

# Insect Interactions of three Trophic Levels on Milkweed Plant *Asclepias sinaica* (Boiss.) Musch

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## ABSTRACT

*Asclepias sinaica*, a host plant for many insects, was chosen for studying the patch occupancy and spatial population structure. Aphid (*Aphis nerii*), ladybird (*Adonia variegata*) and ants (*Monomorium carbonarium*) were chosen for this investigation with the objective to test whether the incidence of these insects is associated with *A. sinaica* plant patches. The temporal fluctuations of each insect species and the distribution of how *A. nerii* affects the distribution of the other two insects were also investigated. The patches were characterized by measuring plant area to assess the influence of patch quality on species occurrence. Plant use by *A. nerii* is influenced by the plant quality and inter-specific interactions. The occurrence of Ladybird on plant patches was associated negatively with ant and positively with *A. nerii* distribution.

**Key Word:** Tritrophic level interaction; *A. sinaica*; Insect-plant interaction; Patch occupancy; Aphid-ant interaction; *A. nerii*

## INTRODUCTION

Interactions between plants and herbivorous insects have provided numerous insights into a range of important ecological and evolutionary processes (Lewinsohn *et al.*, 2005). These have included aspects of community ecology such as the role of multi-trophic interactions, including indirect and apparent competition (Denno *et al.*, 1995). It was noted that a better understanding of co-evolutionary interactions involving plants and their herbivores must include antagonists of the herbivores within a tri- (rather than bi-) trophic framework (Price *et al.*, 1980). Plants possess a variety of biochemical and physical defense mechanisms that can deter, poison, or starve herbivores feeding on them (Hartley & Jones, 1999). Many plant secondary metabolites have toxic effects on a variety of herbivores and pathogens, known from studies *in vitro*, while other plant defenses appear to have indirect effects upon pests and pathogens, such as in attracting predators and inhibiting insect oviposition.

Plant-herbivore interactions provide well studied examples of co-evolution, but little is known about how such interactions are influenced by the third trophic level. The vulnerability of insect herbivores to be attacked by predators and parasitoids is often mediated by interactions with the host plant on which the herbivore feeds. Special herbivores that have overcome certain plant defenses may subsequently co-opt those defenses, for example, by sequestering chemical toxins produced by the plant within their bodies as a defense against their own enemies. Thus, the physical and physiological characteristics of the host plant are thought to be major features influencing the vulnerability of insect herbivores to be attacked by predators and parasitoids and adaptation to those characteristics may be expected to play an important role in avoiding attack by natural enemies (Karban & Baldwin, 1997).

In southern Sinai Egypt, *Asclepias sinaica* is host to a relatively simple community of insect herbivores and their predators. *Aphis nerii* is associated with *A. sinaica*, and feeds on it. Individuals sequester cardiac glycoside toxins from *A. sinaica* and share the plant with a limited set of other species; like weevil *Paramecops sinaica*, and aposematic herbivore *Spilostethus pandurus*. The predator includes syrphid hoverflies (as larvae), coccinellid ladybirds (as larvae & adult) and ant (*Monomorium carbonarium*). This system therefore provides an opportunity to explore how resource use is determined by the interactions between a herbivore and its host plant and by interactions with other herbivores and their shared predators. The purpose of this study was to determine whether the plant quality affect the distribution of all three insects (*A. nerii*, *M. carbonarium* & *A. variegata*) and how these insects distribute on the plant patches, use the host plant, interact on the plant patches and how the presence or absence of any of these three insects affect the distribution of the others.

## MATERIALS AND METHODS

**Study system.** This study was carried out in Wadi Arbaein (33° 57. 28.5. E, 28° 32. 35.65. N, 1620 m altitude), St Katherine's Protectorate, South Sinai, Egypt. The perennial shrub *Asclepias sinaica* (Boiss) is toxic, containing cardenolide glycosides (Elaskary *et al.*, 1995a, & b; Abdel-Azim *et al.*, 1996; Abdel- Azim, 1998). *A. sinaica* is visited by a number of insects. Oleander aphid *A. nerii* (Homoptera: Aphididae) is a bright yellow aposematic insect, which specializes on some genera of Apocynaceae (Martel & Malcolm, 2004) sequestering cardenolides for defense against predators (Malcolm, 1990) and many ladybird species including *A. variegata* (Goeze), present as both larvae and adults and the ants *M. carbonarium*.

**Study method.** Three transects covering 60 plant patches were surveyed. Plant patches were chosen randomly. All

60-host plant patches were surveyed for ant (*M. carbonarium*), aphids (*A. nerii*) and ladybird (*A. variegata*). The data was recorded twice a day for three successive days. The survey was conducted from May, June and July. The data were tabulated and analyzed.

## RESULTS

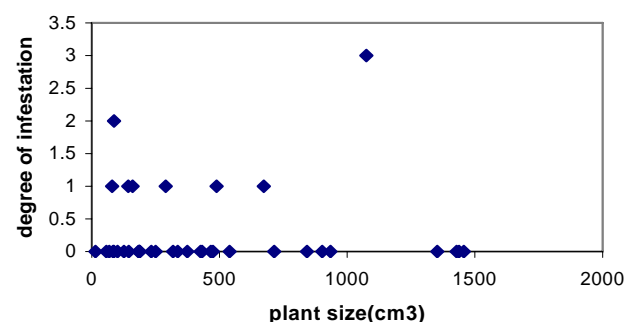
A positive effect of plant size on aphid infestation represented a significant correlation ( $r = 0.0535$ ) between plant size and aphid infestation (Fig. 1). *A. nerii* was therefore more likely to be found on larger plants. Moreover, the presence of *A. variegata* was positively associated with *A. nerii* presence (Fig. 2). By contrast, the presence of *A. variegata* was not associated with plant size; there was no correlation between plant size and ladybird distribution on plant patches (Fig. 3). The ladybird was more likely to be found on the plant with more *A. nerii* infestation rather than plant quality. Meanwhile, aphid infestation had no effect on ant distribution as no correlation was found between aphid infestation and ant distribution. Ant occurrence on plant patches was negatively correlated with ladybird distribution. Data on seasonal abundance showed that there was significant ( $P < 0.01$ ) difference in *A. nerii* abundance during different months. Conversely, there was no difference in ladybird abundance in different months. Meanwhile, there was a significant difference in the ant abundance during different months ( $P < 0.0001$ ). Aphid activity increased from April with activity peak during July (Fig. 4). Ant activity increased abruptly with a peak activity noticed during July. Conversely, activity of ladybird started to decrease until July when the activity highly decreased.

## DISCUSSION

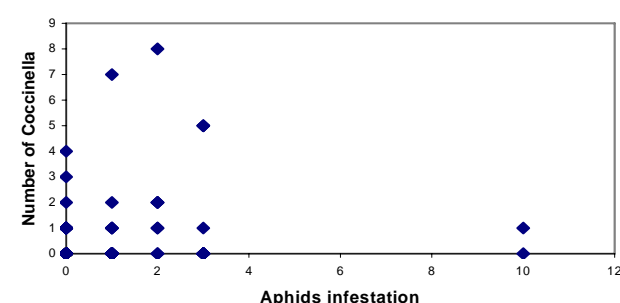
The semiarid ecosystem of St. Katherine has special characteristics like resource-scarce and high variations in flora and fauna. These conditions subject the species to a high risk of predation through their natural enemies (Semida *et al.*, 2001). *A. sinaica* is one of the plants, which contain chemical substances in the form of toxins (Elaskary *et al.*, 1995b). Many plant secondary metabolites defend the plant against herbivores (Sznajder & Harvey, 2003), while other plants appear to have indirect effects upon pests and pathogens (Sirvent *et al.*, 2003). The results of this study clearly demonstrated a positive effect of plant size on aphid infestation. Again a non-random distribution of aphid may be explained by the feeding preference of this species to the fast growing plant particularly the bud. Larger plants represent greater resources in terms of fruit however, since follicle number was positively correlated with plant volume.

On the other hand plant occupancy with *A. variegata* is not only affected by plant size but also strongly affected by aphid infestation, this easily explains by the optimal foraging theory, which was reflected by the preference of

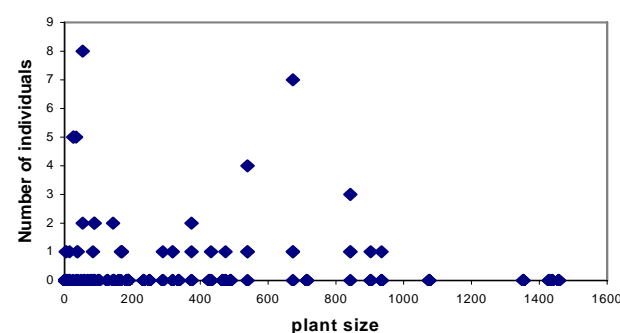
**Fig. 1. The relationship between plant size and infestation by Aphids ( $r = 0.535$ )**



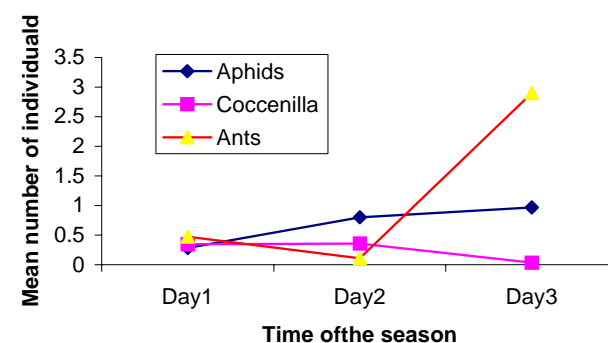
**Fig. 2. The relationship between Coccinella presence Aphids presence ( $r = 0.25$ )**



**Fig. 3. The relationship between plant size and Coccinella in the study area**



**Fig. 4. Temporal pattern of activity**



ladybird to the patches occupied by aphid. This is related to the feeding habit of ladybird on aphid. Resource use by

insects is associated with several biotic factors in the field, such as plant size and the presence of a putative competitor (Van Veen *et al.*, 2006). Our data also supports the idea that the plant volatiles provide a signal to foraging predatory insects about the location of their prey as they found more methyl salicylate was released from the plants infested by aphid than the non-infested plants, which may explain how ladybirds locate the plants with high aphid infestation rather than the one with less infestation (Zhu & Park, 2005). The data on the temporal fluctuations of the three insects showed that there was a synchronized distribution of both the insects. This finding is explained by the abundant of ladybird on aphid as food meal. By contrast, the data illustrates that there was no effect of aphid infestation on ant distribution on the plant patches. This further supports the idea that ant is not dependent on aphid for food.

The ants were abundant on honeydew for feeding rather than aphid. Ladybird population decreased when ant became abundance. This suggested the displacement of ladybird by ants as a predator of aphid traces or honeydew. A negative association between ants and ladybird could arise in a number of ways; but most likely there could be a direct competition for feeding sites or food resources (Denno *et al.*, 1995). The data support the idea of direct and indirect defense mechanism of the plant. Here, it was found the plant *A. sinica* infested with *A. nerii*, which suck the plant sap and may cause plant diseases. The plant *A. sinica* is known to produce secondary metabolites to deter the herbivores. Some insects develop counter adaptations themselves to deal with these chemical substances without any negative impact. Moreover, they may use these substances as a defense against other predator. *A. nerii* was found sequesters and use the plant toxins for defenses (Schoonhoven *et al.*, 2005). There is the consensus that aphids are consistent with their use of plant toxins for defense (Groeters, 1993; Martel & Malcolm, 2004). However, a recent study has shown that population growth varies with respect to inter-specific differences in host plant cardenolide levels, with *A. nerii* doing worse on milkweed species with higher toxin levels (Agrawal, 2004). On the other hand, Martel and Malcolm (2004) found no effect of cardenolide levels on aphid colony biomass (Malcolm, 1990; Groeters, 1993). The plant didn't solve the problem yet, as the aphid persisted to feed on the plant. Our data here supports that *A. sinica* may show defense by secreting plant volatiles to attract predators to get rid of the herbivores. Here, the predators of *A. nerii* are *A. variegata*.

Temporal fluctuations of ladybird and ant activity support the fact that ladybird activity influences the aphid, while ant was not associated with aphid. This makes sense, since ant seems to feed on honeydew but not aphid. Ladybird activity was decreased and displaced by ant. This can also benefit the plant as ant gets rid of honeydew. Negative correlation between ladybird and ants distribution can be explained by the fact that ant secretes volatiles, which deter ladybird. The other possibility is that the ants

protect *A. nerii* against predation by aggressive biting and stinging behavior of the insects inhabiting these plants (Janzen, 1983). Still the plant greatly benefit from tritrophic frame of *A. nerii*, their predator *A. variegata* and ant *M. carbonarium* as a predator to the aphid traces or honey dew.

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