

The Combined Effects of Aphid-Ant Mutualism and Fertilizer on the Resource Allocation of *Valerian edulis*

Danielle M. Dupuy: University of Michigan.

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Abstract

Valerian edulis is a perennial dicot growing in montane and subalpine regions. It is a polygamo dioecious species and popular host plant for aphids. Because of such characteristics, it was chosen to study the effects of resource allocation dependency. This study examined the specific effects of aphid-ant mutualisms, aphid herbivory and the use of fertilizer on the resource allocation of *Valerian edulis*. Results gave no significant evidence that indicated the presence of aphids, ants or the addition of fertilizer effect resource allocation in this plant species. However, results did support previous studies that indicate growth dependence on aphids and ants.

Introduction

Plant resource allocation involves the allotment of carbon, water and other plant nutrients towards varying plant functions including metabolism, growth and reproduction. Both environmental and man-made factors can influence and even change the way a plant allocates its resources. Consequently, whether changes in weather patterns, nutrient availability or herbivory cause a plant to redistribute its energies more towards growth or reproduction, has become a key concern when evaluating fitness and/or evolution of any particular plant species. More specifically, in terms of conservation biology, some significance of resource allocation studies relates to the ability of plant species to adapt to an ever-changing environment.

Studies have been done affirming that, under certain conditions, specific plant species tend to allocate their resources more towards growth. One study, by biologists at the University of Oulu evaluating clonal *Erica* dwarf shrubs (1996), concluded that, following herbivory, the deciduous plant species being tested, allocate their resources more towards compensatory growth than defense mechanisms. On the other hand, opposing studies suggest that herbivory may cause allocation of resources towards defense mechanisms against herbivory of reproductive structures (Siemens, 1994). These, like other similar studies, cover more broad terms such as mammal herbivory or insect herbivory, leaving the effects of specific species, such as aphids, less understood. Overall, I hope to use *Valerian edulis* to evaluate the specific effects of aphid herbivory and ant/aphid mutualisms on resource allocation and to determine how fertilizer may influence growth and resource allocation under the stress of this mutualistic condition.

Aphids naturally and usually exist in a mutualism with ants. Ecologically defined, a mutualistic relationship is considered to be, an interaction between two organisms that is directly and reciprocally beneficial to the other's fitness. In the case of ants and aphids, the tradeoff is primarily protection for food. The feeding aphids excrete a sticky matter known as honey dew, which the ants eat. In turn, the ants provide the aphids with protection from predators. The importance of this type of relationship, especially to the aphids (or in other cases, membracids), has been well documented (Drees 2002; Morales 2000; Cushman, Whitham 1989). "The honey-dew loving ants 'tend' the aphids and prey upon natural enemies of aphids and otherwise unhealthy aphids. Ants also carry aphids to uninfested parts of plants." (Drees 2002). While it is obvious that aphids, being insect herbivores, put a certain level of stress on any plant they infest, the level and nature of these effects have been less quantitatively established. In other words, to what measurable degree does the presence of ants, aphids and/or both affect the allocation of resources within a plant?

The effects of fertilizer in this study were expected to dually correlate with plant growth. Because fertilizers are designed to give plants more nutrients than naturally available, and in turn aid in the overall health of the plant, it is predicted that the fertilizer will have a compensatory effect when combined with aphid herbivory. The plants that are both fertilized and infested with aphids should grow about the same percentage as those without either.

The direct purpose of this study is to use the introduced variable (fertilizer) to test the effects of ant-aphid mutualisms on the resource allocation of *Valerian edulis*. The questions I hope to answer are 1) Does the presence of ants or aphids significantly change a plant's allocation of resources towards growth and/or reproduction? And 2) Does the presence of fertilizer change the level of stress that the plant experiences from aphid herbivory and subsequently cause/prevent a reallocation of plant resources?

Materials and Methods

Study Organism

Valerian edulis is a species of plant belonging to the order Dipsacales and family Valerianaceae. It's a perennial dicot whose closest relatives belong to the Asteraceae family. They are montane and subalpine, usually found in moist meadows and forests and are known to be polygamodioecious. (Weber, 346) This term, more simply, means that some of the plants contain both female and

male plant reproductive systems, while others are unisexual. This plant is of specific interest to this study both because of its reproductive characteristics and because it is a popular host plant among many aphid species. In terms of aphids, there are, in fact, numerous species of aphids ranging in size, color and host plant (Drees 2002). Neither the species of aphid nor the species of ant was controlled for in this experiment.

Protocol

In order to test the dependence of resource allocation on both ant-aphid mutualisms and fertilizer, 80 Valerian *edulis* plants were observed in Gothic, CO (elevation 2900 m.), of which, 3 were excluded because they died. I used 2 ant/ no ant treatments x 2 aphid/ no aphid treatments x 2 fertilizer/ no fertilizer treatments for a total of 8 treatments. Initially ants and aphids were found on all 80 of the plants. Valerian without ants and aphids were ignored to ensure that there were no outside factors influencing their presence. Depending on which of the eight random treatments was assigned to the ten plant groups around the test site, the aphids and ants were systematically removed. Aphids were excluded from specified plants by using Safe-soap, an environmentally safe substance designed to kill aphids and cause no harm to the host plant. The ants were excluded by applying a commonly used gel-like matter known as tanglefoot. It was spread on masking tape around the base of the stalk, which then prevented the ants from climbing.

Miracle-gro was the fertilizer formula used. It contains nitrogen, phosphorus, potassium and other plant nutrients. 500 ml. of Miracle-gro was applied to half of the assigned Valerian and 500 ml. of water was used on the other half as control. These fertilizer treatments were applied once/week for a total of 4 weeks.

The six-week duration (June 24- August 5, 2002) of this project produced four weeks of testable data (July 8- August 5, 2002). This shortage was partially due to time constraints on setting up the treatments. Of the data collected, however, the growth variables (stalk height and leaf length) were measured every week while the reproductive variables (flower # /seed set) were accounted for only once or twice throughout the entire four weeks. The eaten plants and uneaten plants were examined individually in order to get an idea of the true effect of the aphids, ants and fertilizer on the growth of the plant. In terms of

stalk height, the uneaten plants were purposely looked at for the most reliable results because it was certain that herbivory did not affect the data. Analysis was then conducted using a three-way ANOVA.

Independent variables: ant presence, aphid presence and addition of fertilizer. Dependent variables: Stalk height, leaf length, flower number and biomass. In these statistical tests used, however, there is a general requirement for computed data that is normally distributed. Because of this prerequisite, any variable being analyzed by a calculated percentage had to be tested based on the Arcsin % growth or biomass computation rather than just % growth or biomass.

Biomass computations were only calculated for uneaten Valerian. There were 38 Valerian analyzed for biomass, where the arcsin value was used for the statistical results. It was only necessary to compute the data for either reproductive biomass or vegetative, not both. The analysis I conducted was with arcsin calculations of reproductive biomass.

Results

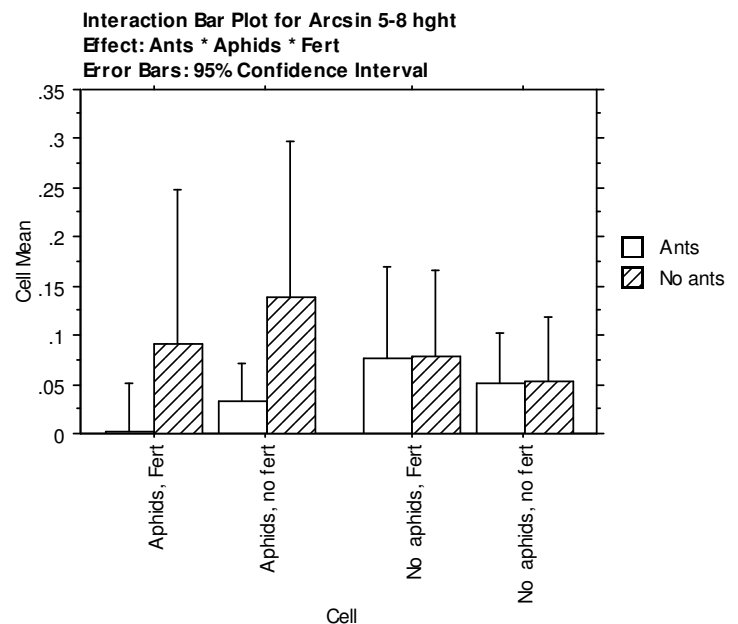
Stalk Height (uneaten)

Table 1: Arcsin % stalk growth

Week 5-8 (July 8- August 5)

	Mean Square	P- Value
Ants	.026	.0361
Aphids	1.514E -5	.8432
Ants *Aphids	.024	.0426
Fert	3.879 E -4	.8443
Ants *Fert	2.214 E -5	.9120
Aphids *Fert	.011	.1896
Ants*Aphids*Fert	2.360 E -4	.9123
Residual	.006	

Figure 1: Arcsin % stalk growth



30 cases were omitted due to missing values.

Week 5-8 (July 8-August 5)

Table 2: Arcsin % leaf growth

Mean Square P- Value

Ants	.003	.4269
Aphids	2.218 E -4	.8267
Ants *Aphids	.005	.2949
Fert	1.754 E -4	.8457
Ants *Fert	2.007 E -6	.9834
Aphids *Fert	6.107 E -5	.9085
Ants*Aphids*Fert	1.957 E -7	.9948
Residual	.005	

Table 4: Flower #

Table 3: Arcsin % reproductive biomass

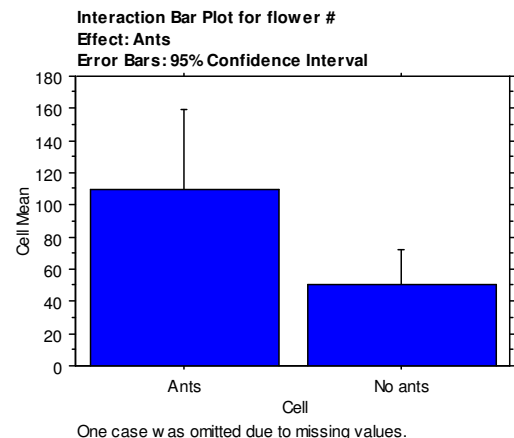
Ants	.9303
Aphids	.6817
Ants *Aphids	.2164
Fert	.9077
Ants * Fert	.9776
Aphids * Fert	.7268
Ants* Aphids* Fert	.7287

ANOVA Table for flower #

	DF	Sum of Squares	Mean Square	F-Value	P-Value
Ants	1	68258.700	68258.700	4.675	.0341
Aphids	1	24718.354	24718.354	1.693	.1975
Ants * Aphids	1	23411.858	23411.858	1.604	.2097
Fert	1	17296.004	17296.004	1.185	.2802
Ants * Fert	1	22.649	22.649	.002	.9687
Aphids * Fert	1	7686.401	7686.401	.526	.4706
Ants * Aphids * Fert	1	22841.761	22841.761	1.564	.2152
Residual	69	1007419.944	14600.289		

One case was omitted due to missing values.

Figure 2: Flower #



Discussion

According to the results the leaf length analysis produced no significant results (Refer to table 2). There was no dependence on the presence of ants, aphids or the addition of fertilizer. Because there was little change in length during the four weeks, this variable was ruled out as a good indicator of growth. On the other hand flower number and stalk height showed a significant dependence on the ants: the presence of ants correlating with a higher number of flowers and the absence of ants correlating with larger stalks. Stalk height analysis showed a dependence on ants and aphids, ants alone but no dependence on the addition of

fertilizer (refer to table 1/ figure 1) This outcome is somewhat unexpected. According to the results, the plants with either aphids and no ants or ants and no aphids grew the most. The results directly contrast with the expected outcome that plants with no aphids, regardless of ant presence, would experience the largest growth rate, suggesting that growth may primarily correlate with ant presence instead of aphid presence as previously predicted.

Although the stalk height results were not as predicted, I believe that this is because of the highly influential role, ants play in the viability of aphid colonies. The ants remove aphid excrement, protect from predators and move aphids to less populated areas of the plant (Drees 2002). Therefore, the absence of ants could have serious consequences, such as overpopulated aphid colonies and starvation which could lead to the death of any aphid colony. The honey dew will begin to form a layer over the stalk inhibiting eating, and the amount of aphids in one area will continue to grow without the ants there to move them around. My observations on many of the plants, in fact support this idea. Many of the plants treated with aphids and no ants eventually had no more aphid colonies, only remnants of honey dew and dead aphids where the colonies used to be. So while statistically it is those plants treated with aphids and no ants that grew the most, these plants actually ended up having either. This could have allowed for an effect where all the nutrients that were previously being sapped by the aphids were now available for plant growth.

In terms of biomass no significance was found in the data. The presence of ants, aphids or the use of fertilizer has no meaningful effect on how *Valerian edulis* allocates its resources towards reproduction or growth.

Due to the conditions of this project, many possible errors could have arisen to skew data. The most influential problem deals with the characteristics of *Valerian edulis*. It is a polygamo dioecious species of plant and reproductive differences were not accounted for in the research. In studies done by Soule, she found that male *Valerian edulis* display lower reproductive efforts each blooming season. More specifically, males allocate less of their resources towards reproduction each year they flower. Not preparing for the difference in the plants could, and probably does, have a significant influence on the results for biomass. Had such variance been noted in the research, the biomass results may have come out significantly which would then have led to far different conclusions. Despite these major complications, this study has found statistical evidence to maintain previous research that has concluded plant growth

dependence on the presence of aphids and ants. Additionally, the use of fertilizer has provided some new insight in the area of growth analysis dependent on insect herbivory.

The essential question of this research dealt with resource allocation. The conclusion was that neither the presence of aphids, the presence of ants nor the use of fertilizer effects the resource allocation of *Valerian edulis*. Overall, however, the results of this study, may work to benefit future studies in the field of resource allocation.