CEC California Ecology and Conservation Research

Natural Reserve System UNIVERSITY OF CALIFORNIA

Creosote (*Larrea tridentata*) influences on canopy and ground dwelling invertebrate communities

Sindhu Bala[1], Lyra Martin[2], Ashley Penaloza[2]

[1]University of California, Davis; [2]University of California, Santa Cruz

ABSTRACT

The morphology and composition of shrubs greatly affects invertebrate communities, and it affects canopy and ground invertebrates in different ways. The number of flowers, in particular, is tied to plant productivity: more flowers means a greater shrub productivity which is attractive to invertebrates. Furthermore, invertebrate abundance and community structure will change depending on the time of day, since day and night conditions attract different species of invertebrates. In the Mojave Desert, a particularly important shrub is the Creosote (*Larrea tridentata*), as it is a common species that fosters many invertebrates, with a number of them being specialists. We used pitfall traps and bush beatings to determine if the number of invertebrates on and under creosote bushes depends on the time of day and percent flower abundance. We found that invertebrates were more common on the bushes at night and under them during the day, which implied that they moved to cooler areas during the hottest part of the day and foraged at night. We also found that increasing percent flower abundance increased the number of invertebrates on but not under creosotes, most likely because of the rewards that flowers provide for insects like pollinators and florivores.

Keywords: Larrea tridentata, creosote bush, day vs night, invertebrates, Mojave Desert

INTRODUCTION

Shrubs provide vital habitats for many organisms. These habitats are unique from their surrounding environments because the morphological aspects of the shrubs shape their immediate surroundings, which allows them to provide a number of different niches (Badano et al. 2016). The invertebrate communities in and around shrubs are often particularly complex (Jiménez-Valverde 2005). The parts of the bushes these invertebrates primarily live in are on the ground under the shrub and in the canopy of a shrub. They are often highly adapted to these areas (Jiménez-Valverde 2005).

A strong influence on ground cover invertebrate communities found under the shrubs is the microclimate that shrubs create. Shrubs influence understory soil characteristics by increasing the availability of nitrogen, decreasing ambient and ground temperatures, and blocking solar radiation (Moro et al. 1997). The existence of this microclimate greatly increases invertebrate community abundance (Zuliani et al. 2021).

Different invertebrates are found in the shrub canopies than under the shrubs (Jiménez-Valverde 2005), and these canopy invertebrates are influenced by different bush conditions than the invertebrates on the ground. For example, invertebrates that live in the canopy cover are more directly influenced by the shrub productivity and if it is flowering or not. Increased bush productivity, which is the amount of living growth on a plant, can lead to increased invertebrate density, particularly in arthropods (Lightfoot and Whitford 1991). Increased productivity is also correlated with increasing flower abundance, which can attract invertebrates such as pollinators and florivores by indicating a larger foraging reward (Minckley et al. 1999, Braun and Lortie 2020).

The microclimates and conditions that affect ground cover invertebrates and canopy invertebrates are not static. For example, the invertebrate communities often shift significantly over the course of the day and night. (Ramírez-Hernández et al. 2021). Changes in light availability, temperature, and soil moisture depending on the time of day alter which invertebrates are able to operate and which are forced to retreat to relative shelter (Ramírez-Hernández et al. 2021). This is particularly important in deserts, where all three of these factors are felt in extremes. In addition, increases and decreases in shrub size change the intensity of the microclimate affects and have led to variations in invertebrate communities. For example, Increases in shrub sizes can lead to increases in invertebrate density in general (Crisp et al. 1998) and arthropods in particular (Sanchez and Parmenter 2001).

Certain plants in particular are known to be huge drivers of local invertebrate diversity (Little et al. 2001). The creosote bush (Larrea tridentata), which is a dominant shrub in the southwestern deserts of the United States, is one such example. Like other shrubs, Creosote provides food and nutrients, temperature mediation, and pollination benefits (Ruttan 2016, Braun and Lortie 2020, Minckley et al. 1999). Furthermore, the abundance, richness, and diversity of insects under creosote bushes is significantly greater than in an open environment (Ruttan 2016). In many cases, the habitats provided by creosote bushes are more important than the rest of the environment combined on invertebrate presence. For instance, when creosote bushes move out of their native desert environments and into grasslands, their invertebrate communities will oftentimes move with them (Sanchez and Parmenter 2001).

One reason for the large influence that creosote has on invertebrate communities, is that creosote causes specialization within the invertebrate communities through its chemical defenses against herbivory (Rhoades and Cates 1976). Creosote bushes have large quantities of diethyl ether soluble phenolic resin on the surface of its leaves and at its terminal stem portions. Although this resin deters many insects, some invertebrate specialists have become adapted to it, and it even acts as an attractant to specialists such as Semiothisa (Rhoades and Cates 1976). Therefore, many invertebrates like the many species of arthropods and over 20 species of native bee (Sanchez and Parmenter 2001, Minckley et al. 1999) rely on creosote as a vital habitat.

Though the invertebrate communities of creosotes are well characterized, it is still unclear how the creosote's morphological features affect their canopy and ground invertebrate communities separately, particularly in relation to day and night time shifts. In order to address this, we examined the effects of flower number, dead growth, bush circumference, and time of day on both ground and canopy creosote invertebrate communities. We expected to see differences in invertebrate communities depending on whether it was day or night. Specifically, we thought there would be a higher density of insects under the creosote and in the canopy of the creosote during the day to avoid the worst of the heat. (Ramírez-Hernández et al. 2021). We expected greater flower abundance to increase the number of invertebrates in the canopy but not on the ground, as more flowers would primarily affect the invertebrates that interact with them. Furthermore, we expected that an increase in bush size would lead to a greater amount of shade and moisture accumulation, and therefore increase invertebrate density under the bushes but not in the canopy since the effect would be primarily on the ground cover microclimate. Finally, we expected to see that an increase in the percentage of a bush consisting of alive growth would increase invertebrate density on the bush canopy due to the increase in productivity.

METHODS

2.1 Field Site

Our study was conducted at the Sweeney Granite Mountains Desert Research Center, which is located in San Bernardino County (34° 48’ 20” N, 115° 39’ 50” W), east of Barstow, as well as directly adjacent to the Mojave National preserve. The field site lies in the east Mojave Desert with an average precipitation of 23 cm at the research center. The habitat consists of a rocky terrain with woodland and succulent shrubs, such as the buckhorn cholla (*Cylindropuntia acanthocarpa*). We collected data between May 4 and 7, 2022.

2.2 Pitfall Traps

We collected Invertebrates under 16 creosote bushes using pitfall traps in order to determine the composition of the ground invertebrate community. We selected bushes if they were taller than 167.64 cm and did not touch any other plants, including other creosotes. We dug two holes 30 cm away from the creosote base in opposite directions for our pitfall traps. We made sure they were deep enough to keep our collection cups (530 mL Solo ® cups) flush with the surface, and then added 3 cm of a soap solution to each cup in order to enhance the capture rate of invertebrates. We reduced soil disturbance around the traps by smoothing out mounded soil and replacing litter. The traps were reset at 6 a.m. and 8 p.m., or at dusk and dawn. Due to time restrictions, we left the pitfall traps for the first 5 bushes out for 3 days and 3 nights and the remainder for only 2 days and 2 nights. This meant that, for the first 5 bushes, there were a total of 6 pitfall trap collections for each bush, and for the last 5, there were only 4.

2.3 Bush Beating

We performed bush beatings at 3 p.m. and 8 p.m to find the composition of the canopy invertebrate community. The 10 creosote bushes we sampled were separate from those used for the pitfall traps. We did each beating on different branches among the bush, so we labeled branches to prevent repetition. We performed 2 day beatings and 2 night beatings at each bush, so there were a total of 4 beatings on 4 different branches for each bush. We chose relatively evenly sized branches based on our capacity to lay down a white beat sheet containing a grid measuring 45.72 x 45.72 cm. One person shook each branch for a total of 15 seconds. Looking at the beat sheet, we then counted the total number of invertebrates and species that fell onto the grid.

2.4 Insect Processing

We sifted invertebrates collected from the pitfall traps through a sieving strainer of 60 mesh (250 microns). We consistently poured water through the sieving strainer to help separate specimens from soil particles and used forceps to retrieve invertebrates of all sizes. We counted the total number of invertebrates per order and the total number of all invertebrates per trial.

2.5 Physiological Measurements

To assess creosote bush physiological and phenological characteristics, we measured the percentage of flowers on bushes, bush diameter, and percentage of the bush that was alive. We visually estimated the flower percentage and percent of the bush that was alive. We selected and measured the longest possible branches to find the first bush diameter, and then we made another diameter measurement exactly perpendicular to the first. We then used these measurements to find an approximate oval-shaped circumference.

2.6 Data Analysis

We analyzed data in JMP statistical software v16 (SAS Institute, Inc.). To ensure accuracy despite differing numbers of samples, we determined the number of invertebrates per sample, therefore looking at invertebrate density, not number. We assessed communities through their abundance per order as well as per trial. We ran discriminant simulations for both the canopy and ground invertebrates to determine the similarities and differences in invertebrate community structures between the day and night. We ran an ANCOVA to determine the effect of flower percentage, day and night, and a combination of these two variables on the number of invertebrates. Two similar ANCOVAs were performed on circumference and percent living growth, once again using day and night as another variable.

RESULTS

3.1 Day vs Night

For both the canopy and ground invertebrates, the creosotes fostered different communities depending on the time. On the ground, more Arachnids, Coleoptera, and Hemiptera were found during the day, and orthoptera were found at night (N = 15, 10.00% misclassified; Fig. 1). In the canopy, more Hymenoptera and Diptera were found during the day, and during the night, more Hemipterans were found (N = 10, 15.63% misclassified; Fig. 2).

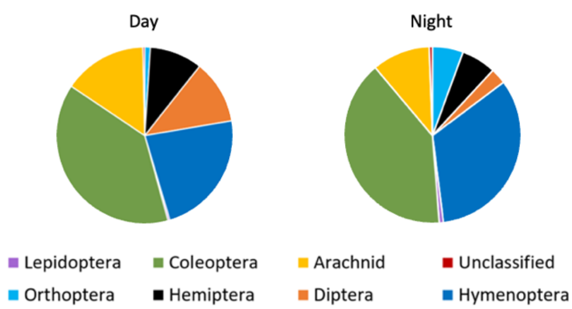

Figure 1. Differences in community between the day and night for creosote (*Larrea tridentata*) ground invertebrates. We sampled invertebrates in the creosote canopy from May 5 and 6, 2022, at Sweeney Granite Mountains Desert Research Center in the Mojave Desert, CA. Pitfall traps were used to determine the ground invertebrates. We placed 2 pitfall traps each at 16 bushes: they were swapped at 6 a.m. and 8 p.m., or dawn and dusk. More Hymenoptera and Diptera were active during the day; more Hemiptera were active at night.

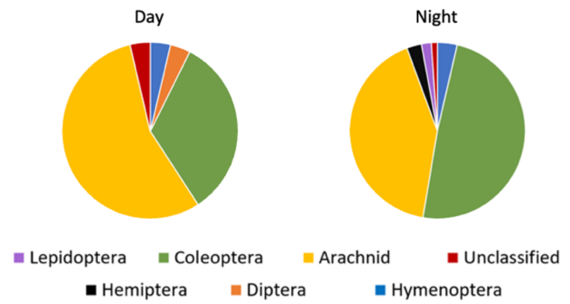

Figure 2. Differences in community between the day and night for creosote (*Larrea tridentata*) canopy invertebrates. We sampled invertebrates in the creosote canopy from May 5 and 6, 2022, at Sweeney Granite Mountains Desert Research Center in the Mojave Desert, CA. Pitfall traps were used to determine the ground invertebrates. Bush beatings were used to determine the number of canopy invertebrates and were performed between 3 p.m. and 8 p.m. on ten different creosote bushes. More arachnids, coleoptera, and Hemiptera were active during the day; more Orthoptera were active at night.

3.2 Flower Percent

When accounting for time of day, the percent of the branches on the bush that were flowering increased the density of invertebrates found on the branches of the creosote (N = 10, t = 2.25, p = 0.00385) regardless of time of day (N = 10, t = -1.03, p = 0.3184). However, flower percent had no effect on the density of invertebrates on the ground (N = 15, t = 0.74, p = 0.4639,) regardless of time of day (N = 15, t = 1.23, p = 0.2290). Controlling for the effects of flower percent, the density of invertebrates during the day and the night changed on the canopy and the ground, but in opposite manners. In the canopy, the density of invertebrates was greater in the night than during the day (N = 10, t = -3.51, p = 0.0029; Fig. 3). On the ground, the density of invertebrates was greater during the day than at night (N = 15, t = 3.44, p = .0018; Fig. 3).

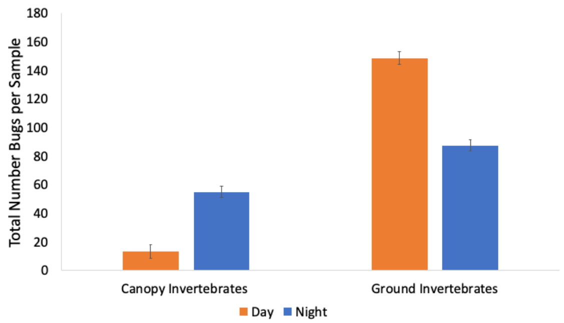

Figure 3. Correlation between day and night and number of invertebrates on creosote bushes. We sampled invertebrates in the creosote canopy from May 5 and 6, 2022, at Sweeney Granite Mountains Desert Research Center in the Mojave Desert, CA. Bush beatings were used to determine the number of canopy invertebrates and performed between 3 p.m. and 8 p.m. on ten different creosote bushes. Pitfall traps were used to determine the ground invertebrates. We placed 2 pitfall traps each at 16 bushes: they were swapped at 6 a.m. and 8 p.m., or dawn and dusk. We observed a greater number of canopy invertebrates during the night than during the day, and we found a greater number of ground invertebrates during the day than the night.

3.3 Bush Circumference

Considering the effects of day and night, the circumference of a bush, and therefore the size of the habitat underneath it, had no effect on the density of invertebrates on or under the creosotes (N_{canopy} = 10, t_{canopy} = 1.52, p_{canopy} = 0.1482; N_{ground} = 15, t_{ground} = 0.42, p_{ground} = 0.6751). This effect remained regardless of time of day for both groups of invertebrates (N_{canopy} = 10, t_{canopy} = -1.40, p_{canopy} = 0.1816; N_{ground} = 15, t_{ground} = -0.04, p_{ground} = 0.9644). Considering the effects of bush circumference, the density of invertebrates during the day and the night once again changed in opposite manners for the canopy and for the ground. In the canopy, the density of invertebrates was greater in the night than during the day (N = 10, t = 0.1482, p = 0.004; Fig. 3). On the ground, the density of invertebrates was greater during the day than at night (N = 15, t = 0.6751, p = 0.0024; Fig. 3).

3.4 Dead vs Alive

With the effects of day and night considered, the percentage of a bush still living has no effect on the density of invertebrates in the canopy or on the ground (N_{canopy} = 10, t_{canopy} = 1.05, p_{canopy} = 0.3093; N_{ground} = 15, t_{ground} = 0.86, p_{ground} = 0.8754). Again, considering the effects of the percentage of a bush alive, the density of invertebrates increased during the night in the canopy (N = 10, t = -3.12, p = 0.0066; Fig. 3) and in the day for the ground (N = 15, t = 3.32, p = 0.0025; Fig. 3).

DISCUSSION

Overall, invertebrate communities changed depending on if it was day or night and how much of a bush was flowering. However, the bush size and the percentage of a bush still alive did not affect invertebrate densities.

There were significant differences in bug community structure at day compared to at night. Hymenoptera and Diptera seemed particularly resilient to the heat, as they were able to move on the bushes at midday, whilst Coleoptera and Arachnid remained under the bushes for most of the day. Hemiptera were on the bushes during the night, and then moved under the bushes during the day. Research on different forms of invertebrates, including Hymenoptera and Coleoptera, have found that activity differences between the day and the night are primarily due to temperature shifts (Albrecht and N.J. Gotelli 2001, Parmenter et al. 1989). It is possible they were moving to avoid higher heat, and that being in the bush means being more directly exposed to sunlight than they would be under the bush. A good avenue for future research would be tagging invertebrates to determine whether they move around creosotes depending on the temperature or time of day.

We saw an increase in the density of ground invertebrates during the day which was expected. This may be because, as temperatures are generally lower under bushes, it is possible that the ground invertebrates use the creosote bushes as an escape from the midday heat, whereas at night, they are able to go out foraging (Moro et al. 1997). However, in the canopy, we saw an increase in invertebrate density during the night when we had expected to see a decrease. Since other studies have found that desert invertebrate activity peaks at dawn and dusk for ground insects, it is possible that both canopy and ground invertebrates are more active during the day overall, but fewer bugs are active specifically at 3 p.m. than at 8 p.m., which is when we collected data on the canopy invertebrates (Ramírez-Hernández et al. 2021). As the ground measurements looked over the course of the entire day, they would not have been affected by these trends. Alternatively, it may be that even the bush canopy is too exposed to heat and solar radiation during the day for invertebrates to want to stay in it. So, the invertebrates only forage in the canopy during the night when conditions are more mild.

There was evidence to support our prediction that the abundance of flowers affected the density of bugs on a creosote bush. Percent flowering most likely had an effect because it indicated a higher amount of productivity, which can increase arthropod density (Lightfoot and Whitford 1991). An increase in productivity of these plants can lead to an increase in nutrient availability, particularly nitrogen, which attracts invertebrates (Lightfoot and Whitford 1991). This phenomenon is particularly important in creosotes, as the chemical defense these plants use against herbivory decreases the availability of usable nitrogen for invertebrates (Lightfoot and Whitford 1991). The increase in flowers could also have increased pollinator and florivore density (Minckley et al. 1999).

The percent of a bush that had living growth did not affect the density of invertebrates on the creosote bush. This does not follow our predictions, as we thought that an increase in living growth would lead to an increase in invertebrate density. However, it does follow previous research to an extent. There is precedent that shows that living growth on a bush is more valuable for some invertebrates who will use the extra nutrients, such as arthropods, but that dead growth can be easier to establish in (Schowalter et al. 1999). It is possible that, as more dead growth helped some communities and harmed others, no overall trend could be established: a possible future study would be examining responses to dead growth on a community level.

Contrary to our predictions, we saw that the size of the bush had no effect on the density of invertebrates under the bush. This means that the number of invertebrates increased proportionally with an increase in bush size, leading to an equal invertebrate density. This is also contrary to findings in prior research where increasing bush size was found to increase invertebrate density (Crisp et al. 1998, Sanchez and Parmenter 2001). Therefore, the effects of bush size might not be as important to take into account for future invertebrate studies as this past research implies.

The differences in trends that we saw between day and night invertebrate communities as well as between canopy and ground communities have important implications for invertebrate community structure in general. These differences imply that conclusions made about day invertebrate communities cannot be applied to night communities, nor the inverse. Similarly, analyses of ground invertebrates cannot be extrapolated to canopy ones, and canopy cannot be projected onto ground. Future studies must consider the differences inherent to these groups and either account for them or only examine one community.

ACKNOWLEDGEMENTS

This work was performed at the University of California's Sweeney Granite Mountains. Desert Research Center, doi:10.21973/N3S942.

We would like to thank the California Natural Reserve System for sponsoring our research. We deeply appreciate the work Ryan Kinsel, Sarah Euchner, Siddarth Malik, Paloma Lobos, Mason Rogers, Rushi Tawade, Tim Chen, and Adrien Castillo did helping us collect data when one of our group members was indisposed. Finally, we would like to thank Tim Miller and Renske Kirchholtes for their consistent advice and aid in setting up and managing difficulties with this project.

REFERENCES

Albrecht, M., and N. J. Gotelli. 2001. Spatial and temporal niche partitioning in grassland ants. *Oecologia* **126**: 134–141.

Badano, E. I., O. R. Samour-Nieva, J. Flores, J. L. Flores-Flores, J. A. Flores-Cano, and J. P. Rodas-Ortíz. 2016. Facilitation by nurse plants contributes to vegetation recovery in human-disturbed desert ecosystems. *Journal of Plant Ecology* **9**:485–497.

Braun, J., and C. J. Lortie. 2020. Facilitation with a grain of salt: reduced pollinator visitation is an indirect cost of association with the foundation species creosote bush (*Larrea tridentata*). *American Journal of Botany* **107**:1342–1354.

Crisp, P. N., K. J. M. Dickinson, and G. W. Gibbs. 1998. Does native invertebrate diversity reflect native plant diversity? A case study from New Zealand and implications for conservation. *Biological Conservation* **83**:209–220.

Jiménez-Valverde, A., and J. M. Lobo. 2005. Determining a combined sampling procedure for a reliable estimation of Araneidae and Thomisidae assemblages (Arachnida, Araneae). *The Journal of Arachnology* **33**:33–42.

Lightfoot, D. C., and W. G. Whitford. 1991. Productivity of creosote bush foliage and associated canopy arthropods along a desert roadside. *The American Midland Naturalist* **125**:310–322.

Little, R., T. J. Swiecki, and W. D. Tietje. 2001. Oak Woodland invertebrates: The little things count.

University of California, Agriculture and Natural Resources Communication Services, Davis, California, USA.

Minckley, R., J. Cane, L. Kervin, and T. H. Roulston. 1999, May. Spatial predictability and resource specialization of bees (Hymenoptera: Apoidea) at a superabundant, widespread resource. *Biological Journal of the Linnean Society* **67**:119–147.

Moro, M. J., F. I. Pugnaire, P. Haase, and J. Puigdefábregas. 1997. Effect of the canopy of *Retama sphaerocarpa* on its understorey in a semiarid environment. *Functional Ecology* **11**:425–431.

Parmenter, R. R., C. A. Parmenter, and C. D. Cheney. 1989. Factors influencing microhabitat partitioning in arid-land darkling beetles (Tenebrionidae): temperature and water conservation. *Journal of Arid Environments* **17**:57–67.

Ramírez-Hernández, A., A. P. Martínez-Falcón, M. Ávila-Argáez, and J. Flores. 2021. Soil moisture shapes diversity and network structure of insect communities associated with *Cylindropuntia imbricata* (Cactaceae). *Arthropod-Plant Interactions* **15**:897–906.

Rhoades, D. F., and R. G. Cates. 1976. Toward a general theory of plant antiherbivore chemistry. *Springer US* **10**:168–213.

Ruttan, A., A. Filazzola, and C. Lortie. (n.d.). 2016. Shrub-annual facilitation complexes mediate insect community structure in arid environments. *Journal of Arid Environments* **134**:1–9.

Sanchez, B., and R. Parmenter. 2001. Patterns of shrub-dwelling arthropod diversity across a desert shrubland-grassland ecotone: a test of island biogeographic theory. *Journal of Arid Environments* **50**:247–265.

Schowalter, T. D., D. C. Lightfoot, and W. G. Whitford. 1999. Diversity of arthropod responses to host-plant water stress in a desert ecosystem in southern New Mexico. *The American Midland Naturalist* **142**:281–290.

Zuliani, M., N. Ghazian, and C. J. Lortie. 2021. Shrub density effects on the community structure and composition of a desert animal community. *Wildlife Biology* **2**:wlb.00774.