the 117 morphospecies identified, 57 are coleopteran, of

which 26 belong to the Nitidulidae and 12 Staphylinidae families. Furthermore, Drosophilid flies were found in great numbers in each fruit and locality where we sampled. The genus *Drosophila* is separated in two subgenera: Sophophora, which include to *D. melanogaster*, and Drosophila. I classified all Drosophilid flies in the subgenus Sophophora, in the repleta species group in the subgenus Drosophila or in the genus *Zaprionus*. Ecological diverse groups were found together within decomposed fruits, such as omnivorous earwigs (Dermaptera), true bugs (Hemiptera), various parasitoid wasps and several omnivorous ants (Hymenoptera), an adult moth (Lepidoptera), predaceous green lacewings (Neuroptera) and thrips (Thysanoptera). Different groups in the Arachnida were present, such as predaceous pseudoscorpiones (Pseudoscorpiones), omnivorous harvestmen (Opiliones), distinct species of spiders (Araneae) and four mite species (Acari). The last group was found infesting some coleopteran species, and this forms the basis of Chapter 2 of this thesis. Finally, only one Collembola was sampled and two Crustacean morphospecies (Isopoda).

In the four arthropod assemblages that were compared (GPP: Guanajuato/Prickly pear; SPP: Sonora/Prickly pear; GO: Guanajuato/Orange; and SO: Sonora/Orange) four orders were overrepresented: Coleoptera, Diptera, Hymenoptera and Acari (Figure 2). Relative abundances differed between assemblages, but all showed that Coleoptera is the richest in species. Almost half of the species are coleopteran in each assemblage. In SPP, 38% of total abundance belongs to Hymenoptera, however, they are only two species of ants. Similar, in SO, three mite species comprised approximately 40% of the individuals. Most of those mites were attached to nitidulid beetles.

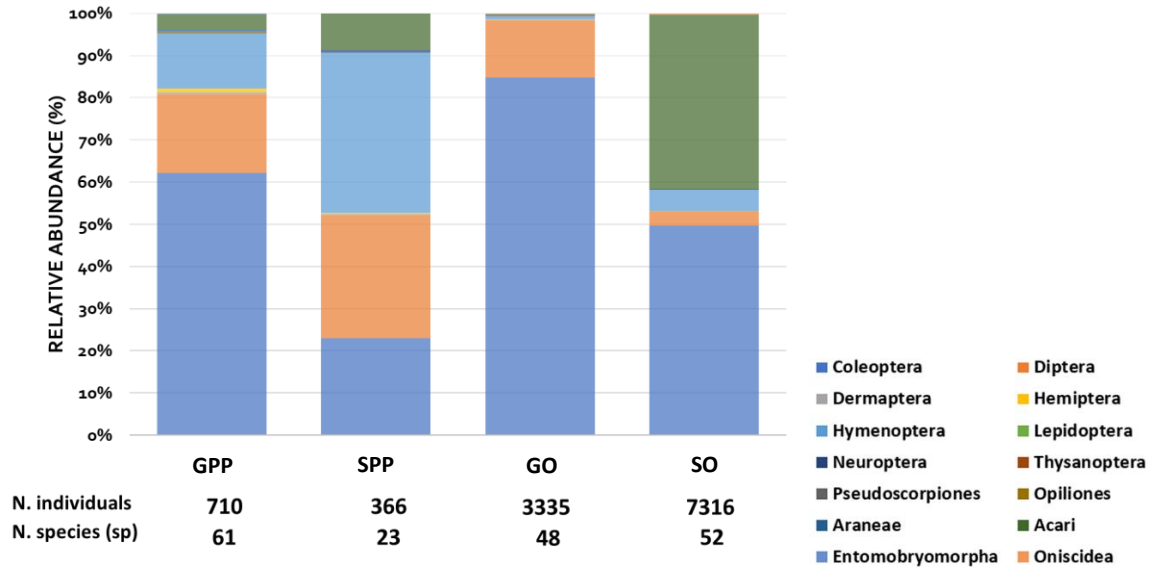


Figure 2. Relative abundance of arthropods by order.

Species richness varied across assemblages (Table 4). GPP had more observed species (61) and more species are expected if sampling effort is increased, based on the estimators that account for rare species. The SPP assemblages have the lowest observed and estimated species richness, even though that sampling effort was higher than for GO. In both orange assemblages, the number of individuals was huge. SO is two times the number of individuals of GO, nevertheless, species richness in both was lesser than GPP. I used a species accumulation curve to compare species richness among assemblages (Figure 3). As sampling efforts were not similar, data were extrapolated to 150 fruit samples using EstimateS. In prickly pear assemblages, fruits from Guanajuato were richer in species. In orange assemblages, Guanajuato had more observed species when the same number of samples are compared. However, in the rarefaction curve data overlap when 50 fruits are plotted and continuous overlapping until 150 samples. When the area of the curve overlaps with other, it does not provide strong statistical support, thus, both orange assemblages are not statistical different. In four assemblages, any accumulation curve was close to saturation (plateau), indicating that more samples are needed to estimate the real arthropod diversity in decomposed fruits.

Table 4. Species richness estimations. Numbers of samples, number of arthropod individuals (N), species richness (S), Chao01 and Chao02, and ACE and ICE indices.

Fruit type	State	Number of samples	Individuals (N)	Species richness (S obs)	Chao01	Chao02	ACE	ICE
Prickly pear	Guanajuato	126	710	61	117.25	95.98	101.89	101.1
	Sonora	77	366	23	39.62	64.7	39.64	45.29
Orange	Guanajuato	48	3335	48	44.36	59.99	55.71	61.96
	Sonora	118	7316	52	70.28	72.08	72.39	73.68

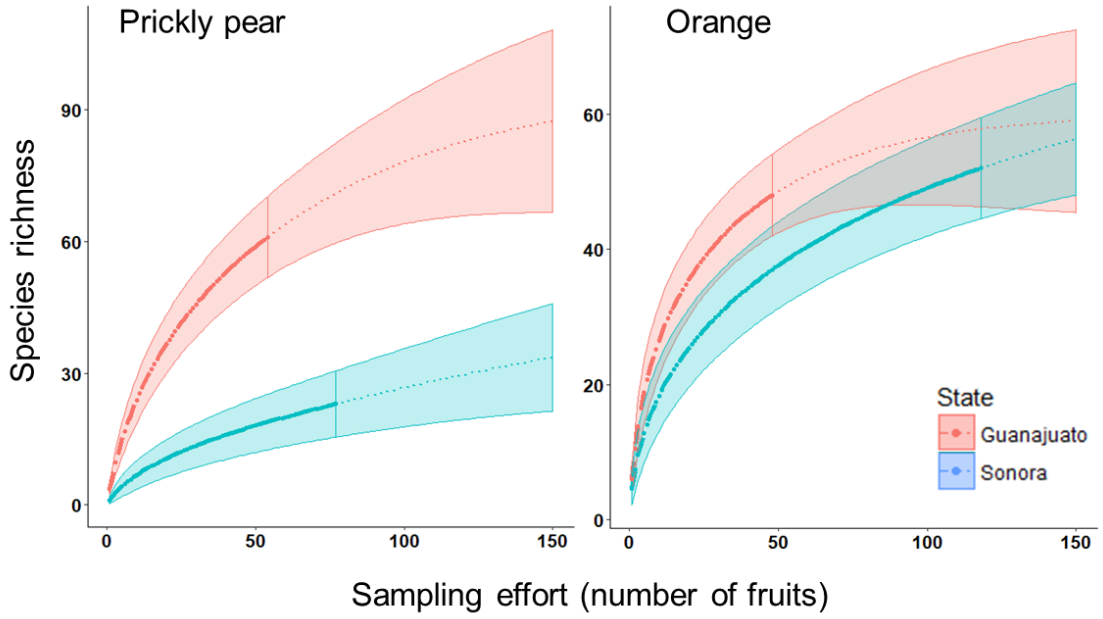


Figure 3. Species accumulation curves in localities and fruit type. Dashed lines indicate sampling effort done and dotted points indicate samples extrapolated to 150 fruits using EstimateS. Shaded regions indicate the 95% confidence intervals.

Species diversity estimations indicate that the GPP assemblage is the most diverse (Table 5). Shannon evenness (J) indices indicate whether abundances of species are similar in the assemblages. If they are more similar in distribution, species diversity will be greater. Evenness was greater in GPP and lower in SO. The Shannon index (H') and its transformed

value (Exp Shannon) showed that GPP has more species diversity, followed by GO. Sonoran assemblages are two times lower than Guanajuato in their transformed Shannon index. The Simpson (D) index, the probability that two randomly chosen individuals belong to the same species reveals that the GPP and GO assemblages had lower probability than those of SPP and SO.

Table 5. Diversity measurements for states and fruit types. Number of samples, number of individuals (N), Shannon Evenness (J), Shannon index (H'), Exp Shannon and Simpson index (D).

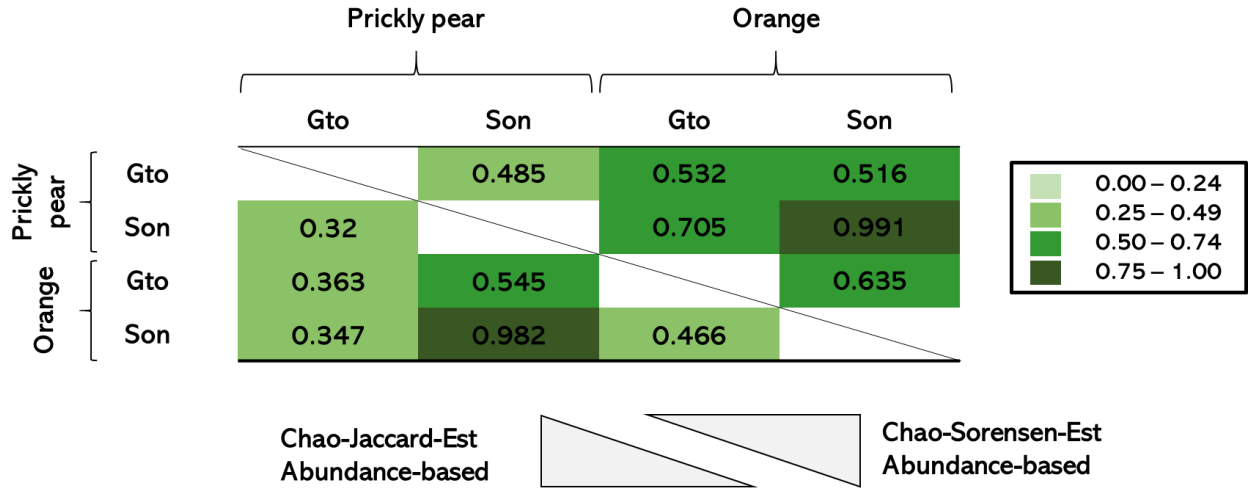
Fruit type	State	Number of samples	Individuals (N)	Shannon evenness (J)	Shannon (H')	Exp Shannon	Simpson (D)
Prickly pear	Guanajuato	126	710	0.701	2.88	17.75	0.109
	Sonora	77	366	0.603	1.89	6.64	0.226
Orange	Guanajuato	48	3335	0.641	2.48	11.91	0.147
	Sonora	118	7316	0.473	1.87	6.48	0.240

Arthropod compositional differences between fruit type and state were compared with a Venn diagram (Figure 4). GPP has more unique species when compared with Sonora and oranges. Many species are shared among all assemblages: the nitidulid beetles *Epuraea luteola*, *Carpophilus funebris*, *Conotelus mexicanus* and *Stelidota geminata*, the mycetophagid *Litargus* sp., *Drosophila* flies of the subgenus Sophophora and subgenus Drosophila repleta group, *Zaprionus* sp. and the earwig *Euborellia* sp. Interesting particular species in each aggrupation were found. For example, *C. lugubris*, *Lobiopa insularis* and the genus *Colopterus* (Nitidulidae) were found in both fruits but only in Guanajuato. *Aethina villosa* and *Aethina* sp. were specific of decaying prickly pears in Guanajuato. Additional taxa were unique in each assemblage but in lower numbers (Table S2). Pairwise comparisons based on similarity indices show that geographic location, more than fruit type, influences compositional differences in arthropod assemblages (Table 6). Sonoran assemblages (SPP and SO) are the most similar in arthropod composition, despite fruit type.

Moreover, assemblages from Guanajuato have elevated values (0.705 with Chao-Sorensen index) of similarity.

Figure 4. Comparisons of species composition in each assemblage by fruit type and state.

Table 6. Pairwise similarity indices among localities and fruit type. Chao-Sorensen abundance-based index (above) and Chao-Jaccard abundance-based index (below). Closer to 1 is more similar in species composition.



2. Ecology of the arthropod community and succession of decomposition.

To obtain information about the successional process of the arthropods in the necrotic fruit model, I classified prickly pears and oranges into five decomposed stages: fresh (F), early decay (E), active decay (Ac), advanced decay (Ad), and remains (R). I based my classification in color change, texture of tissue and water content (Zanetti et al., 2015). During the early decay, the fruit is typically damaged by an injury, but the tissue looks fresh and it's not covered with microorganisms. Throughout active decay stage the tissue is moist and soft, colonized by microorganisms (fermentation), while during the advanced decay the tissue is almost dry. In the remains stage the fruit is totally dry, with dark color and only the skin remains on the ground (Figure 5).

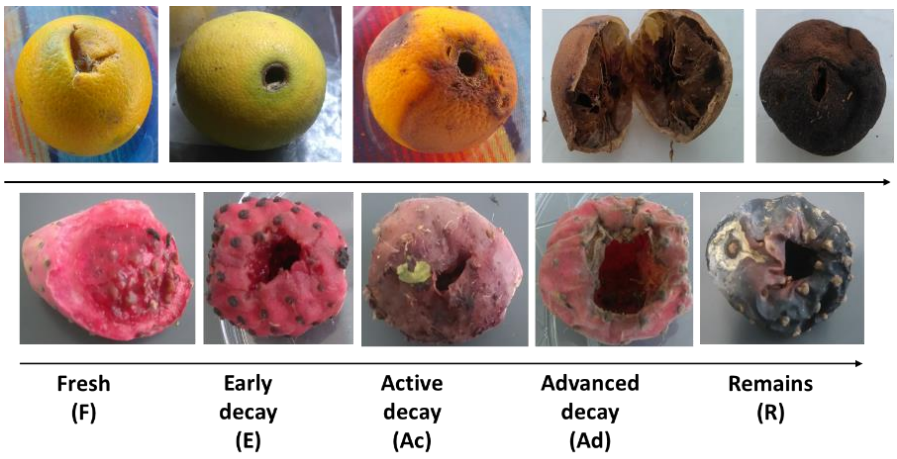


Figure 5. Stages of decomposition in oranges (top) and prickly pears (bottom).

Arthropods were categorized in five functional guilds (saprophagous, predators, parasitoids, parasites and omnivorous) depending on their feeding habits described in literature. When no information was found, species were positioned in the most appropriate guild to better known related species. Within the saprophagous, I added those species which are usually found in decomposed habits, where feed on detritus or microorganisms (bacteria and fungi). Nitidulids, drosophilid flies, Aleocharinae beetles,

ants, mites and woodlice were positioned in the saprophagous group. Omnivorous were the species with facultative diet preferences, such as detritus, microorganisms and other arthropods. Predators include those which feed on adult and larvae insects. Wasps were classified as parasitoids, whose progeny feed on beetle and fly larvae. Any species were cataloged as parasites since it was not clear if this relationship exist among given species. Likewise, some species were not included because no clear data was found.

Incidence of functional guilds showed that a succession of species is occurring during decomposition of fruits (Figure 6). In both fruits is observed an increment in the occurrence of functional groups. Saprophagous arthropods arrived earlier in decomposition while predators, omnivorous and parasitoids arrived later.

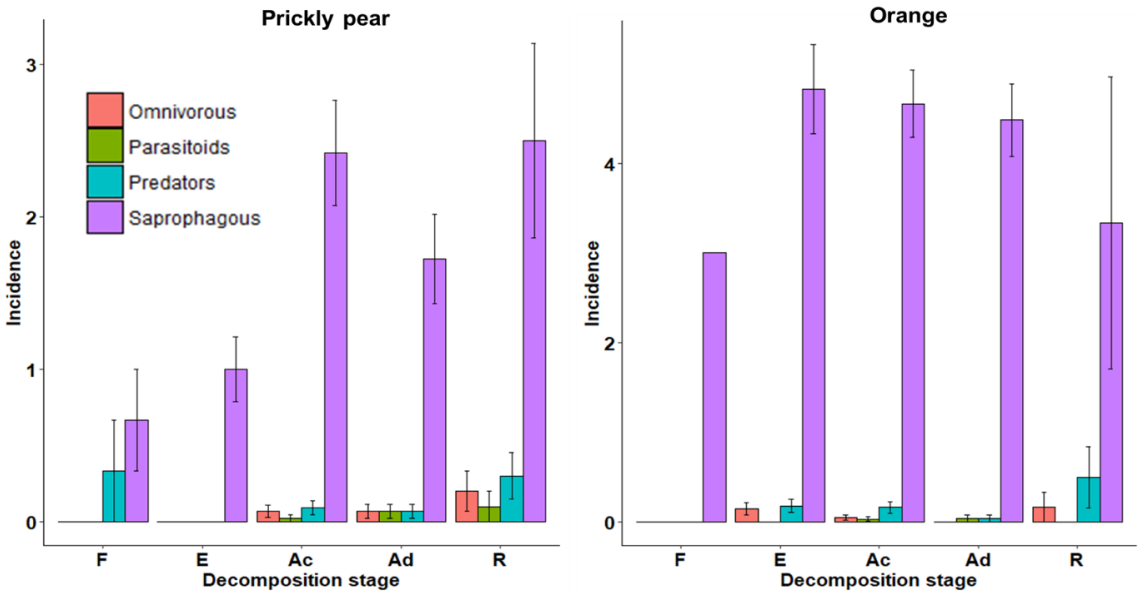


Figure 6. Mean incidence (±SE) of functional arthropod guilds in different decomposition stages of the fruit. Stages: fresh (F), early decay (E), active decay (Ac), advanced decay (Ad), and remains (R). Guilds: Omnivorous, Parasitoids, Predators and Saprophagous.

3. Population genetics of Nitidulidae species.

The coleopteran Nitidulid family was the most abundant and diverse in each arthropod assemblage. Almost each sample collected had at least one nitidulid specimen. I chose

seven species to ask if geographic differentiation exists between populations, since we found that geographic locality strongly influences arthropod community species diversity. Those species are: *Epuraea luteola*, *Carpophilus mutilatus*, *C. nepos*, *C. hemipterus*, *Urophorus humeralis*, *Stelidota geminata* and *Aethina villosa*. The mitochondrial CO1 gene fragment indicate varied patterns of genetic variability among nitidulid species. Nucleotide (π) and haplotype (*h*) diversity are highly variable in the seven nitidulid species (Table 7). *Epuraea luteola* had the lowest haplotype and nucleotide diversity, while *A. villosa* had the largest although *S. geminata* have greater number of haplotypes. High nucleotide and haplotype diversity are observed in populations from Guanajuato of *S. geminata* and *U. humeralis* and slightly of *C. hemipterus* when are estimated by separately. Negative Tajima's D and Fu's Fs values indicate purifying selection in mitochondrial CO1 fragments, but not significant.

Table 7. Summary of genetic diversity indices and results of neutrality tests (Tajima's D and Fu's FS) in the CO1 gene segments from *E. luteola, C. mutilatus*, *C. nepos*, *C. hemipterus*, *U. humeralis*, *S. geminata*, and *A. villosa* species.

Beelte species	N	L	k	K	*h* (±SD)	π (±SD)	Tajima's D	Fu's *Fs*
E. luteola	16	658	1	2	0.125 ± 0.106	0.00019 ± 0.00016	-1.16221	-0.700
C. mutilatus	11	615	4	4	0.673 ± 0.123	0.00177 ± 0.00054	-0.73668	-0.555
C. nepos	5	658	3	3	0.700 ± 0.218	0.00183 ± 0.00074	-1.04849	-0.186
C. hemipterus	13	618	5	6	0.821 ± 0.082	0.00205 ± 0.00043	-1.26863	-2.405
U. humeralis	10	618	2	3	0.378 ± 0.181	0.00090 ± 0.00046	-0.69098	-0.594
S. geminata	43	658	10	13	0.682 ± 0.078	0.00221 ± 0.00041	-1.08928	-7.576
A. villosa	16	606	7	7	0.825 ± 0.071	0.00245 ± 0.00052	-1.04598	-2.678
Guanajuato								
E. luteola	8	658	1	2	0.250 ± 0.180	0.00038 ± 0.0000001	-1.05482	-0.182
C. nepos	5	658	3	3	0.700 ± 0.218	0.00183 ± 0.00074	-1.04849	-0.186
C. hemipterus	5	618	3	3	0.700 ± 0.218	0.00227 ± 0.00092	-0.17475	0.061
U. humeralis	4	658	3	3	0.833 ± 0.222	0.00228 ± 0.00083	-0.75445	-0.288
S. geminata	24	613	4	6	0.721 ± 0.082	0.00190 ± 0.00029	0.24387	-1.506
A. villosa	16	606	7	7	0.825 ± 0.071	0.00245 ± 0.00052	-1.04598	-2.678

Sonora								
E. luteola	8	658	0	1	0	0	0	0
C. mutilatus	11	615	4	4	0.673 ± 0.123	0.00177 ± 0.00054	-0.73668	-0.555
C. hemipterus	8	618	3	4	0.750 ± 0.139	0.00150 ± 0.00040	-0.81246	-1.387
U. humeralis	6	618	2	3	0.600 ± 0.215	0.00140 ± 0.00055	-0.05002	-0.427
S. geminata	19	658	6	6	0.538 ± 0.133	0.00187 ± 0.00067	-0.92136	-1.734

Abbreviations: **N**, number of sequences; **L**, sequence length; **k**, number of variable sites; **K**, number of haplotypes; ***h***, haplotype diversity; **π**, nucleotide diversity.

The AMOVA analysis showed that genetic variation is distributed within *E. luteola*, *S. geminata* and *U. humeralis* populations and no genetic structure was observed. For *E. luteola* and *U. humeralis*, fixation index was very small. Small values imply that the frequencies of their haplotypes in both populations are very similar to each other. More than 22% of the genetic variation in *C. hemipterus* occurs between populations and a significant genetic structure was demonstrated.

Table 8. Hierarchical analysis of molecular variance (AMOVA) of mitochondrial CO1 fragments.

Beetle species	Source of variation	d.f.	Sum of squares	Variance components	Percentage of variation	Fixation index *Fst* (p value)
E. luteola	Among populations	1	0.063	0.00000	0.00	0.00000 (1.0000)
	Within populations	14	0.876	0.06256	100.00	
	Total	15	0.938	0.06256		
C. hemipterus	Among populations	1	1.621	0.16984	22.79	0.22788 (0.01466)
	Within populations	11	6.330	0.57547	77.21	
	Total	12	7.951	0.74531		
U. humeralis	Among populations	1	0.334	0.01306	4.59	0.04592 (0.47116)

	Within populations	8	2.170	0.27131	95.41	
	Total	9	2.504	0.28436		
S. geminata	Among populations	1	2.267	0.07422	9.68	0.09682 (0.00391)
	Within populations	41	28.387	0.69237	90.32	
	Total	42	30.654	0.76659		

d.f. = degrees of freedom.

Genetic diversity is reflected in haplotype networks and similar topology is observed when sequences are separated by state or fruit type (Figure 7 and Figure 8). No grouping is observed between Sonora and Guanajuato localities or prickly pear and orange fruits. On the other hand, different numbers of haplotypes are observed. *Stelidota geminata* has one common haplotype and several singletons (only one individual) haplotypes that vary by a small number of nucleotides. Contrasting networks are observed, such as in *E. luteola*, that has only one common haplotype while *A. villosa* has many. In *C. hemipterus* one shared haplotype is observed but two additional (one from Guanajuato and one from Sonra) appear to be frequent.

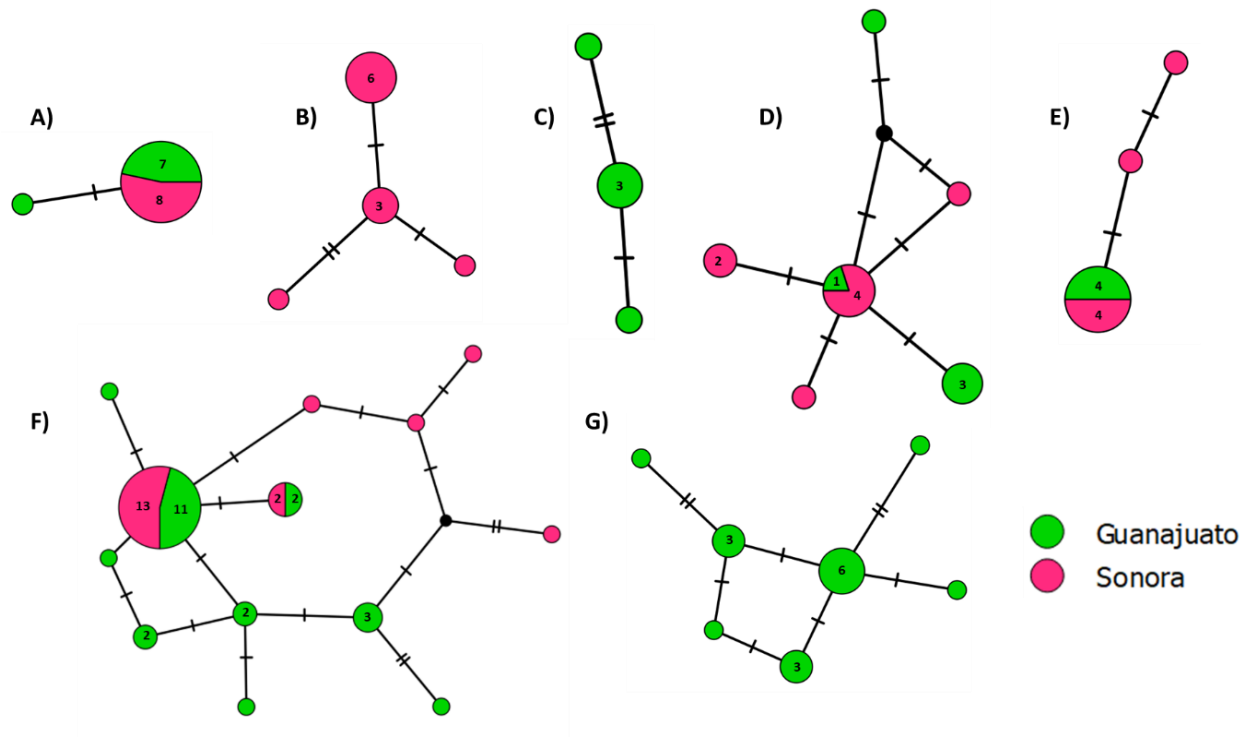


Figure 7. TCS haplotype networks based on mCO1 fragments of selected nitidulid species: A) *E. luteola*; B) *C. mutilatus;* C) *C. nepos;* D) *C. hemipterus*; E) *U. humeralis*; F) *S. geminata*; G) *A. villosa*. Diameter is relative to the number of specimens with a particular haplotype where the smaller circle represents a single individual. Colors correspond to different geographic origin of specimens. Each line mark represents a single nucleotide substitution and dots on branches represent inferred missing haplotypes.

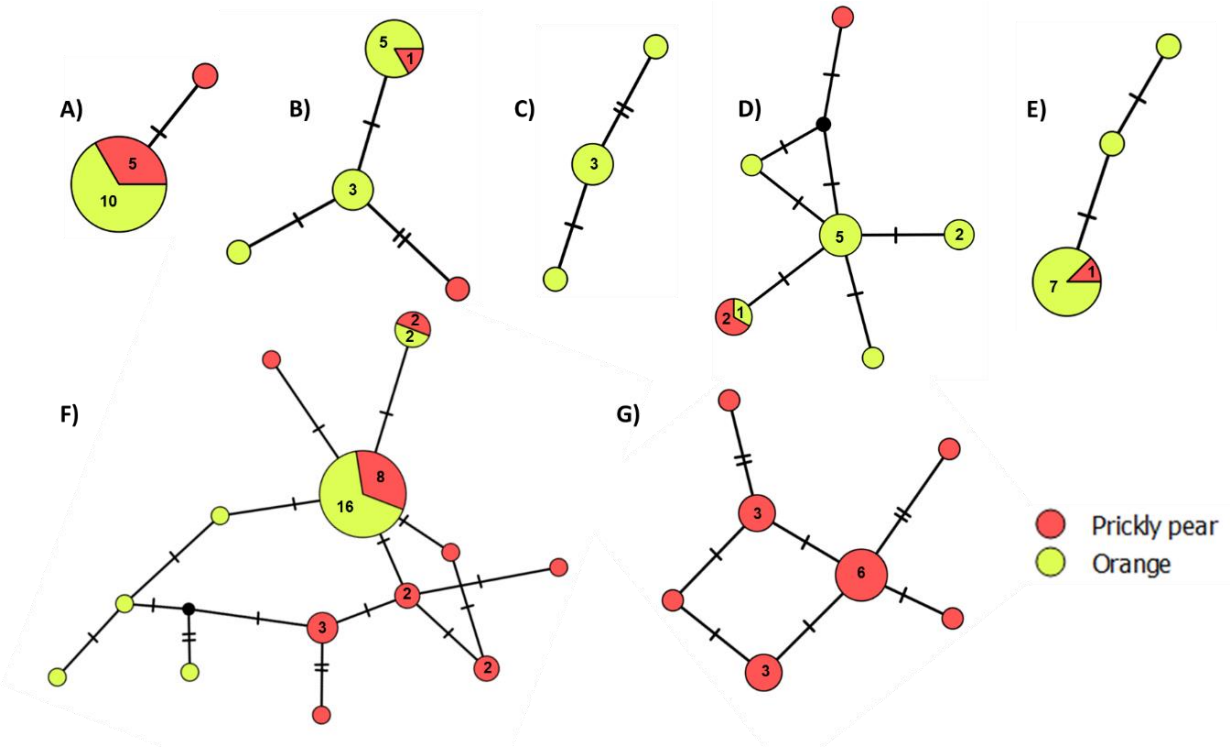


Figure 8. TCS haplotype networks based on mtCO1 fragments of selected nitidulid species: A) *E. luteola*; B) *C. mutilatus;* C) *C. nepos;* D) *C. hemipterus*; E) *U. humeralis*; F) *S. geminata*; G) *A. villosa*. Diameter is relative to the number of specimens with a particular haplotype where the smaller circle represents a single individual. Colors correspond to different decomposed fruit origin of specimens. Each line mark represents a single nucleotide substitution and dots on branches represent inferred missing haplotypes.

Discussion

Arthropod communities associated with decaying prickly pear and orange fruits were highly diverse. Geographic location, more than the fruit type, was considerably the major factor affecting arthropod diversity. The most abundant and diverse arthropod taxa found here were Coleoptera, Diptera, Hymenoptera and Acari. Within those groups, the coleopteran family Nitidulidae was the most diverse group that exploit the rotting fruit resource.

Similar to previous studies made in cacti (Castrezana & Markow, 2001; Delgado-Fernández, Escobar-Flores, & Franklin, 2017; Ferro et al., 2013; Richmond, Reese, Mejía, & Markow, *in revision*) and *Citrus* fruits (Mutinelli et al., 2016), the beetles, flies, hymenopterans and mites are the dominant arthropod fauna on the decomposed plant niche. Nevertheless, arthropod communities in decaying fruits harbored more species and taxa groups than those characterized in cactus. Unlike rotting fruits that are rich in carbohydrates, cactus substrate is unique in chemical content, since are deficient in nutrients and produce several compounds that are harmful for several insects, like alkaloids, sterol diols or terpenoids (Fogleman & Danielson, 2001) that limit the occurrence of several species.

Coleoptera was the most diverse arthropod group in decaying fruits, which agrees with proportional expectations. Beetles are one of the most successful orders of arthropods due to their variety of lifestyles and in terms of number of species, they comprise over one-third of all insects, followed by Diptera and Hymenoptera (Stork et al., 2015; Zhang, 2013). During morphological and molecular identification of specimens, my collaborator Dr. Andrew R. Cline realized that some Nitidulidae are probably undescribed new species. As result, coleopteran diversity is actually higher than we calculated before. Future research should consider that underestimations in species diversity could be a consequence of poor identification of specimens. Here, decaying fruits offered an excellent model to explore the variety of arthropods as a new hotspot in biodiversity studies.

The most common arthropod group in fruits I recovered, Nitidulidae, is a worldwide distributed family, that feeds and breeds on a variety of decomposed fruits and vegetables (David & Brown, 2009; Majka, Webster, & Cline, 2008). Certain nitidulids, principally

Carpophilus spp., are considered important pests in orchards, agricultural crops or stored products (e. g. grains and dried fruits), because they damage plants by chewing or are vectors of pathogenic microorganisms (Bartelt, Diana, Petroskl, & Baker, 1995; David & Brown, 2009; Emekci & Moore, 2015). Because of their lifestyle, nitidulids are easily dispersed by human trade, making them frequent fauna in fruits. Among the nitidulids considered as pests, I found *Carpophilus hemipterus*, *C. mutilatus*, *C. lugubris*, *C. nepos*, *Urophorus humeralis, Epuraea luteola* and *Stelidota geminata*.

Although I observed some specific species from each assemblage, arthropods were mostly generalists in rotten fruits. *Aethina villosa*, *Aethina* sp. and *Lobiopa insularis* were particularly interesting due its restricted distribution to Guanajuato. While *A. tumida* is an important invasive species through all the world, which infest honeycomb colonies (Li et al., 2018), information in the biology of *A. villosa* is poor. *Lobiopa insularis* is an extensively distributed species that feeds upon a variety of substrates, such as decaying fruits (mango, strawberry, grape, apple, tomato, pineapple and guava), plant inflorescences and sap flows (Ellis, Delaplane, Cline, & Mchugh, 2008; Hernández-Torres et al., 2018). Hernández-Torres et al. (2018) had found *L. insularis* in Durango, Mexico, revealing that its distribution is not restricted to Guanajuato but limited in the Sonoran Desert.

A number of factors can determine the diversity of species or the occurrence of a particular arthropod on decaying fruits. Arthropod diversity differed between fruit type and among geographic location. The GPP assemblage was the most diverse but also more samples were collected. Sampling effort is a crucial factor in comparative diversity studies. For example, Richmond et al. (*in revision*) found that cardón cactus (Pachycereus pringlei) was more diverse in arthropod communities than organ-pipe (*Stenocereus thurberi*), and senita (*Lophocereus schottii*), but more cardons had been sampled and they are much larger cacti than the other two. I found that even with a large number of samples, the number of arthropod species expected is huge. Thus, additional sampling will reveal even more species. Besides, four *Opuntia* species were used and more sampling effort was performed in Guanajuato. While for oranges only one species was used and in Mexico, *Citrus sinenis* is commonly found only in orchards.

In the Sonoran Desert, both landscape and plant use shapes arthropod distributions in decaying cactus (Pfeiler & Markow, 2011). Herein, I observed that arthropods associated with rotten fruits are mainly determined by the place they inhabit. Guanajuato and Sonora landscapes share semiarid climate, but in Sonora temperatures are normally elevated. Sampling of prickly pears in Sonora was performed in the Sonoran Desert, while oranges were in a private orchard within the desert. In Guanajuato, sampling of *Opuntia* fruits was done in the wild, but in semi urban habits with semiarid climate. Oranges were collected on private Guanajuato gardens with very variable climate conditions (arid to humid).

Fruit type was not a considerable factor shaping decaying fruit communities. Both prickly pears and oranges are rich in many compounds and have an elevated quantity of free-sugars. The pulp of *Opuntia ficus-indica* fruit, for example, is richer in carbohydrates (58%; principally of glucose and fructose) than in proteins and lipids (< 5.9% and < 0.9%, respectively) (El Kossori, Villaume, El Boustani, Sauvaire, & Méjean, 1998; Salim, Abdelwaheb, Rabah, & Ahcene, 2009) and contains high levels of potassium (Salim et al., 2009). Oranges, in addition, also are rich in carbohydrates. Around 80% of solid components are sugars, but principally sucrose (Kelebek & Selli, 2014). As well, both fruits are rich in organic acids, principally citric (Kefford, 1960; Stintzing et al., 2005). In addition, during decomposition, the microorganisms, principally yeast, that invade and ferment the tissue changing the composition of the substrate or increasing or decreasing the toxicity (Fogleman & Danielson, 2001; Ganter, Morais, & Rosa, 2017). Here, I did not attempt characterize microbial communities or nutritional composition of decaying prickly pear and orange fruits during decay. Moreover, given similar results in arthropods composition, both fruits provide similar nutritional resources to arthropods, along with similar microbial communities, assuming that given arthropods are vectors of those microorganisms.

One of the most interesting observations in arthropod communities was the coexistence of many phylogenetically related species of the same functional guild in the fruit. In decaying oranges, a huge number of beetles of the genus *Carpophilus* spp. and *Colopterus* spp. were co-occurring at the same time (e. g. around 800 specimens of a number of species per fruit). More remarkable is that ephemeral habits are limited in resources. Experimental

tests have showed that *Carpophilus* spp. are very attracted to the aggregation pheromones of its relatives or even from other species to find mate or food (Bartelt et al., 1995), which explains the high numbers of beetles but not their coexistence. Ecological theory explains that biodiversity is maintained if niche differences between coexisting species exist (Levine & HilleRisLambers, 2009). If two species are competing for the same resource, one is predicted to disappear. Instead, if one differs in its strategy to obtain resources, species will coexist as result of stabilizing effects of niche differences (Levine & HilleRisLambers, 2009). Little is known about niche partitioning in nitidulid communities. One hypothesis is that beetles are specializing upon different microorganisms within the fruit and/or colonizing the fruit at different stages of decomposition. *Carpophilus lugubris*, for example, have shown strong preference for one substrate, whole wheat bread dough over rotten fruits, when was compared with *S. geminata*, *Glishrochilus fasciatus* and *G. quadrisignatus* (Blackmer & Phelan, 1991). Temporal partitioning has been observed in decaying oranges colonized by *Drosophila*, as different species arrive at different stages of decomposition (Nunney, 1990). Therefore, future research should explore resource and temporal partitioning in Nitidulidae.

The arthropod communities changed over time in both decomposed fruits. Species with specific nutritional requirements may be dominating early stages while species with a broad diet dominate afterward. The first organisms who arrived were saprophagous, principally represented by Drosophila and Nitidulidae, which feed and breed on rotting orange and prickly pear fruits. Saprophagous arthropods dominated the fruits during all the process of decomposition, only varying in their incidence and abundance. Secondly, several predators (24 species) were found in decaying communities. They appeared in the more advanced stages of decomposition when dipteran and coleopteran eggs, larvae and pupae are available. A similar succession pattern was observed in barrel cacti (Ferro et al., 2013) wherein the abundance of predators is directly related to advanced cactus decay. Parasitoid and predator species should be regulating populations within the community. For example, *D. starmeri* is controlled by the action of a number of species, including *Pseudomyrmex* and *Zacryptocera* ants, both larvae and adult Staphylinidae in decaying

Pilosocereus lanuginosus cacti (Escalante & Benado, 1990). I found several staphilinids, hysterids, pseudoscorpions, spiders and even hemiptera, any of which could be feeding upon the saprophagous species.

Arthropods that exploit ephemeral resources previously have been hypothesized to have high dispersal capabilities and thus an absence of population structure (Pfeiler & Markow, 2011). Nitidulids exploit ephemeral resources, are distributed worldwide and utilize a variety of substrates (Emekci & Moore, 2015; Newton, 2015). Thus, if nitidulid species are strong dispersers, I expect, even if there is high genetic diversity, a lack of population structure across geographic locations. The examined species, with exception of *E. luteola* and *U. humeralis*, showed high nucleotide and haplotype diversity in the mitochondrial CO1 gene. The low genetic diversity in *E. luteola* and *U. humeralis* may be the result of a founder effect or selective sweep. Lack of genetic differentiation, however, was detected in three of four species found both in Sonora and Guanajuato. *Carpophilus hemipterus* was the only beetle showing genetic differentiation among Sonora and Guanajuato populations. Either *C. hemipterus* is not as strong disperser as the others, possibly because its dispersal is restricted by factors like human transport or different fruit preferences. Neutrality test indicated possible purifying selection in CO1 fragment, that indicate exists a selective pressure in nitidulid populations.

A broad died must be important in maintaining genetic variation in nitidulid species since analyzed species have a wide spectrum of hosts. Besides, gene flow between populations from Sonora and Guanajuato is probably acting in shaping beetle variability. Gene flow could be consequence of its own dispersal capability, since some species could travel up to 4 km, but more plausible is that dispersion is facilitated by human commerce (Emekci & Moore, 2015).

Despite the importance of some nitidulids as potential pest and invasive species, little work has explored their genetic variation, population analysis or even exploring patterns of invasion (David & Brown, 2009). In comparison, genetic studies of the invasive fruit fly *Zaprionus indianus* using the gene marker CO1 revealed no genetic structure among six

populations in Mexico and one from Panama. Additionally, haplotype variation is given by more than one possible invasion from Africa to South and North America (Markow et al., 2014). Similar to *Z. indianus*, the genetic variation in Nitidulidae may reflect several introductions of beetles to Mexico, facilitated by human trade. Future comparisons of haplotypes from source populations could resolve this question.

The barcode region of the mitochondrial gene CO1 has been successfully used for species discovery and phylogenics for many years (Hebert et al., 2016). This 650 bp region is particularly useful to identify species and in population genetic studies to answer ecological and evolutionary questions (Barrowclough & Zink, 2009; Hebert, Cywinska, Ball, & deWaard, 2003). Several sequence databases are available, such as the public database BOLD (Andújar et al., 2018) that can be used in several comparative studies and as a global bioidentification tool. Here, CO1 allowed us to identify and infer evolutionary relationships between nitidulid species. Besides, it had been useful to infer phylogenetic relationships within the genus *Carpophilus* (David & Brown, 2009). Also, this region can be successfully used to detect potential invasive nitidulids by employing the real-time PCR assay (Li et al., 2018).

Decomposition of organic matter is one of the most important ecological services performed by arthropods by creating patches of energy and nutrient flow (Barton et al., 2013). As well, ecological services are directly affected by biodiversity complexity. For example, litter decomposition rate increased when more plant functional groups were added (Scherer-Lorenzen, 2008). Arthropods associated with decomposed plants or animals are highly varied. Here, decaying prickly pears and oranges provided an excellent opportunity to study the diversity of the arthropods associated with decomposed plants. I found a great amount of species diversity within the fruits, included high genetic diversity in the nitidulids examined. While this diversity likely contributes to ecosystem functioning, future studies would be required to address the roles of the individual species in these processes.

Conclusion

The diversity of arthropods associated with decaying prickly pear and orange fruits was greatly diverse and many species are expected to exist. Indeed, decaying fruits demonstrated to be a strong system to study the diversity and ecology of arthropods. Several species were sampled from four classes (Insecta, Arachnida, Collembola and Crustacea) keyed in 14 orders and 29 families. The most abundant groups were Coleoptera, Diptera, Hymenoptera and Acari and of those the most diverse was the worldwide distributed family Nitidulidae. Decaying fruits harbored more arthropod species than those previously studied in necrotic cactus in the Sonoran Desert (Castrezana & Markow, 2001, Ferro et al., 2013; Richmond et al., in press). Differences in diversity must be given by the chemical differences between substrates, since fruits provide more sugars while cactus are richer in detrimental compounds, besides the specificity of the species than can survive under awkward conditions. Arthropod composition was highly influenced by the geographic location more than the fruit type. Climatic conditions or dispersion are a possible restricting arthropods distribution. Although many species were specific from one location or fruit type, the species were mostly generalist. Likewise, succession pattern might be influencing arthropod communities, since I observed that different species can colonize the fruit at different times: saprophagous arrived first while other functional groups arrived later. Nitidulidae are worldwide distributed and considered invasive species. Those beetles have a broad range of diets and are mostly transported by human trade. Of four examined species, only one, *Carpophilus hemipterus*, showed geographic differentiation between populations. This difference could be given by restricted dispersal giving rise to local adaptations among localities.

Resumen

La foresis es una forma de comensalismo en la que una especie es dispersada por otra por un período de tiempo limitado y con el único propósito de dispersión. La foresis es una estrategia ventajosa para los organismos con movilidad limitada, principalmente para aquellos que viven en ambientes efímeros. Los ácaros son fauna común en hábitats irregulares, como plantas en descomposición, pero requieren la asistencia de huéspedes altamente móviles, como escarabajos, para trasladarse de un lugar a otro. La selección del huésped es un factor importante para los ácaros, ya que de esto depende su éxito en la dispersión a un nuevo hábitat, apareamiento o la obtención de recursos nutricionales. Por lo tanto, muchas interacciones están restringidas a un rango limitado de hospederos. En tunas y naranjas podridas, siete miembros de las familias Nitidulidae y Staphylinidae fueron infestados por dos especies de ácaros foréticos en el estado de Sonora, México. Las deutoninfas de *Histiostoma* sp. (Astigmata: Histiostomatidae) mostraron preferencia de infestación por escarabajos nitidúlidos. El sesgo en la infestación se confirmó experimentalmente mediante una prueba de elección por pares. Sin embargo, no hubo diferencia en la infestación entre las cuatro especies infestadas por *Macrocheles* sp. (Mesostigmata: Macrochelidae). Además, ambos ácaros foréticos mostraron diferentes patrones en su distribución en el cuerpo del hospedero, derivados de las estrategias fisiológicas y de defensa de cada especie. Muchos factores deben determinar las preferencias de los ácaros, como la disponibilidad de escarabajos, el tamaño del hospedador o las señales químicas. Las relaciones foréticas están ampliamente distribuidas en la naturaleza, pero poco estudiadas en los escarabajos de las familias Nitidulidae y Staphylinidae. Nuevos estudios deberían explorar la extensión de estas interacciones y cómo podrían afectar la ecología de cada especie.

Abstract

Phoresy is a form of commensalism in which one species is dispersed by another for a limited period of time and with the only purpose of dispersion. Phoresy is an advantageous strategy for organisms with limited mobility, principally for those that live in ephemeral habitats. Mites are common fauna in patchy habitats, such as decaying plants, but require the assistance of highly mobile hosts, like beetles, to move to new places. Selection of the host is an important factor for mites since on this depends their success in dispersion to a new habitat, mating or the obtention of nutrition resources. Therefore, many interactions are restricted to a limited range of hosts. In rotten prickly pears and orange fruits, seven members of the Nitidulidae and Staphylinidae families were infested by two phoretic mite species in the state of Sonora, Mexico. Deutonymphs of *Histiostoma* sp. (Astigmata: Histiostomatidae) showed infestation preference for nitidulid beetles. Bias in infestation was experimentally confirmed through a pairwise choice test. However, there was not difference in infestation among the four infested species by *Macrocheles* sp. (Mesostigmata: Macrochelidae). Besides, both phoretic mites showed different patterns of distributions on host body, derived from physiological and defense strategies by each species. Many factors should be determining mite preferences, such as beetle availability, host size or chemical cues. Phoretic relationships are broadly distributed in nature, but poorly studied in the Nitidulidae and Staphylinidae beetles. Further studies must explore the extensiveness of given interactions and how can affect in the ecology of each species.

Chapter 2: Phoretic mites on Nitidulidae and Staphylinidae beetles on rotten fruits.

Introduction

Phoresy is form of commensalism in which one animal is used by another, called the phoretic, for a limited period of time and with the primary purpose of dispersal from one place to another (Camerik, 2009; Houck & OConnor, 1991). In spatial and temporal ephemeral habits, such as carrion, dung or decaying plant material, phoresy is an advantage for a variety of arthropods with restricted mobility, giving them a strategy for the rapid colonization of new resources. In the necrotic cactus, for example, the pseudoscorpion *Dinocheirus arizonensis* (Pseudoscorpiones) preys upon several cactophilic insects but still has limited dispersal. To colonize new fresh rots, *D. arizonensis* attaches the legs of the neriid cactus fly *Odontoloxozus longicornis* and occasionally to other insects (Pfeiler et al., 2009; Pfeiler & Markow, 2011).

Mites are particularly common in ephemeral environments, where they feed on decaying material, microorganisms or prey on small arthropods or even on fly larvae (Castrezana & Markow, 2001; Perez-Leanos, Loustalot-Laclette, Nazario-Yepiz, & Markow, 2017; Perotti & Braig, 2009; Walter & Proctor, 2013). In Acari, phoresy is widely distributed and is a fixed life history trait because they require assisted dispersal by a highly mobile host (Houck & OConnor, 1991). Mites have developed morphological and behavioral strategies that allow them to improve their chances to infest a potential host. In many Mesostigmata species, only adult gravid females are transported or rarely, some immature stages. In contrast, in the Astigmata, the immature deutonymph is the phoretic stage (OConnor, 1982). Deutonymphs are principally characterized principally by the absence of a mouth and a foregut and a ventral attachment organ (known as sucker disk), for holding against the host (Houck & OConnor, 1991; Walter & Proctor, 2013).

Phoretic associations between mites and beetles are particularly common owing to the flight capabilities of these hosts. Several interactions are described in literature, exhibiting the complexity of phoretic symbiosis. Phoresy is transitory and mites can benefit from several host, but many interactions become restricted to a range of host. For example, Perez-Leanos et al. (2017) found that the generalist *Macrocheles subbadius* (Mesostigmata) infests a wide range of Drosophilid flies although there appears to be a preference for the phylogenetically related flies of the repleta group. At the same time, distribution of mites on host body is nonrandom and can vary by species and sex of the host (Cross & Bohart, 1969). In the phoretic relationship between the bee *Nomia melanderi* and four mite species, mite distribution on the host depends on the species, and for two of them depends on the sex (Cross & Bohart, 1969).

Sap beetles belong to the worldwide-distributed family Nitidulidae (Cucujoidea). Approximately 2800 species are divided into nine subfamilies (Cline et al., 2014). Nitidulids have a variety of lifestyles, principally saprophagous, feeding on decaying plants and fruits, carrion, fungi, pollen andhoney bees (Emekci & Moore, 2015). Some species are of interest as they are minor pests in orchards and stored products. Nitidulids are carriers of phoretic mites in other ephemeral habits. For example, in tree-sap exudate, some species of the genus *Hericia* (Astigmata: Algophagidae) utilize *Soronia fracta* (Nitidulinae), *Librodor japonicus*, *Amphicrossus lewisi* (Amphicrossinae) and *Glischrochilus obtusus* (Cryptarchinae) to colonize new sap flux of oak trees (*Quercus* spp.) (Fashing, 2008; Hayashi, Ichikawa, & Yasui, 2011). *Aethiophenax luteoli* (Trombidiformes: Acarophenacidae), an egg parasitoid on several beetle species, has been found under the elytra of *Epuraea luteola* (Epuraeinae) (Katlav, Hajiqanbar, & Talebi, 2015). The phoretic relationship between *Mystrops* spp. (Nitidulidae) beetles and mites of the genus *Xanthippe* (Mesostigmata: Ascidae) is suggested by their presence in the inflorescences of the palm *Socratea exorrhiza* in Venezuela (Naskrecki & Colwell, 1995).

Staphylinidae is one of the major coleopteran groups with more than 47,000 species described (Navarrete-Heredia et al., 2002). Staphylinids, or rove beetles, have a variety of life styles, including those which feed on decomposed material (plants, animals or fungi),

predators and some others are parasitoids in insect nests (Navarrete-Heredia et al., 2002). Rove beetles have been found carrying mesostigmatan (*Macrocheles glaber*, *Crassicheles holsaticus*, *Thinoseius spinosus* and *Uroobovella pyriformis*) and astigmatan mites (*Pelzneria* sp. and *Spinanoetus* sp.) in human carrion (Perotti & Braig, 2009).

Here, I explored the association between phoretic mites and beetles of the families Nitidulidae and Staphylinidae associated with decomposed prickly pear and orange fruits in Mexico discussed in Chapter 1. I asked if a bias exists in the infestation of the phoretic mites and their potential beetle hosts.

Methods

Sampling of mites. As the first observations of phoretic mites were on coleopterans in the state of Sonora, my sampling was confined to this state (Figure 1). A total of 134 oranges were sampled in the locality of Navojoa in three separated collections: April, October 2017, and March 2018. In November 2017, I sampled 33 prickly pears in the locality of Batacosa. Beetles were collected individually with forceps, taken to the laboratory and examined under a stereomicroscope. I separated those beetles infested with mites and recorded the number and body part of the beetle to which they were attached.

Figure 1. Localities sampled in the state of Sonora, Mexico.

Identification of beetles. Beetles were examined under a ZEISS stereo-microscope and classified initially by morphospecies (Table 1) using morphological keys (Ewing and Cline 2005; Navarrete-Heredia et al. 2002) and verified by Dr. Andrew R. Cline from the Plant Pest Diagnostics Center in California, USA. I subsequently verified the morphological identification by sequencing the 658-base pair barcode region of the cytochrome oxidase subunit I (CO1) mitochondrial gene using the primer sequences described by Folmer et al. (1994) under the following PCR conditions: one cycle of denaturation at 94 °C for 3 min, 35

cycles of 94 °C for 30 s of denaturation, 52 °C for 1 min of annealing, and 72 °C for 1 min of extension, and a final extension of 7 min at 72 °C.

Identification of mites. Morphological identification of mites was performed by placing each specimen in "glycerine jelly" mounting medium. Mites first were washed in PBS and incubated with KOH 10 % at 99 °C for 10 min. They were washed with sterile water, ethanol 70 % and ethanol 100 % to remove the KOH solution from sample. After incubation in lactic acid:ethanol (1:1 v⁄v) for 30 min at room temperature and they were placed in a drop of heated glycerine jelly on a slide.

For molecular identification, total DNA was obtained by mashing mites in "squishing buffer" [10 mM Tris-Cl, pH 8.0, 1 mM EDTA, 25 mM NaCl, 200 µg/ml freshly diluted Proteinase K solution]. A pipette tip was used to macerate the mite before incubating at 37 °C for 60 min. Proteinase K then was inactivated by heating to 95°C for 2 min. We used the 530-base pair fragment 18S rDNA gene to identify the mites with the primer sequence Fw1230 (5'-TGAAACTTAAAGGAATTGACG-3') from Skoracka and Dabert (2010) and ConsR18S (5'-ATTCAATCGGTAGTAGCGACG-3') from Perez-Leanos et al. (2017). We used 1.5 µl in a 10 µl PCR reaction volume. PCR conditions were: one cycle of denaturation at 95 °C for 3 min, 35 cycles of 95 °C for 30 s of denaturation, 54 °C for 45 s of annealing, and 72 °C for 90 s of extension, and a final extension of 30 min at 72 °C. Amplicons were purified with the QIAquick™ PCR Purification Kit (QIAGEN® Inc., Valencia, CA). PCR products were sequenced by Sanger technology at the LANGEBIO core DNA sequencing facility.

Species preference test. Because no free-living mites were encountered in nature, I used mites taken from infested beetles collected from the oranges. Mites of *Histiostoma* sp. found infesting *C. mutilatus* were detached for the choice test. Mites were very tiny and did not support a lot of time without a beetle host on the petri dish, thus, they were immediately used for choice experiments. A preliminary trial used dead beetles to maintain them immobile. Subsequently live specimens were utilized and, since no difference in treatments was observed (Fisher's exact test, p-value = 0.5396), the observations were pooled. Live beetles, immobilized in Eppendorf tubes placed on ice to carefully remove

their legs, were placed on a Petri dish with agarose 0.5 %. Two beetle species were placed in the center of the dish about 15 mm apart. A single mite was placed between the two beetles and observed for one hour. I measured the time from mite introduction until a beetle was chosen or until the hour was up.

Results

Twenty-nine coleoptera species from eight families were found living both in rotten fruits. Nitidulidae and Staphylinidae were the most diverse families found, in species richness and abundance, *Carpophilus mutilatus* and Aleocharinae 01 being the most abundant species respectively (Table 1). Seven of the 29 beetle species were infested with phoretic mites. Five nitidulids and two staphylinids were infested with the mite *Histiostoma* sp. (Astigmata), while only three nitidulids and one staphylinid were infested with the uncharacterized *Macrocheles* sp. (Table 2). Non-simultaneous infestations per individual were found. Most of the beetles were from oranges. From prickly pears, only one nitidulid species had mites.

Table 1. Number of Coleoptera species collected from decaying prickly pears and oranges in Sonora.

Family	Species	Prickly pear (n=33)	Orange (n=161)
NITIDULIDAE	*Epuraea luteola*	4	422
	Carpophilus hemipterus		35
	Carpophilus mutilatus	57	1831
	Carpophilus delkeskampi		1
	Carpophilus nepos		4
	Carpophilus funebris	1	
	Urophorus humeralis	1	146
	Colopterus denticulatus		9
	Conotelus mexicanus	2	7
	Stelidota geminata		142
	Stelidota sp.2		1
	Stelidota sp.3		1
MYCETOPHAGIDAE	*Litargus* sp.	7	2
HISTERIDAE	*Carcinops consors*		4
STAPHYLINIDAE	Aleocharinae 1	1	3022
	Aleocharinae 2		85
	Aleocharinae 3		1
	Aleocharinae 5	1	1
	Staphylininae 1		4

	Belonuchus apiciventris		2
	Tachyporinae 2	1	
	Staphylinidae 6		1
ZOPHERIDAE	*Bitoma* sp.		16
MONOTOMIDAE	*Europs* sp.		5
SILVANIDAE	Ahasverus rectus	1	3
TENEBRIONIDAE	Tenebrionidae 1		1
	Tenebrionidae 2		2
	Tenebrionidae 3		4
	Coleoptera 10		28

While the overall percentage of infestation of both mites was low, *Histiostoma* sp. was the most prevalent, occurring in 197 beetle specimens while *Macrocheles* sp. occurred in 41 (Table 2). There was a difference in prevalence of *Histiostoma* sp. between beetle species collected from oranges ($X^2 = 163.0$, df = 6, $p < 0.0001$), but did not differ among species collected from oranges versus prickly pears ($X^2 = 162.3$, df = 6, $p < 0.0001$). Likewise, *Histiostoma* sp. were more prevalent on nitidulid (6.42%) over staphylinid beetles (0.80%) ($X^2 = 141.1$, df = 1, $p < 0.0001$). Despite *U. humeralis* having the highest percentage of infestation, the difference with *E. luteola*, *C. mutilatus, C. hemipterus*, and Aleocharinae 02 was not significant. Infestation of *U. humeralis* was significantly higher, however, compared to *S. geminata* and Aleocharinae 01 (Fisher's exact test, $p = 0.011$ and $p < 0.0001$, respectively). Meanwhile, the prevalence of the mite *Macrocheles* sp. between species was not significantly different ($X^2 = 4.174$, df = 3, $p = 0.2432$) nor between families ($X^2 = 3.761$, df = 1, $p = 0.0525$).

Table 2. Mite associations and beetle species collected from rotten orange or prickly pear fruits and percentage of infestation.

Fruit	Family	Species	Mite species	Number of beetles	Infested beetles	% Infestation
ORANGE	NITIDULIDAE	*E. luteola*	Histiostoma sp. (Astigmata)	422	29	6.87 [a]
		C. mutilatus		1831	122	6.66 [a]
		U. humeralis		146	12	8.22 [a]
		C. hemipterus		35	1	2.86 [a]
		S. geminata		142	2	1.41 [b]
	STAPHYLINIDAE	Aleocharinae 01		3022	20	0.66 [b]
		Aleocharinae 02		85	5	5.88 [a]
PRICKLY PEAR	NITIDULIDAE	*C. mutilatus*	Histiostoma sp. (Astigmata)	57	3	5.26
ORANGE	NITIDULIDAE	*E. luteola*	*Macrochles* sp. (Mesostigmata)	422	3	0.71 [a]
		C. mutilatus		1831	8	0.44 [a]
		U. humeralis		146	1	0.68 [a]
	STAPHYLINIDAE	Aleocharinae 01		3022	29	0.96 [a]

Different letters indicate significant differences in infestation ($p < 0.05$).

The number of mites infesting any given beetle was highly variable. Some were infested with very few mites, while others had vast numbers (Figure 2). While most beetles had less than 10 *Histiostoma* sp. mites, some *E. luteola* had up to 40, and one *C. mutilatus* was found with 271 mites. *Carpophilus mutilatus* had the higher mean mite load ($X = 19.4 \pm 3.52$) that differed statistically from Aleocharinae 01 ($t = -4.966$, $p < 0.0001$), from *E. luteola* ($t = -3.3181$, $p = 0.0011$), and from *U. humeralis* ($t = -4.811$, $p < 0.0001$) (Table 3). *Macrocheles* sp. mite loads were lower than for the astigmatid mite with less than 10 per individual. The mean load was higher in *E. luteola* ($X = 4.67 \pm 3.18$) but it was not significative different between species (Table 3).

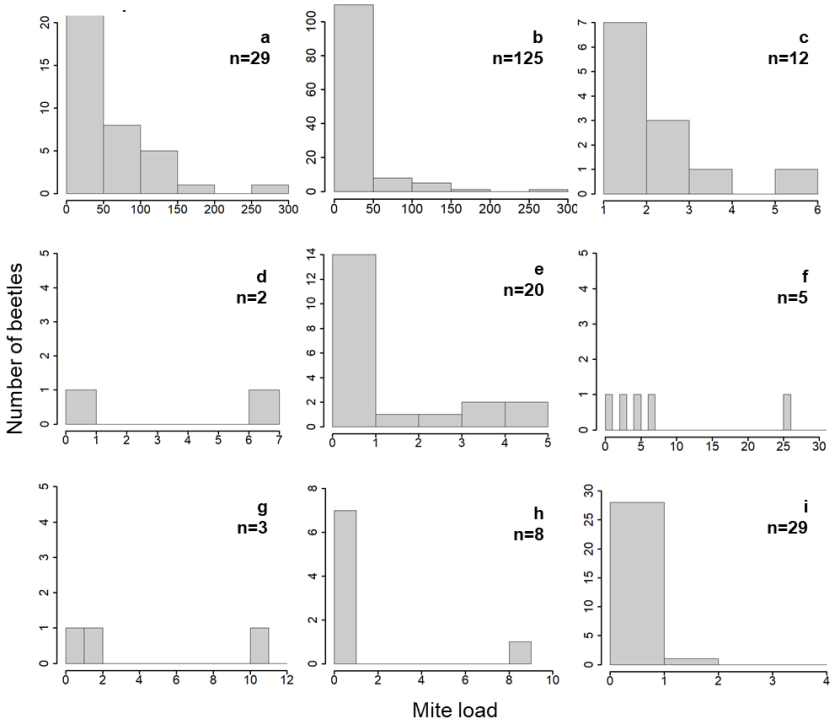


Figure 2. Mite load distributions on beetles, where n = number of infested beetles: a, *E. luteola*; b, *C. mutilatus*; c, *U. humeralis*; d, *S. geminata*; e, Aleocharinae 1; f, Aleocharinae 2; g, *E. luteola*; h, *C. mutilatus*; i, Aleocharinae 1. a-f are *Histiostoma* sp. load distributions and g-i are *Macrocheles* sp. load distributions.

Table 3. Mean mite loads on their beetle hosts and standard deviations (S.E.).

Mite species	Beetle species	Number of beetles	Mean mite load	S.E.
Histiostoma sp.	*E. luteola*	29	6.45 ^{a,b}	1.69
	C. mutilatus	125	19.4 ^{b,c}	3.52
	U. humeralis	12	2.33 ^{a,b}	0.45
	C. hemipterus	1	1.00	-
	S. geminata	2	4.00 ^{a,b,c}	3.00
	Aleocharinae 01	20	1.85 ^{a,b}	0.33
	Aleocharinae 02	5	8.40 ^{a,b,c}	4.52
Macrocheles sp.	*E. luteola*	3	4.67 ^{a}	3.18
	C. mutilatus	8	2.00 ^{a}	1.00
	U. humeralis	1	1.00	-
	Aleocharinae 01	29	1.03 ^{a}	0.03

Different letters are significantly different ($p < 0.05$).

Prevalence of *Histiostoma* sp. in male and female nitidulid beetles varied among species. When analyzed by species, females were more heavily infested in *E. luteola* ($X^2 = 9.14$, df = 1, p = 0.0025) and males in *U. humeralis* ($X^2 = 4.45$, df = 1, p = 0.0349). However, no differences in size within the species exist. No difference in infestation between sexes was detected in *C. mutilatus* ($X^2 = 1.05$, df = 1, p = 0.3055) (Table 4).

Table 4. Number of sexed Niditulidae associated with *Histiostoma* sp. mites.

Beetle species	Males	Females	X^2	P value
E. luteola	6	22	9.14	p = 0.0025**
C. mutilatus	52	63	1.05	p = 0.3055
U. humeralis	9	2	4.45	p = 0.0349*
C. hemipterus		1		
S. geminata	1	1		

The two mite species were not randomly distributed on host body parts (Table 5). The majority of *Histiostoma* sp. mites (44.11 %) were found attaching to the abdomen of the beetle, followed by the thorax (15.36 %) and elytra (12.98 %). Most of *Macrocheles* sp. mites were found attached to legs (55.32 %), abdomen (23.4 %) and thorax (10.64 %). *Macrocheles* sp. attachment, however, was not consistent between species, since it preferred to attach the legs of Aleocharinae 01 but to the abdomen of *C. mutilatus*.

Table 5. Spatial distribution of mites on host body.

Histiostoma sp.									
Site of attachment (# beetles)	*C. mutilatus* (125)	*E. luteola* (29)	*U. humeralis* (12)	*S. geminata* (2)	*C. hemipterus* (1)	Aleocharinae 01 (20)	Aleocharinae 02 (5)	Number of mites	%
Abdomen	1122	55	5			11	10	1203	44.11
Thorax	381	19	2	2		4	11	419	15.36
Elytra	304	38	6			3	3	354	12.98
Prosternum	160	14	4				1	179	6.56
Leg	133	5	7			7	15	167	6.12
Pronotum	110	16	3	3		2	1	135	4.95
Mesosternum	97	16						113	4.14
Head	60	9	1			2	1	73	2.68
Mandible	41	14			1	3		59	2.16
Eye	17	1						18	0.66
Genitalia	1							1	0.04
Antenna	1					5		6	0.22
Macrocheles sp.									
Site of attachment (# beetles)	*C. mutilatus* (8)	*E. luteola* (3)	*U. humeralis* (1)	*S. geminata*	*C. hemipterus*	Aleocharinae 01 (29)	Aleocharinae 02	Number of mites	%
Abdomen	8					3		11	23.40
Thorax	3					2		5	10.64
Elytra	1					1		2	4.26
Leg	2					24		26	55.32
Mesosternum	1		1			0		2	4.26
Head	1					0		1	2.13

In the choice test, where *Histiostoma* sp. mites could choose between two different host species, there no preference among the nitidulids. When a nitidulid and a staphylinid were tested, however, despite the small sample size, the nitidulid was prefered (Table 6). Once a *Histiostoma* sp. selected its host, it did not choose again. This mite spent in average 27:03 ± 7:08 min before choosing a beetle. Despite the variation in time before selecting a host, it did not differ among beetle species (Table 7).

Table 6. Results of choice experiments of the mite *Histiostoma* sp. for Nitidulidae and Staphylinidae beetle species. The sum of times a mite chose a nitidulid or a staphylinid is shown below.

Species to choose		Number of attachments		Number of no attachments
Species 1	Species 2	Species 1	Species 2	
E. luteola	*C. mutilatus*	0	4	3
E. luteola	*U. humeralis*	0	3	1
E. luteola	Aleocharinae	5	0	1
C. mutilatus	*U. humeralis*	1	4	2
C. mutilatus	Aleocharinae	3	0	0
U. humeralis	Aleocharinae	1	0	0
Nitidulidae	Staphylinidae	9	0	1

Values are significantly different ($p < 0.05$).

Table 7. Mean and standard errors for amount of time mites spent before attaching a beetle.

Species to choose		Time to attach (number of attachments)	
Species 1	Species 2	Species 1	Species 2
E. luteola	*C. mutilatus*	0 (0)	16:20 ± 8:30 (3)
E. luteola	*U. humeralis*	0 (0)	31:30 (2)
E. luteola	Aleocharinae	25:00 ± 9:00 (5)	0 (0)
C. mutilatus	*U. humeralis*	6:00 (1)	26:15 ± 5:17 (4)
C. mutilatus	Aleocharinae	30:39 ± 15:38 (3)	0 (0)
U. humeralis	Aleocharinae	74:00 (1)	0 (0)

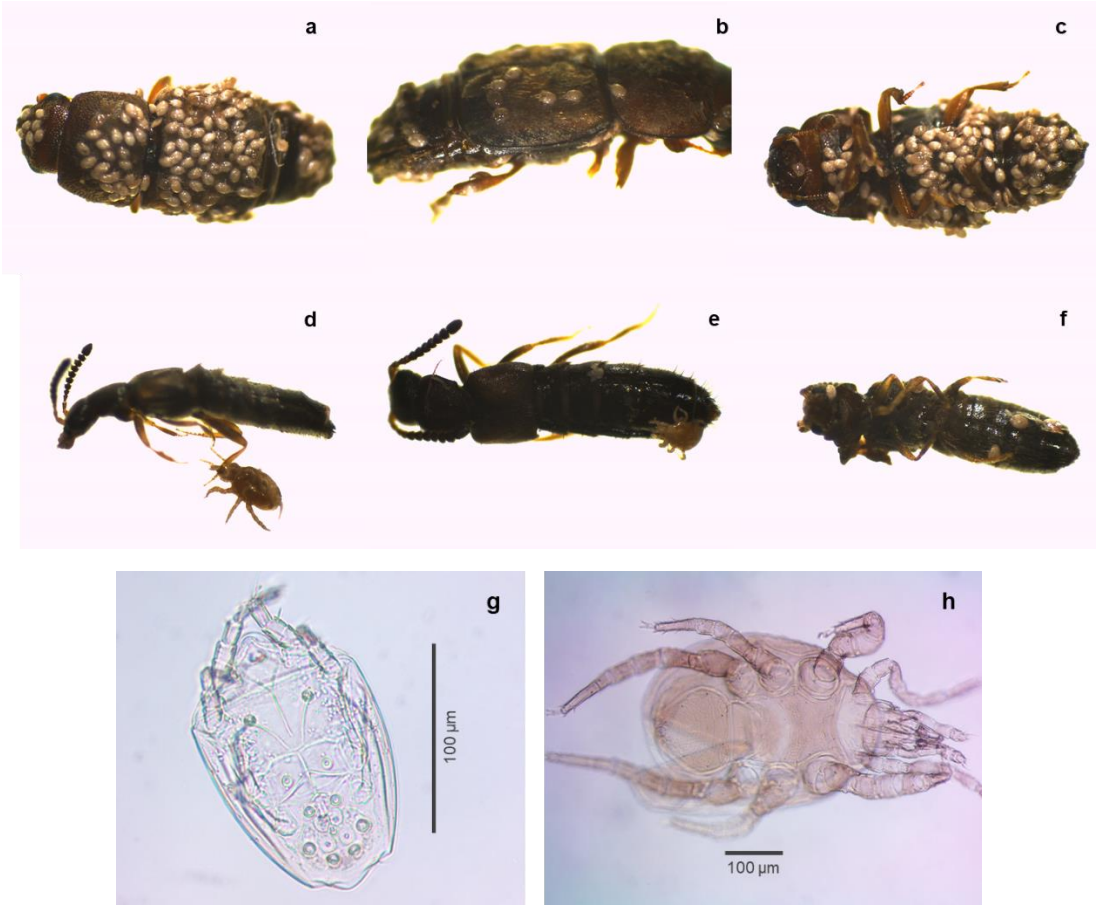


Figure 3. Images of infested beetles and their phoretic mites. **a-c**, *Histiostoma* sp. attached to *C. mutilatus*; **d-e**, Melicharidae attached to Aleocharinae 01; **f**, *Histiostoma* sp. attached to Aleocharinae 01; **g**, *Histiostoma* sp. deutonymph and; **h**, adult *Macrocheles* sp. female.

Discussion

Seven beetle species of the Nitidulidae and Staphylinidae families were infested by two phoretic mites, *Histiostoma* sp. and a *Macrocheles* sp. While the two-mite species infested both coleopteran families, *Histiostoma* sp. preferred to infest nitidulids, while *Macrocheles* sp. did not shown preferences. Additionally, mites differed in their spatial distribution on the host's body.

Mites belong to very different groups within Acari. *Histiostoma* is classified in the Histiostomatidae family that belongs to the Astigmata suborder. Astigmatan mites are one of the most successful in forming phoretic relationships and more commonly found with flies, bees ad beetles (Houck & OConnor, 1991). Astigmatid mites are dispersed by arthropods during their immature stage, the deutonymph, and complete their life cycle when detached from host. I found the deutonymph stage attached to the examined beetles. In addition, when mites were removed from host outer surface, no damage was observed, suggesting only a phoretic relationship. Meanwhile, the *Macrocheles* sp. (Macrochelidae) belongs to the suborder Mesostigmata, which is the most diverse and globally distributed group of mites (Walter & Proctor, 2013). *Macrocheles* species are ubiquitous in ephemeral habits, where they feed on bacteriophagic nematodes and small arthorpods, including fly larvae (Krantz, 1998). Mesostigmatid mites have been found on staphylinid beetles associated with human carrion, included to *Macrocheles glaber*, but not Aleocharinae species (Perotti & Braig, 2009). To my knowledge, this is the first report of Nitidulidae and Staphylinidae beetles associated with *Histiostoma* sp. and the first example of Nitidulidae and Aleocharinae species infested by *Macrocheles* sp.

While several other coleopteran species also were in rotten fruits, only Nitidulidae and Staphylinidae families were infested. However, those families were the most abundant in the samples, with more than 98% of total individuals. The bias in infestation is probably a function of their overrepresentation as they are the most available carriers. The other four nitidulid and six staphylinid species were found without mites, but they were only 0.63% in abundance. Increased sampling may raise the chances of finding additional infested insects.

For example, I found only one *C. hemipterus* infested, but the abundance is minor compared with other species. The remaining beetle families were found without phoretic mites but, as mentioned before, it reflects the small number of individuals collected.

There was a tendency in preference of *Histiostoma* sp. for nitidulid over staphylinid hosts, but there was no preference for a specific species. Despite the large number of Aleocharinae 01, representing more than 50 % of total individuals, they were less infested than nitidulids. In constrast, Aleocharinae 02 were equally infested than nitidulids. Bias in infestation could be due to many factors (chemical, physical or ecological) (Krantz, 1998), for example, differences in size between beetles, since given species vary in their body size. *Histiostoma* sp. was more prevalent in *U. humeralis*, which is the largest beetle, with around 4.7 mm in length and 9 mm^2 of body surface. Moreover, Aleocharinae 01 was the smallest and less infested potential host (2.7 mm in length and 2.5 mm^2 of body surface). Aleocharinae 02 is slightly bigger than Aleocharinae 01 and had higher prevalence (3.3 mm in length, 6 mm^2 of body surface), what fits size hypothesis. Selection of the largest host will increase the chances of success in finding a new habit, new feeding resources or a mate. Previous studies have shown that phoretic mites prefer larger than smaller beetles, that are more likely to increase their fitness. In the case of Hydrophilidae beetles, infestation by *Uropoda orbicularis* (Mesostigmata: Uropodidae) deutonymphs was strongly related to its body size: the largest beetles were occupied by phoretic mites and smaller were uninfested (Bajerlein & Przewoźny, 2012). Likewise, the preference of *Poecilochirus carabi* (Mesostigmata: Parasitidae) for larger *Nicrophorus investigator* (Silphidae) beetles was tested and confirmed on nature and laboratory tests (Grossman & Smith, 2008). Nevertheless, a more extensive analysis is required to confirm the size bias, for example, testing preferences of mites using a broad range of possible hosts that vary in size.

Even with the small number of experiments, choice-tests confirmed the bias of *Histiostoma* sp. mites for nitidulids, since the mite on no occasion chose the Aleocharinae beetles as a host. However, it remains unclear if selection is based on size, chemical or ecological signals. If chemical cues are responsible for nitidulid preferences, those molecules must be similar among related beetles, since no differences between nitidulid species was

observed. The bias in *Macrocheles subbadius* infestation for members of the repleta species group of the subgenus Drosophila may reflect differences in epicuticular hydrocarbons on the fly's surface recognizable by mites (Perez-Leanos et al., 2017). Drosophilid hydrocarbons serve as pheromone attractants for mating recognition (Ferveur, 1997). Many nitidulid species produce aggregation pheromones from a disk located in the abdomen (Dowd & Bartelt, 1993). Those pheromones are hydrocarbons that attract equally males and females and even other sap beetles (Emekci & Moore, 2015; Bartelt, Dowd, Platter, & Weisleder, 1990: Bartelt, Dowd, Vetter, et al., 1992), possibly to find others for mating or food exploitation.

For *Macrocheles* sp., there was no difference in prevalence among families nor between beetle species. Unlike *Histiostoma* sp., *Macrocheles* sp. appears to select it host by its availability, explaining why more were on the most abundant beetles, such as *C. mutilatus*, *E. luteola*, *U. humeralis* and Aleocharinae 01.

As expected, both mite species were usually found in low numbers on host bodies. Too many mites attached to a single host would impair dispersal and be disadvantageous for the mites. Depending upon the number of mites on a host and the distance that host travel with its phoretics, phoresy may have some costs for the host. Despite this, six remarkable *C. mutilatus* individuals were found with more than 100 *Histiostoma* sp. deutonymphs attached and one unusual beetle had almost 300 mites (Figure 3). Secondly, *C. mutilatus* had the highest *Histiostoma* sp. load, with an average of 19.4 mites per individual, a cost is expected. Perhaps, the presence of one mite on a host could attract to others possibly by chemical cues. Moreover, a possibility is that a beetle is attracting many mites because it is more advantageous for dispersion and will increase the establishment of mites at the new site.

The presence of the mites in high numbers and in body parts that can interfere with beetle functions, like flying, vision or reproduction, must have a cost. The presence of *Macrocheles* affected flight aerodynamics of infested flies, by reducing flight time even when mites were one hour previously removed (Luong, Penoni, Horn, et al., 2015). Unfortunately, I was

unable, given time and sample limitations, to identify if a cost exists for examined beetles, but I suspect that it must to occur at least for heavily infested.

Sex bias in *Histiostoma* sp. infestation was observed for *E. luteola* and *U. humeralis*. However, no sexual dimorphism or differences in size has been observed in this species. Within the nitidulids, *Librodor japonicus* (Cryptarchinae) males show bigger mandibles than females (Okada & Miyatake, 2004). Additionally, the number of infested beetles is rather small compared to *C. mutilatus*, where which no sex bias was observed.

Mite distributions on host body were not random and differed between mite species. Here, *Macrocheles* sp. had a strong preference for attaching the legs of Aleocharinae 01, while *Histiostoma* sp. chosen abdomen or even other flat surfaces of *C. mutilatus, E. luteola* and Aleocharinae 01. Because the small number of beetles, there is not a clear evidence of preference for the other species. However, differences could rely on physiological characteristics given by each mite species. Adult *Macrocheles* use their chelicera to attach to its host, thus, the legs are more suitable to bite the beetle, while *Histiostoma* sp. utilize their sucker organs located on the ventral surface to bind the beetle (OConnor, 1982). A flat surface, such as the abdomen, should be more appropriate for deutonymphs to reside on the beetle. My results indicate that *Macrocheles* sp. attaches to Aleocharinae beetles by biting the legs, but it can also use other body parts, perhaps the pulvillis (lobes between the tarsal claws that help the mite bind to a surface). *Histiostoma* sp. prefer flatter surfaces and it is possible that mites found in different sides were exploring the beetle before the attaching. Moreover, the site of attachment could be a form of defense against the host, to avoid mite removal by grooming behavior or chemical defense. For example, *Macrocheles* spp. preferred to attach behind the coxae of the hind legs of the beetles *Nicrophorus* spp., a site less accessible for the beetle to eliminate the mites (Schwarz, Starrach, & Koulianos, 1998). Factors influencing mite distributions warrant more extensive investigation, increasing the number of beetles and experimental trials.

Conclusion

Five Nitidulidae and two Staphylinidae beetles have a phoretic interaction with an astigamatid and a mesostigmatid mites in decaying fruits. The astigmatan, *Histiostoma* sp., infested to *Carpophilus mutilatus*, *Epuraea luteola*, *Urophorus humeralis*, *Stelidota geminata*, Aleocharinae 01 and Aleocharinae 02 during the immature stage deutonymph. *Macrocheles* sp., the mesostigmatan, infested, but as an adult, to *C. mutilatus*, *E. luteola*, *U. humeralis* and Aleocharinae 01. This is the first report of those beetles with phoretic mites. Both phoretic mites have different behavioral preferences for their hosts. *Histiostoma* sp. had a preference for nitidulid species over the staphylinids, while there was not a bias for *Macrocheles* sp. Selection of the host is especially important in phoretic relationships, since on this depends the success in finding a new habit, a mate or food. Several factors must be determining host choice in *Histiostoma* sp. and *Macrocheles* sp. species. Beetle availability must be an important component since the most abundant species were infested. Despite the availability, the largest beetles were infested, which implies that size could be playing an important role in host bias. Additionally, chemical cues, like pheromone hydrocarbons, should be attracting *Histiostoma* sp. to potential nitidulid hosts. Distributions on host body varied among mite species. *Histiostoma* sp. preferred to attach the abdomen of the beetles while *Macrocheles* sp. had a preference for the legs. The pattern of distributions should be result of physiological and behavioral differences between mites, to avoid mechanical or chemical defenses from the host.

References

Andújar, C., Arribas, P., Yu, D. W., Vogler, A. P., & Emerson, B. (2018). Why the COI barcode should be the community DNA metabarcode for the Metazoa. *Molecular Ecology*, 0–3. https://doi.org/10.1111/mec.14844

Bajerlein, D., & Przewoźny, M. (2012). When a beetle is too small to carry phoretic mites? A case of hydrophilid beetles (Coleoptera: Hydrophilidae) and *Uropoda orbicularis* (Acari: Mesostigmata). *Canadian Journal of Zoology*, *90*(3), 368–375. https://doi.org/10.1139/z2012-005

Barrowclough, G. F., & Zink, R. M. (2009). Funds enough, and time: mtDNA, nuDNA and the discovery of divergence. *Molecular Ecology*, *18*, 2934–2936.

Bartelt, R. J., Diana, S. V., Petroskl, R. J., & Baker, T. C. (1995). Pheromone Combination Lures for Carpophilus (Coleoptera : Nitidulidae) Species. *Ecology and Behavior*, *88*(4), 864–869.

Bartelt, R. J., Dowd, P. F., Plattner, R. D., & Weisleder, D. (1990). Aggregation Pheromone of Driedfruit Beetle, Carpophilus hemipterus, Wind-Tunnel Bioassay and Identification of Two Novel Tetraene Hydrocarbons. *Journal of Chemical Ecology, 16*(4), 1015-1039.

Bartelt, R. J., Dowd, P. F., Vetter, R. S., Shorey, H. H., & Baker, T. C. (1992). Responses of Carpophilus hemipterus (Coleoptera: Nitidulidae) and Other Sap Beetles to the Pheromone of C. hemipterus and Host-Related Coattractants in California Field Tests. *Environmental Entomology, 21*(5), 1143-1153

Barton, P. S., Cunningham, S. A., Macdonald, B. C. T., McIntyre, S., Lindenmayer, D. B., & Manning, A. D. (2013). Species Traits Predict Assemblage Dynamics at Ephemeral Resource Patches Created by Carrion. *PLoS ONE*, *8*(1). https://doi.org/10.1371/journal.pone.0053961

Becher, P. G., Flick, G., Rozpedowska, E., Schmidt, A., Hagman, A., Lebreton, S., … Bengtsson, M. (2012). Yeast, not fruit volatiles mediate Drosophila melanogaster attraction, oviposition and development. *Functional Ecology*, *26*(4), 822–828. https://doi.org/10.1111/j.1365-2435.2012.02006.x

Blackmer, J. L., & Phelan, P. L. (1995). Ecological analyses of Nitidulidae: seasonal occurrence, host choice and habitat preference. *Journal of Applied Entomology*, *119*, 312–329. https://doi.org/10.1111/j.1439-0418.1995.tb01294.x

Blackmer, J. L., & Phelan, P. U. (1991). Effect of physiological state and fungal inoculation on chemically modulated host-plant finding by Carpophilus hemipterus and Carpophilus lugubris. *Entomologia Experimentalis et Applicata*, *61*, 33–43.

Borrow, D. J., & White, R. E. (1970). A Peterson Field Guide to Insects: America North of Mexico. Houhton

Mifflin Company.

Camerik, A. M. (2009). Phoresy revisited. *Trends in Acarology: Proceedings of the 12th International Congress*, 333–336. https://doi.org/10.1007/978-90-481-9837-5

Castrezana, S., & Markow, T. A. (2001). Arthropod diversity in necrotic tissue of three species of columnar cacti (Cactaceae). *The Canadian Entomologist*, *133*, 301–309.

Chao, A., Chazdon, R. L., Colwell, R. K., & Shen, T.-J. (2005). A new statistical approach for assessing similarity of species composition with incidence and abundance data. *Ecology Letters*, *8*, 148–159. https://doi.org/10.1111/j.1461-0248.2004.00707.x

Clement, M., Posada, D., & Crandall, K. A. (2000). TCS: A computer program to estimate gene genealogies. *Molecular Ecology*, *9*(10), 1657–1659. https://doi.org/10.1046/j.1365-294X.2000.01020.x

Cline, A. R., Smith, T. R., Miller, K., Moulton, M., Whiting, M., & Audisio, P. (2014). Molecular phylogeny of Nitidulidae: assessment of subfamilial and tribal classification and formalization of the family Cybocephalidae (Coleoptera: Cucujoidea). *Systematic Biology*. DOI: 10.1111/syen.12084; Molecular

Colwell, R. K., & Elsensohn, J. E. (2014). EstimateS turns 20: Statistical estimation of species richness and shared species from samples, with non-parametric extrapolation. *Ecography*, *37*(6), 609–613. https://doi.org/10.1111/ecog.00814

Cross, E. A., & Bohart, G. E. (1969). Phoretic Behavior of Four Species of Alkali Bee Mites as Influenced by Season and Host Sex Author. *Journal of the Kansas Entomological Society*, *42*(2), 195–219.

Date, P., Crowley-Gall, A., Diefendorf, A. F., & Rollmann, S. M. (2017). Population differences in host plant preference and the importance of yeast and plant substrate to volatile composition. *Ecology and Evolution*, *7*(11), 3815–3825. https://doi.org/10.1002/ece3.2993

David, S., & Brown, J. (2009). *Molecular systematics and colour variation of Carpophilus species (Coleoptera: Nitidulidae) of the South Pacific*.

Delgado-Fernández, M., Escobar-Flores, J. G., & Franklin, K. (2017). El cardón gigante (Pachycereus pringlei) y sus interacciones con la fauna en la península de Baja California, México. *Acta Universitaria*, *27*(5), 3–12.

Dowd, P. F., & Bartelt, R. J. (1993). Aggregation pheromone glands of Carpophilus freemani (Coleoptera: Nitidulidae) and gland distribution among other sap beetles. *Annals of the Entomological Society of America, 86*(4), 464-469.

El Kossori, R. L., Villaume, C., El Boustani, E., Sauvaire, Y., & Méjean, L. (1998). Composition of pulp, skin and

seeds of prickly pears fruit (Opuntia ficus indica sp.). *Plant Foods for Human Nutrition*, *52*(3), 263–270. https://doi.org/10.1023/A:1008000232406

Ellis, J. D., Delaplane, K. S., Cline, A., & Mchugh, J. V. (2008). The association of multiple sap beetle species (Coleoptera: Nitidulidae) with western honey bee (Apis mellifera) colonies in North America. *Journal of Apicultural Research*, *47*(3), 188–189. https://doi.org/10.3827/IBRA.1.47.3.04

Emekci, M., & Moore, D. (2015). Sap Beetles. In *Sustainable Pest Management in Date Palm: Current Status and Emerging Challenges* (pp. 205–236). https://doi.org/10.1007/978-3-319-24397-9

Escalante, A., & Benado, M. (1990). Predation on the Cactophilic Fly , Drosophila starmeri , in the Columnar Cactus , Pilosocereus lanuginosus. *Biotropica*, *22*(1), 48–50.

Ewing, C. P., & Cline, A. R. (2005). Key to Adventive Sap Beetles (Coleoptera: Nitidulidae) in Hawaii, with Notes on Records and Habits. *The Coleopterists Bulletin*, *59*(2), 167–183. https://doi.org/10.1649/736

Excoffier, L. and Lischer, H. E. L. (2010). An Integrated Software Package for Population Genetics Data Analysis. *Molecular Ecology Resources*, *10*, 564–567. https://doi.org/10.1111/j.1755-0998.2010.02847.x

Fashing, N. J. (2008). Biology of the genus Hericia (Algophagidae: Astigmata), with the description of a new species from the eastern United States. *Systematic and Applied Acarology*, *13*(1), 3–25. https://doi.org/10.11158/saa.13.1.1

Fellows, D. P., & Heed, W. B. (1972). Factors Affecting Host Plant Selection in Desert-Adapted Cactophilic Drosophila. *Ecology*, *53*(5), 850–858.

Ferro, M. L., Nguyen, N. H., Tishechkin, A., Park, J.-S., Bayless, V., & Carlton, C. E. (2013). Coleoptera Collected from Rotting Fishhook Barrel Cacti (Ferocactus wislizeni (Engelm.) Britton and Rose), with a Review of Nearctic Coleoptera Associated with Succulent Necrosis. *The Coleopterists Bulletin*, *67*(4), 419–443. https://doi.org/10.1649/0010-065X-67.4.419

Ferveur, J. (1997). The pheromonal role of cuticular hydrocarbons in Drosophila melanogaster. *BioEssays, 19*(4), 353-358.

Fogleman, J. C., & Danielson, P. B. (2001). Chemical Interactions in the Cactus-Microorganism- Drosophila Model System of the Sonoran Desert. *American Zoologist*, *41*, 877–889. https://doi.org/10.1093/icb/41.4.877

Folmer, 0, Black, M., Hoeh, W., Lutz, R., & Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, *3*(5), 294–299.

Frost, C. J., & Hunter, M. D. (2004). Insect canopy herbivory and frass deposition affect soil nutrient dynamics and export in oak mesocosms. *Ecology*, *85*(12), 3335–3347.

Fu, Y. X. (1997). Statistical tests of neutrality of mutations against population growth, hitchhiking and background selection. *Genetics*, *147*(2), 915–925. https://doi.org/genetics.org//147/2/915

Ganter, P. F., Morais, P. B., & Rosa, C. A. (2017). Yeasts in Cacti and Tropical Fruit. In *Yeasts in Natural Ecosystems: Diversity* (pp. 225–264). Retrieved from http://eproofing.springer.com/books/mainpage.php?token=UvmqBPOEvphfmkvgP6FwjxcRmF29kLkOLZ_8VjxdcI4

Gotelli, N. J., & Chao A. (2013) Measuring and Estimating Species Richness, Species Diversity, and Biotic Similarity from Sampling Data. In: Levin S.A. (ed) *Encyclopedia of Biodiversity*, second edition (5), 195-211. Waltham, MA: Academic Press.

Gouy, M., Guindon, S., & Gascuel, O. (2010). Sea view version 4: A multiplatform graphical user interface for sequence alignment and phylogenetic tree building. *Molecular Biology and Evolution*, *27*(2), 221–224. https://doi.org/10.1093/molbev/msp259

Grossman, J. D., & Smith, R. J. (2008). Phoretic Mite Discrimination Among Male Burying Beetle (*Nicrophorus investigator*) Hosts. *Annals of the Entomological Society of America, 101*(1), 266-271.

Hamilton, M. B. (2009). Population Genetics. Willey-Blackwell. First edition.

Hayashi, K., Ichikawa, T., & Yasui, Y. (2011). Critical factors in the limited occurrence of the Japanese tree sap mite Hericia sanukiensis (Acari: Astigmata: Algophagidae) inhabiting the sap of the oak Quercus acutissima. *Experimental and Applied Acarology*, *54*(4), 395–409. https://doi.org/10.1007/s10493-011-9463-3

Hebert, P. D. N., Cywinska, A., Ball, S. L., & deWaard, J. R. (2003). Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences*, *270*(1512), 313–321. https://doi.org/10.1098/rspb.2002.2218

Hebert, P., Ratnasingham, S., Zakharov, E. V., Telfer, A. C., Levesque-Beaudin, V., Milton, M. A., … DeWaard, J. R. (2016). Counting animal species with DNA barcodes: Canadian insects. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *371*. https://doi.org/10.1098/rstb.2015.0333

Hedrick, P. (1999). Genetics of Populations. Jones and Barlett. Second edition.

Hernández-Torres, H., García-Martínez, O., Romero-Nápoles, J., Sánchez-Valdez, V. M., Aguirre-Uribe, L. A., & Sánchez-Peña, S. R. (2018). Escarabajos de la Savia de Coahuila , México y Atrayentes Efectivos Para su Recolecta. *Southwestern Entomologist*, *43*(1), 151–166.

Houck, M. A., & OConnor, B. M. (1991). Ecological And Evolutionary Significance Of Phoresy In The Astigmata. *Annual Review of Entomology*, *36*(1), 611–636. https://doi.org/10.1146/annurev.ento.36.1.611

Katlav, A., Hajiqanbar, H., & Talebi, A. A. (2015). First record of the genus Aethiophenax (Acari: Acarophenacidae) from Asia, redefinition of the genus and description of a new species. *Journal of Asia-Pacific Entomology*, *18*(3), 389–395. https://doi.org/10.1016/j.aspen.2015.03.011

Kefford, J. F. (1960). The chemical consituents of Citrus fruits. In C. C. (Ed.) (Ed.), *Advances in Food Research* (p. 285–372.).

Kelebek, H., & Selli, S. (2014). Identification of phenolic compositions and the antioxidant capacity of mandarin juices and wines. *Journal of Food Science and Technology*, *51*(6), 1094–1101. https://doi.org/10.1007/s13197-011-0606-7

Kim, K. C. (1993). Biodiversity, conservation and inventory: why insects matter. *Biodiversity and Conservation*, *2*, 191–214. https://doi.org/10.1007/BF00056668

Krantz, W. G. (1998). Reflections on the biology, morphology and ecology of the Macrochelidae. *Experimental and Applied Acarology*, *22*, 125–137.

Leigh, J. W., & Bryant, D. (2015). POPART: Full-feature software for haplotype network construction. *Methods in Ecology and Evolution*, *6*(9), 1110–1116. https://doi.org/10.1111/2041-210X.12410

Levine, J. M., & HilleRisLambers, J. (2009). The importance of niches for the maintenance of species diversity. *Nature*, *461*(10), 254–258. https://doi.org/10.1038/nature08251

Li, D., Waite, D. W., Fan, Q., George, S., Semeraro, L., & Blacket, M. J. (2018). Molecular detection of small hive beetle Aethina tumida Murray (Coleoptera : Nitidulidae): DNA barcoding and development of a real-time PCR assay. *Scientific Reports*, *8*(February), 1–13. https://doi.org/10.1038/s41598-018-27603-x

Luong, L. T., Penoni, L. R., Horn, C. J., & Polak, M. (2015). Physical and physiological costs of ectoparasitic mites of host flight endurance. *Ecological Entomology, 40*, 518-524.

Majka, C. G., Webster, R., & Cline, A. R. (2008). New records of Nitidulidae and Kateretidae (Coleoptera) from New Brunswick, Canada. *ZooKeys*, *2*, 337–356. https://doi.org/10.3897/zookeys.2.23

Marguran, A. E. (2004). Measuring Biological Diversity. Blackwell Science Ltd.

Markow, T. A., Hanna, G., Riesgo-Escovar, J. R., Tellez-Garcia, A. A., Richmond, M. P., Nazario-Yepiz, N., … Pfeiler, E. (2014). Population genetics and recent colonization history of the invasive drosophilid Zaprionus indianus in Mexico and Central America. *Biological Invasions*, *16*, 2427–2434. https://doi.org/10.1007/s10530-014-0674-5

Mejía, D. (2016). *The Diversity, Distribution, and Ecology of Rove Beetles (Staphylinidae) in the Sonoran Desert Necrotic Cactus Niche*.

Miller, M. W., & Mrak, E. M. (1953). Yeasts associated with dried-fruit beetles in figs. *Applied Microbiology*, *1*(4), 174–178. https://doi.org/10.1001/jama.1909.02540490052022

Mimura, M., Yahara, T., Faith, D. P., Vï¿½zquez-Domï¿½nguez, E., Colautti, R. I., Araki, H., … Hendry, A. P. (2017). Understanding and monitoring the consequences of human impacts on intraspecific variation. *Evolutionary Applications*, *10*(2), 121–139. https://doi.org/10.1111/eva.12436

Mutinelli, F., Federico, G., Carlin, S., Montarsi, F., & Audisio, P. (2016). Preliminary investigation on other Nitidulidae beetles species occurring on rotten fruit in Reggio Calabria province (South-western Italy) infested with small hive beetle (Aethina tumida). *Journal of Apicultural Research*, *54*(3), 233–235. https://doi.org/10.1080/00218839.2016.1142733

Naskrecki, P., & Colwell, R. K. (1995). New Genus and Two New Species of Melicharini From Venezuela (Acari, Mesostigmata, Ascidae). *Annals of the Entomological Society of America*, *88*(3), 284–293. Retrieved from http://apps.webofknowledge.com.ezproxy.stanford.edu/full_record.do?product=UA&search_mode=GeneralSearch&qid=74&SID=2F7yDDP6mVSa1XzqU2h&page=1&doc=9%5Cn%3CGo to ISI%3E://A1995QY12800005

Navarrete-Heredia, J. L., Newton, A. F., Thayer, M. K., Ashe, J. S., & Chandler, D. S. (2002). *Guía illustrada para los géneros de Staphylinidae (Coleoptera) de México. Illustrated guide to the genera of Staphylinidae (Coleoptera) of Mexico. Universidad de Guadalajara y CONABIO, México.*

Newton, A. F. (2015). Beetles (Coleoptera) of Peru: A Survey of the Families. Staphylinidae Latreille, 1802. *Journal of the Kansas Entomological Society*, *88*(2), 283–304. https://doi.org/10.2317/kent-88-02-283-304.1

Nunney, L. (1990). Drosophila on oranges: colonization, competition, and coexistence. *Ecology*, *71*(5), 1904–1915. https://doi.org/10.2307/1937598

OConnor, B. M. (1982). Evolutionary Ecology of Astigmatid Mites. *Annual Review of Entomology*, *27*(1), 385–409. https://doi.org/10.1146/annurev.en.27.010182.002125

Okada, K. & Miyatake, T. (2004). Sexual Dimorphism in Mandibles and Male Aggressive Behavior in the Presence and Absence of Females in the Beetle Librodor japonicus (Coleoptera: Nitidulidae). *Annals of the Entomological Society of America, 97*(6), 1342-1346.

Perez-Leanos, A., Loustalot-Laclette, M. R., Nazario-Yepiz, N., & Markow, T. A. (2017). Ectoparasitic mites and

their Drosophila hosts. *Fly*, *11*(1), 10–18. https://doi.org/10.1080/19336934.2016.1222998

Perotti, M. A., & Braig, H. R. (2009). Phoretic mites associated with animal and human decomposition. *Experimental and Applied Acarology*, *49*(1–2), 85–124. https://doi.org/10.1007/s10493-009-9280-0

Pfeiler, E., Bitler, B. G., Castrezana, S., Matzkin, L. M., & Markow, T. A. (2009). Genetic diversification and demographic history of the cactophilic pseudoscorpion Dinocheirus arizonensis from the Sonoran Desert. *Molecular Phylogenetics and Evolution*, *52*(1), 133–141. https://doi.org/10.1016/j.ympev.2008.12.020

Pfeiler, E., & Markow, T. A. (2011). Phylogeography of the cactophilic Drosophila and other arthropods associated with cactus Necroses in the Sonoran Desert. *Insects*, *2*(2), 218–231. https://doi.org/10.3390/insects2020218

Reese, E. M., & Swanson, A. P. (2017). A Review of the Cactophilic Carcinops Marseul (Coleoptera: Histeridae) of the Sonoran Desert Region, with Descriptions of Six New Species. *The Coleopterists Bulletin*, *71*(1), 159–190. https://doi.org/10.1649/0010-065X-71.1.159

Reichle, D. E. (1977). The Role of Soil Invertebrates in Nutrient Cycling. *Ecological Bulletins*, *25*, 145–156. Retrieved from http://www.jstor.org

Richmond, M. P., Reese, E. M., Mejía, D., & Markow, T. A. (n.d.). Arthropod community diversity in the ecological niche associated with necrotic cactus in the Sonoran Desert. *In Revision*, 1–32.

Root, R. B. (1967). The Niche Exploitation Pattern of the Blue-Gray Gnatcatcher. *Ecological Monographs*. https://doi.org/10.2307/1942327

Rozas, J., Ferrer-Mata, A., Sánchez-DelBarrio, J. C., Guirao-Rico, S., Librado, P., Ramos-Onsins, S. E., & Sánchez-Gracia, A. (2017). DnaSP 6: DNA Sequence Polymorphism Analysis of Large Data Sets. *Molecular Biology and Evolution*, *34*(12), 3299–3302. https://doi.org/10.1093/molbev/msx248

Ruiz, A., & Heed, W. B. (1988). Host-Plant Specificity in the Cactophilic Drosophila mulleri Species Complex. *Source: Journal of Animal Ecology Journal of Animal Ecology*, *57*(57), 237–249. Retrieved from http://www.jstor.org/stable/4775

Salim, N., Abdelwaheb, C., Rabah, C., & Ahcene, B. (2009). Chemical composition of Opuntia ficus-indica (L.) fruit. *African Journal of Biotechnology*, *8*(8), 1623–1624. Retrieved from http://www.academicjournals.org/AJB

Scherer-Lorenzen, M. (2008). Functional diversity affects decomposition processes in experimental grasslands. *Functional Ecology*, *22*(3), 547–555. https://doi.org/10.1111/j.1365-2435.2008.01389.x

Schwarz, H., Starrach, M., & Koulianos, S. (1998). Host specificity and permanence of associations between mesostigmatic mites (Acari: Anactinotrichida) and burying beetles (Coleoptera: Silphidae: Nicrophorus). *Journal of Natural History, 32*(2), 159-172. DOI: 10.1080/00222939800770101

Skoracka, A., Dabert, M. (2010). The cereal rust mite *Abacarus hystrix* (Acari: Eriophyoidea) is a complex of species: evidence from mitochondrial and nuclear DNA sequences. *Bulletin of Entomological Research*, 100(3), 263-272.

Starmer, W. T., Lachance, M.-A., & Phaff, H. J. (1987). A comparison of yeast communities found in necrotic tissue of cladodes and fruits of Opuntia stricta on Islands in the Caribbean Sea and where introduced into Australia. *Microbial Ecology*, *14*, 179–192. https://doi.org/10.1007/BF02013022

Stintzing, F. C., Herbach, K. M., Mosshammer, M. R., Carle, R., Yi, W., Selllappan, S., … Felker, P. (2005). Color, Betalain Pattern, and Antioxidant Properties of Cactus Pear (Opuntia spp.) Clones. *Journal of Agricultural and Food Chemistry*, *53*, 442–451. https://doi.org/10.1021/jf048751y

Stork, N. E., Mcbroom, J., Gely, C., & Hamilton, A. J. (2015). New approaches narrow global species estimates for beetles , insects , and terrestrial arthropods, *112*(24), 7519–7523. https://doi.org/10.1073/pnas.1502408112

Swift, M. J., Heal, O. W., & Anderson, J. M. (1979). Decomposition in Terrestrial Ecosystems. Studies in Ecology. Vol(5). Blackwell, Oxford.

Tajima, F. (1989). Statistical method for testing the neutral mutation hypothesis by DNA polymorphism. *Genetics*, *123*(3), 585–595. https://doi.org/PMC1203831

Walter, D. E., & Proctor, H. C. (2013). *Mites: Ecology , Evolution & Behaviour* (Second Edi). Springer.

Whitford, W. G., & Parker, L. W. (1989). Contributions of soil fauna to decomposition and mineralisation processes in semiarid and arid ecosystems. *Arid Soil Research and Rehabilitation*, *3*(April 2015), 199–215. https://doi.org/10.1080/15324988909381199

Wright, G. A. (2015). Olfaction: Smells Like Fly Food. *Current Biology*, *25*(4), R144–R146. https://doi.org/10.1016/j.cub.2014.12.052

Zanetti, N. I., Visciarelli, E. C., & Centeno, N. D. (2015). Trophic roles of scavenger beetles in relation to decomposition stages and seasons. *Revista Brasileira de Entomologia*, *59*(2), 132–137. https://doi.org/10.1016/j.rbe.2015.03.009

Zhang, Z. Q. (2013). Phylum Athropoda. In: Zhang, Z.-Q. (Ed.). *Animal Biodiversity: An Outline of Higher-Level Classification and Survey of Taxonomic Richness (Addenda 2013)*, *3703*(1), 17–26. https://doi.org/10.11646/zootaxa.3703.1.6

Supplementary material

Table S1. Sites and dates of sampling.

Date	Fruit type	Location	Latitude	Longitude
01/09/2016	Prickly pear	Los Nicolases, Guanajuato	20831127	-101319550
05/09/2016	Prickly pear	Celaya-Dolores Hidalgo highway, San Miguel de Allende, Guanajuato	20910060	-100759341
07/09/2016	Prickly pear	Cerro de Arandas, Irapuato, Guanajuato	20725849	-101404323
07/09/2016	Prickly pear	San Miguel de Allende, Guanajuato	20.6737777	-103.405454
14/09/2016	Prickly pear	Aldama, Guanajuato	208369599	-1013155596
26/09/2016	Prickly pear	Highway Irapuato-Guanajuato	20890284	-1013512558
25/09/2016	Prickly pear	Los Nicolases, Guanajuato	20831127	-101319550
25/09/2016	Prickly pear	Cerro de Arandas, Irapuato, Guanajuato	20725849	-101404323
25/09/2016	Prickly pear	Aldama, Guanajuato	208369599	-1013155596
25/09/2016	Prickly pear	Irapuato-Guanajuato highway, Guanajuato	20890284	-1013512558
03/04/2017	Orange	Irapuato, Guanajuato	206924393	-10135603
24/04/2017 - 29/04/2017	Orange	Navojoa, Sonora	270719761	-109309379
17/05/2017	Orange	Guanajuato, Guanajuato	21.0016861	-101.286105
23/05/2017	Orange	Guanajuato, Guanajuato	21.0016861	-101.286105
09/06/2017	Orange	Irapuato, Guanajuato	206924393	-10135603
12/06/2017	Orange	Irapuato, Guanajuato	206924393	-10135603
24/06/2017	Orange	Irapuato, Guanajuato	206924393	-10135603
01/07/2017	Orange	Guadalajara, Jalisco	20.6737777	-103.405454
19/08/2017	Prickly pear	Los Nicolases, Guanajuato	20831127	-101319550
21/09/2017	Prickly pear	Los Nicolases, Guanajuato	20831127	-101319550
24/09/2017	Prickly pear	San Miguel de Allende (Charco del Ingenio), Guanajuato	20.9177411	-100.729609
04/10/2017	Prickly pear	Irapuato, Guanajuato	20.7232423	-101.346188
05/10/2017	Prickly pear	Irapuato, Guanajuato	20.7232423	-101.346188
07/10/2017	Prickly pear	San Juan de los Lagos - Lagos de Moreno highway, Jalisco	21,309,236	-102,091,169
09/10/2017	Orange	Navojoa, Sonora	270719761	-109309379
11/10/2017	Prickly pear	Navojoa, Sonora	270719761	-109309379
16/10/2017	Orange	Irapuato, Guanajuato	20.7232423	-101.346188
24/11/2017	Orange	Irapuato, Guanajuato	20.7232423	-101.346188
01/11/2017	Prickly pear	Batacosa, Sonora	20.678565	-101.348045
11/12/2017	Orange	Irapuato, Guanajuato	20.7232423	-101.346188
15/03/2018	Orange	Navojoa, Sonora	270719761	-109309379
04/08/2018	Orange	Irapuato, Guanajuato	20.7232423	-101.346188
24/08/2018	Prickly pear	Batacosa, Sonora	20.678565	-101.348045

Table S2. Taxonomic diversity and abundance of adult arthropods found in decaying prickly pears and orange fruits and number of fruits.

										Prickly pears			Oranges			
									Number of fruits	126	77	13	48	134	-	398
Class	Order	Suborder	Superfamily	Family	Subfamily	Genus	Species	Current name	Guild	Guanajuato	Sonora	Jalisco	Guanajuato	Sonora	Jalisco	Overall
Insecta	Coleoptera	Adephaga	Caraboidea	Carabidae	Harpalinae	Lebia	natalensis	Lebia natalensis	Predator	0	0	0	0	1	0	1
Insecta	Coleoptera	Adephaga	Caraboidea	Carabidae	Harpalinae	Lebia	grandis	Lebia grandis	Predator	0	1	0	0	0	0	1
Insecta	Coleoptera	Adephaga	Caraboidea	Carabidae				Carabidae	Predator	0	1	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Epuraeinae	Epuraea	luteola	Epuraea luteola	Saprophagous	12	7	0	45	290	0	354
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	hemipterus	Carpophilus hemipterus	Saprophagous	12	0	0	84	15	0	111
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	mutilatus	Carpophilus mutilatus	Saprophagous	0	58	0	14	1233	0	1305
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	funebris	Carpophilus funebris	Saprophagous	10	1	0	48	1	0	60
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	lugubris	Carpophilus lugubris	Saprophagous	2	0	0	12	0	0	14
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	delkeskampi	Carpophilus delkeskampi	Saprophagous	0	0	0	0	1	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	nepos	Carpophilus nepos	Saprophagous	0	0	0	377	1	0	378
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	sp.	Carpophilus (Caplothorax) sp.	Saprophagous	0	0	0	2	0	0	2
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	sp.	Carpophilus sp. 1	Saprophagous	0	0	0	4	1	0	5
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Carpophilus	sp.	Carpophilus sp. 2	Saprophagous	0	0	0	0	1	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae	Urophorus	humeralis	Urophorus humeralis	Saprophagous	6	0	0	32	93	0	131
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Carpophilinae			Carpophilininae	Saprophagous	0	0	0	76	0	0	76
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Conotelus	mexicanus	Conotelus mexicanus	Saprophagous	1	3	0	43	5	0	52
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Colopterus	denticulatus	Colopterus denticulatus	Saprophagous	8	0	0	275	0	0	283
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Colopterus	posticus	Colopterus posticus	Saprophagous	4	0	0	218	0	0	222
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Colopterus	truncatus	Colopterus truncatus	Saprophagous	1	0	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Colopterus	macropterus	Colopterus macropterus	Saprophagous	11	0	1	125	0	0	137
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Cillaeinae	Colopterus	sp.	Colopterus sp.	Saprophagous	0	0	0	236	0	0	236
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Lobiopa	insularis	Lobiopa insularis	Saprophagous	33	0	0	4	0	0	37
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Stelidota	geminata	Stelidota geminata	Saprophagous	148	2	6	6	142	11	315
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Stelidota	sp.	Stelidota sp.1	Saprophagous	2	0	0	0	1	0	3
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Stelidota	sp.	Stelidota sp.2	Saprophagous	0	0	0	0	1	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Stelidota	sp.	Stelidota sp.3	Saprophagous	1	0	1	0	0	0	2
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Aethina	villosa	Aethina villosa	Saprophagous	153	0	0	0	0	0	153
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae	Aethina	sp.	Aethina sp.	Saprophagous	1	0	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Nitidulidae	Nitidulinae			Nitidulinae	Saprophagous	2	0	0	0	0	0	2
Insecta	Coleoptera	Polyphaga	Cucujoidea	Mycetophagidae		Litargus	sp.	Litargus sp.	Saprophagous	1	7	0	66	2	0	76
Insecta	Coleoptera	Polyphaga	Cucujoidea	Coccinellidae				Coccinellidae	Predator	1	0	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Cucujoidea	Silvanidae	Silvaninae	Ahasverus	rectus	Ahasverus rectus	Saprophagous	0	1	0	3	4	0	8
Insecta	Coleoptera	Polyphaga	Cucujoidea	Monotomidae	Monotominae	Europs	sp.	Europs sp.	Saprophagous	4	0	0	52	2	0	58
Insecta	Coleoptera	Polyphaga	Hydrophiloidea	Histeridae	Dendrophilinae	Carcinops	consors	Carcinops consors	Predator	0	0	0	0	4	0	4
Insecta	Coleoptera	Polyphaga	Scarabaeidae	Scarabaeidae	Cetoniinae	Cotinis	sp.	Cotinis sp.	Saprophagous	1	0	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Scarabaeidae	Scarabaeidae				Scarabaeidae	Saprophagous	4	0	0	0	0	0	4

Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 1	Saprophagous	0	1	2	1094	1786	6	2889
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 2	Saprophagous	0	0	0	0	38	0	38
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 3	Saprophagous	0	0	0	3	1	0	4
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 4	Saprophagous	0	1	0	2	0	0	3
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 5	Saprophagous	0	0	0	1	2	0	3
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Aleocharinae			Aleocharinae 6	Saprophagous	0	0	0	0	1	0	1
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Staphylininae	Belonuchus	apiciventris	Belonuchus apiciventris	Predator	5	0	0	0	2	0	7
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Staphylininae	Belonuchus	godmani	Belonuchus godmani	Predator	1	0	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Staphylininae	Belonuchus	sp.	Belonuchus sp.	Predator	2	0	0	0	0	0	2
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Staphylininae			Staphylininae 1	Predator	0	0	0	0	4	0	4
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae				Staphylinidae 2	-	0	0	0	2	0	0	2
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Tachyporinae	Coproporus	hepaticus	Coproporus hepaticus	Saprophagous	8	0	0	0	0	0	8
Insecta	Coleoptera	Polyphaga	Staphylinoidea	Staphylinidae	Tachyporinae			Tachyporinae 1	Omnivorous	0	1	0	0	0	0	1
Insecta	Coleoptera	Polyphaga	Tenebrionoidea	Zopheridae	Colydiinae	Bitoma	sp.	Bitoma sp.	Saprophagous	0	0	0	0	2	0	2
Insecta	Coleoptera	Polyphaga	Tenebrionoidea	Tenebrionidae				Tenebrionidae 1	Saprophagous	0	0	0	0	1	0	1
Insecta	Coleoptera	Polyphaga	Tenebrionoidea	Tenebrionidae				Tenebrionidae 2	Saprophagous	0	0	0	0	2	0	2
Insecta	Coleoptera	Polyphaga	Tenebrionoidea	Tenebrionidae				Tenebrionidae 3	Saprophagous	0	0	0	0	4	0	4
Insecta	Coleoptera							Coleoptera 1	Saprophagous	0	0	0	3	0	0	3
Insecta	Coleoptera							Coleoptera 2	-	1	0	0	0	0	0	1
Insecta	Coleoptera							Coleoptera 3	-	2	0	0	0	0	0	2
Insecta	Coleoptera							Coleoptera 4	-	1	0	0	0	0	0	1
Insecta	Diptera	Brachycera	Ephydroidea	Drosophilidae	Drosophilinae	Drosophila	Sophophora (subgenus)	Sophophora (subgenus)	Saprophagous	48	89	2	211	120	0	470
Insecta	Diptera	Brachycera	Ephydroidea	Drosophilidae	Drosophilinae	Drosophila	Repleta (subgroup)	Repleta (subgroup)	Saprophagous	23	15	2	63	87	0	190
Insecta	Diptera	Brachycera	Ephydroidea	Drosophilidae	Drosophilinae	Zaprionus	sp.	Zaprionus sp.	Saprophagous	47	2	0	172	10	0	231
Insecta	Diptera	Calyptratae	Muscoidea	Muscidae				Muscidae	Saprophagous	0	0	0	0	1	0	1
Insecta	Diptera	Nenmatocera		Culicidae				Mosquito	-	0	0	0	2	0	0	2
Insecta	Diptera							Diptera 1	-	13	1	0	2	0	0	16
Insecta	Diptera							Diptera 2	-	1	0	0	0	0	0	1
Insecta	Diptera							Diptera 3	-	0	0	0	0	5	0	5
Insecta	Diptera							Diptera 4	-	0	0	0	0	12	0	12
Insecta	Dermaptera	Forficulina		Anisolabididae		Euborellia	sp.	Euborellia sp.	Omnivorous	1	1	0	8	5	6	21
Insecta	Dermaptera	Forficulina		Forficulidae				Forficulidae	Omnivorous	1	0	0	0	2	0	3
Insecta	Dermaptera							Dermaptera	Omnivorous	1	0	0	0	0	0	1
Insecta	Hemiptera	Heteroptera		Anthocoridae		Orius	sp.	Orius sp.	Predator	0	1	0	2	12	0	15
Insecta	Hemiptera	Heteroptera		Largidae		Stenomacra	marginella	Stenomacra marginella	Saprophagous	0	0	0	3	0	0	3
Insecta	Hemiptera	Heteroptera						Heteroptera 1	-	1	0	0	0	0	0	1
Insecta	Hemiptera	Heteroptera						Heteroptera 2	-	4	0	0	0	0	0	4
Insecta	Hemiptera	Heteroptera		Cydnidae				Cydnidae	Saprophagous	1	0	0	0	0	0	1
Insecta	Hymenoptera	Apocrita	Ichneumonoidea	Braconidae				Braconidae	Parasitoid	5	0	0	0	0	0	5
Insecta	Hymenoptera	Apocrita	Cynipoidea	Figitidae				Figitidae	Parasitoid	6	0	0	13	0	0	19
Insecta	Hymenoptera	Apocrita	Proctotrupidae	Proctotrupidae				Proctotrupidae	Parasitoid	1	0	0	7	3	0	11
Insecta	Hymenoptera	Aculeata	Vespoidea	Vespidae	Polistinae	Polistes	fuscatus	Polistes fuscatus	Parasitoid	0	0	0	1	0	0	1
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae	Myrmicinae	Atta	sp.	Atta sp.	Saprophagous	12	0	0	0	1	0	13
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 1	Saprophagous	3	0	0	0	15	0	18
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 2	Saprophagous	0	0	0	1	0	0	1
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 3	Saprophagous	0	0	0	1	0	0	1
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 4	Saprophagous	0	0	0	0	0	0	0
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 5	Saprophagous	39	0	0	0	0	0	39

Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 6	Saprophagous	8	0	0	0	0	0	8
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 7	Saprophagous	17	0	0	0	0	0	17
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 8	Saprophagous	0	134	0	0	159	0	293
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 9	Saprophagous	0	5	0	0	13	0	18
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 10	Saprophagous	0	0	0	0	174	0	174
Insecta	Hymenoptera	Aculeata	Formicoidea	Formicidae				Formicidae 11	Saprophagous	0	0	0	0	0	3	3
Insecta	Lepidoptera							Lepidoptera	Saprophagous	1	0	0	0	0	0	1
Insecta	Neuroptera			Chrysopidae				Neuroptera	Omnivorous	0	2	0	0	0	0	2
Insecta	Thysanoptera							Thysanoptera	Omnivorous	2	0	0	3	0	0	5
Insecta								Insecta 1	-	1	0	1	0	0	0	2
Insecta								Insecta 2	-	1	0	0	0	0	0	1
Collembola	Entomobryomorpha		Entomobryoidea	Entomobryidae	Lepidocyrtinae	Seira	sp.	Seira sp.	Saprophagous	1	0	0	0	4	0	5
Arachnida	Pseudoscorpiones							Pseudoscorpiones 1	Predator	1	0	0	0	0	0	1
Arachnida	Pseudoscorpiones							Pseudoscorpiones 2	Predator	0	0	1	0	0	0	1
Arachnida	Pseudoscorpiones							Pseudoscorpiones 3	Predator	0	0	0	0	1	0	1
Arachnida	Opiliones							Opiliones	Omnivorous	4	0	0	0	0	0	4
Arachnida	Araneae		Araneomorphae	Salticidae				Salticidae 1	Predator	0	0	0	3	0	0	3
Arachnida	Araneae		Araneomorphae	Salticidae				Salticidae 2	Predator	0	0	0	1	0	0	1
Arachnida	Araneae		Araneomorphae	Salticidae				Salticidae 3	Predator	0	0	0	1	0	0	1
Arachnida	Araneae							Araneae 1	Predator	0	0	0	0	1	0	1
Arachnida	Araneae							Araneae 2	Predator	1	0	0	0	0	0	1
Arachnida	Araneae							Araneae 3	Predator	1	0	0	0	0	0	1
Arachnida	Araneae							Araneae 4	Predator	0	0	0	0	1	0	1
Arachnida	Araneae							Araneae 5	Predator	0	0	0	2	0	0	2
Arachnida	Araneae							Araneae 6	Predator	1	0	0	0	0	0	1
Arachnida	Araneae							Araneae 7	Predator	0	0	0	1	0	0	1
Arachnida	Araneae							Araneae 8	Predator	0	0	0	1	0	0	1
Arachnida	Acari	Astigmata		Histiostomatide		Histiostoma	sp.	Histiostoma sp.	Saprophagous	7	24	0	0	2808	0	2839
Arachnida	Acari	Mesostigmata		Macrochelidae		Macrocheles	sp.	Macrocheles sp.	Saprophagous	0	0	0	0	178	0	178
Arachnida	Acari							Acari 1	Saprophagous	18	8	0	1	41	0	68
Arachnida	Acari							Acari 2	Saprophagous	1	0	0	0	0	0	1
Crustaceae	Oniscidea							Oniscidea 1	Saprophagous	0	0	0	9	0	0	9
Crustaceae	Oniscidea							Oniscidea 2	Saprophagous	0	0	0	0	22	0	22
										710	366	16	3335	7316	26	11769