

BENEFITS AND CONSTRAINTS ON PLANT DEFENSE AGAINST  
HERBIVORES: SPINES INFLUENCE THE LEGITIMATE AND  
ILLEGITIMATE FLOWER VISITORS OF YELLOW STAR THISTLE,  
*CENTAUREA SOLSTITIALIS* L. (ASTERACEAE)

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**ABSTRACT**—The evolutionary ecology of plant defenses against herbivores is generally understood in a cost-benefit framework. We studied the ecological consequences of the spines of *Centaurea solstitialis* L. (yellow star thistle) which are typically thought of as defenses against mammalian herbivores. By experimentally removing spines, which consist of dead tissue, we show that “nectar robbing” lepidopteran visitors are deterred by naturally occurring spines (a benefit). Unlike the lepidoptera, legitimate pollinators (bees and flies) were not more likely to visit spineless flower heads, although bees and flies did spend 20% more time per visit on spineless flower heads. One potential cost of having spines is the lower time per visit of the bees. The net result of spine removal from flower heads was a 22% reduction in the percentage of filled seeds of spineless flower heads compared to spined controls. Thus, spines of yellow star thistle may not only deter mammalian herbivory, but also deter lepidoptera which are illegitimate flower visitors. Although the spines may be energetically costly (not measured in this experiment), or costly because of other ecological trade-offs, it appears that these costs are outweighed by the benefits of spines as a plant defense.

**RESUMEN**—La ecología evolutiva de las defensas de las plantas contra herbívoros es generalmente entendida bajo una estructura de costos y beneficios. Estudiamos las consecuencias ecológicas de las espinas de *Centaurea solstitialis* L. las cuales son típicamente consideradas como defensas contra mamíferos herbívoros. Al eliminar experimentalmente las espinas, que consisten de tejido muerto, demostramos que las especies lepidópteras que roban néctar de estas flores son repelidas cuando las espinas están naturalmente presentes (un beneficio). A diferencia de las lepidópteras, las polinizadoras legítimas (abejas y moscas) no fueron más propensas a visitar las cabezas florales sin espinas, aunque las abejas y las moscas pasaron 20% más tiempo en cada visita en las cabezas florales sin espinas. Un costo posible de tener espinas es menos tiempo por visita de abejas. El resultado neto de la eliminación de espinas de las cabezas florales fue una reducción del 22% en el porcentaje de semillas llenas en las cabezas florales sin espinas comparadas con los controles con espinas. Así, las espinas de esta planta no sólo repelen a los mamíferos herbívoros, también repelen a especies lepidópteras las cuales son visitadoras ilegítimas de flores. A pesar de ser las espinas energéticamente costosas (lo que no se midió en este experimento), o costosas por otros balances ecológicos, parece que estos beneficios que tienen las espinas como defensas para las plantas sobrepasan sus costos.

The evolutionary ecology of plant defenses has largely been studied in a cost-benefit framework (Rhoades, 1979; Simms and Rausher, 1987). Plant defenses are considered beneficial because they ultimately reduce herbivory and increase the fitness of plants compared to plants without defenses. However, in

environments without herbivory, plant defenses are thought to impose a net fitness cost to the plant because of unnecessary resource allocations away from growth and reproduction to defense. This line of reasoning assumes that plants have a finite amount of resources and increased allocation to one process (e.g., de-

fense) reduces the amount of resources available for other processes (principle of allocation). Such allocation costs of defense have been proposed as the primary reason why plants are not maximally defended (Simms, 1992). However, allocation costs of plant defense against herbivores have been difficult to detect empirically. In a recent review of the measured costs of plant defense, it was found that nearly two thirds of controlled studies reported that fitness of resistant and susceptible plants was indistinguishable in the absence of herbivory (i.e., no cost of defense—Bergelson and Purrington, 1996).

One potential reason for this lack of empirical evidence for allocation costs of plant defenses is that costs may not be due to allocation trade-offs, but rather due to ecological trade-offs (Simms, 1992; Mole, 1994). An ecological cost of defense is expressed as a reduction in fitness due to effects of the defense on other organisms that interact with the plant. For example, depending on whether generalist or specialist herbivores are present, cucurbitacins, which are plant secondary chemicals, may provide a net benefit (resistance) or cost (susceptibility) to plants in the Cucurbitaceae (Chambliss and Jones, 1966; DaCosta and Jones, 1971).

Several alternative costs and benefits of plant defense have been proposed. For example, plant defenses may impose a cost to the plant because chemical compounds or structures that deter herbivores also may deter mutualists of the plant, such as pollinators (L. S. Adler, pers. comm.). Alternatively, chemical plant defenses have occasionally been implicated in deterring illegitimate "nectar-robbing" flower visitors (Stephenson, 1982; Pyrs-Jones and Willmer, 1992). Similar results have not been previously reported for physical defenses such as thorns and spines. Plant defenses that deter nectar robbers provide an unappreciated benefit to the plant; conversely, plant defenses that deter plant mutualists such as pollinators may represent a cost to the plant.

In this study we investigated the consequences of defensive spines of yellow star thistle with relation to its flower visitors. We manipulated spines to determine their effects on flower visitors and, ultimately, on seed set. We were particularly interested in determining effects of spines on lepidopteran flower visitors that feed on floral nectar but do not pollinate the plant (M. J. Pit-

cairn, pers. comm.). Although spines are thought to be a defense primarily against mammalian herbivores (Gowda, 1996), deterring illegitimate floral visitors may be an additional benefit of spines. We also determined effects of spines on legitimate, non-lepidopteran pollinators, to test whether spines imposed an ecological cost by deterring real pollinators. However, in this study we do not investigate the potential allocation costs associated with spine production.

We asked the following specific questions in our study: 1) Does the presence of spines on bracts affect the number of visits to flowers made by individual illegitimate, non-pollinating (lepidoptera) and legitimate, pollinating (non-lepidoptera) flower visitors?; 2) Does the presence of spines affect the length of each flower visit by lepidopteran and non-lepidopteran flower visitors?; and 3) Does the presence of spines affect the percentage of seeds filled per flower head?

**METHODS AND MATERIALS—*Natural History of Yellow Star Thistle***—Yellow star thistle, *Centaurea solstitialis* L. (Asteraceae) is an invasive annual plant in California that is native to Eurasia and naturalized in temperate regions throughout the world (Holm et al., 1979). It flowers from May to October in California. Yellow star thistle is a highly outcrossing species that is often limited by pollinators, especially late in the growing season (Maddox et al., 1996; Sun, 1997; M. J. Pitcairn, pers. comm.). Plants are very spiny; containing stiff, needle sharp spines up to 2 cm in length surrounding each involucre (Fig. 1). The spines are avoided by cattle and are thought to be effective defenses against some mammalian herbivores (McHenry et al., 1990). The spines have been known to cause bleeding mouths in grazing mammals and also cause a condition called pink eye in which the eyes of livestock are pierced by the spines and become infected (M. J. Pitcairn, pers. comm.).

**Experimental Manipulation of Spines**—In October and November 1997, we selected 15 individuals of yellow star thistle growing in a heavily disturbed site in the Orchard Park vicinity of the University of California at Davis campus. To test for effects of spines on flower visitors and seed set we employed a paired treatment design. Each plant was visually divided in half. On a randomly chosen half we removed all of the spines using a small pair of scissors (Fig. 1). Mature spines consist of dead tissue, and thus our manipulation was not likely to cause an induced response in the plants (Karban and Baldwin, 1997). Spines were trimmed as necessary through the remainder of the flowering season to create a balanced



Fig. 1—Line drawing of a manipulated *Centaurea solstitialis* L. (Asteraceae).

pair of opened flowers on the spined and spineless side of each plant.

**Effects of Spines on Visitors**—We recorded numbers and identities of flower visitors and the length of each visit on 12 occasions for approximately 1 hour each between 1100 and 1700 h, the peak time of floral visitation (total observation time = 12 h). We divided visitors into two classes: lepidoptera and non-lepidoptera (i.e., mostly hymenoptera and some diptera). We made this distinction because lepidopteran visitors of yellow star thistle are not legitimate pollinators but rather consume floral nectar without gathering or transferring any pollen (see Maddox et al., 1996; M. J. Pitcairn, pers. comm.; A. A. Agrawal et al., pers. obser.). European honey bees (*Apis mellifera*), common visitors to the plants in our study, are known to be the major legitimate pollinators of yellow star thistle that actually transfer pollen (Barthell et al., 1994).

**Effects of Treatment on Seed Set**—After flowers senesced, we harvested two similarly aged flower heads from each plant, one from the spined and one from the spineless side of the plant. The flower heads were dissected in the laboratory and the number of filled and unfilled seeds was determined. We then calculated the percentage of filled seeds for each flower as an indication of pollination success and seed set.

**Statistical Analysis**—We determined whether flower visitors were influenced by spines by using a Wilcoxon Signed Rank test. Total number of visits to a plant by each individual pollinator was considered an independent data point. Mean length of each polli-

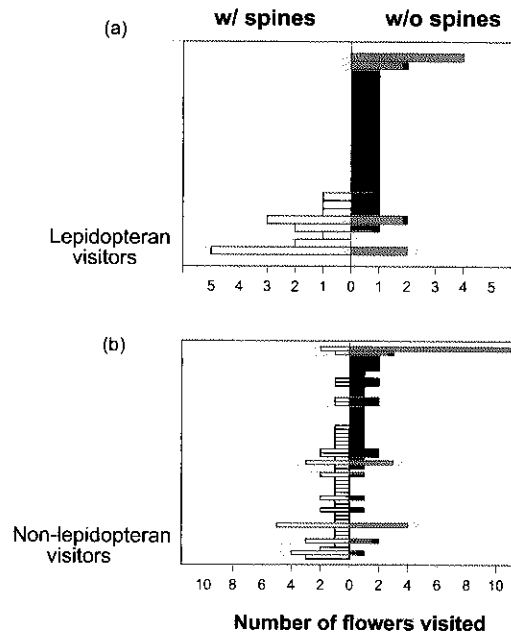


Fig. 2—Visitation of a) lepidoptera (non-pollinators) and b) non-lepidoptera (pollinating bees and flies) to flower heads with spines and with spines removed. Each ratio of visits to spine and spineless flower heads represents an individual pollinator. Although lepidopteran pollinators were more likely to visit spineless flower heads, spines did not influence visitation by bees and flies.

nator's visits to spined versus spineless flowers was compared using a Friedman test. We also compared the percentage of filled seeds in spined versus spineless flowers using a Wilcoxon Signed Rank test.

**RESULTS**—Most lepidopteran flower visitors were butterflies in the families Pieridae, Hesperidae, and Nymphalidae. These illegitimate flower visitors were significantly deterred by spines of yellow star thistle. Lepidoptera made more visits to spineless than to spined flowers ( $n = 26$  visits,  $Z = 2.138$ ,  $P = 0.032$ , Fig. 2a). The legitimate pollinating flower visitors were dominated by Hymenoptera (including the European honeybee) and some Diptera. In contrast to the butterflies, the number of visits by bees and flies was not influenced by the presence of spines ( $n = 54$  visits,  $Z = 0.815$ ,  $P = 0.415$ , Fig. 2b). Although spines did not affect the length of time Lepidoptera spent on flowers (Friedman Test Statistic = 0.421,  $df = 1$ ,  $P = 0.516$ , Fig. 2), the presence of spines reduced the time spent on flowers by bees and

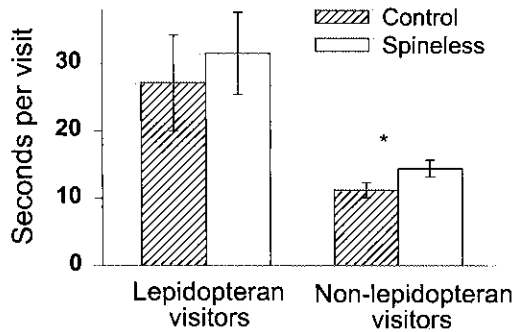


Fig. 3—The length of each visit to flower heads by lepidopteran and non-lepidopteran visitors. Lepidoptera were unaffected by our treatment (spines). Each visit by bees and flies was half as long as that of lepidoptera, and the length of each visit was significantly longer on flower heads with spines removed. Bars are  $X \pm SE$ .

flies more than 22% (Friedman Test Statistic = 4.939,  $df = 1$ ,  $P = 0.026$ , Fig. 3). Finally, flower heads with spines had over 20% more filled seeds than flower heads from which we removed spines ( $Z = 2.542$ ,  $P = 0.011$ , Fig. 4).

**DISCUSSION**—Our results suggest that a novel benefit of spines for yellow star thistle is that they deter illegitimate lepidopteran flower visitors. Although length of time per visit was unaffected, Lepidoptera were more than twice as likely to visit a spineless flower than a spined flower. The net effect of our manipulation was a reduction in the percentage of filled seeds in spineless flower heads. One potential explanation for our results lies in the triggering mechanism of star thistle flowers. Each hermaphroditic flower presents the male and female stages separated temporally and spatially (Maddox et al., 1996). After stimulation by a floral visitor, the filaments retract by approximately 2 mm and the stigma and upper style are exerted, but not yet receptive. This retraction and exertion mechanism causes the pollen to be caught in a ring of hairs on the style, making pollen available to pollinators (Maddox et al., 1996). Although it has not been documented, it is possible that lepidopteran visitors trigger this response without collecting or transferring any pollen. These visitors do generally not get pollen on them because they can sit on top of the flower head and collect nectar with their long proboscises. Thus, lepidopteran

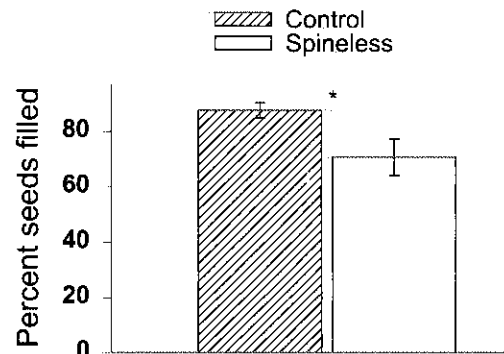


Fig. 4—Percentage of filled seeds in the flower heads of yellow star thistle with spines, and with spines removed. Spine removal resulted in a net effect of significantly reduced seed set. Bars are  $X \pm SE$ .

visitors may cause loss of pollen to the wind or even cause increased rates of selfing in this generally self-incompatible species (self-incompatibility: Maddox et al., 1996; Sun, 1997).

Few other studies have found that illegitimate flower visitors may be deterred by plant defenses, and ours is the first to show that a physical plant defense can serve this function. Stephenson (1982) found that iridoid glycosides in the nectar of *Catalpa speciosa* flowers deterred nectar thieves but did not affect legitimate pollinators. Additionally, Pyrs and Willmer (1992) suggest that high pH and pungent chemical compounds in the nectar of *Lathraea clandestina* provide protection against nectar thievery by ants. If illegitimate flower visitors reduce plant fitness, as has been shown in several systems (Roubik, 1982; Galen, 1983; Roubik et al., 1985), then we predict that natural selection would favor defenses that reduce nectar thievery.

Plant defenses against folivores and nectar thieves are likely to require an investment from the plant. Such investment costs or other ecological costs may constrain plants to be less than maximally resistant to their enemies. Spines of yellow star thistle appear to benefit the plant by reducing both mammalian herbivory and nectar thievery. Spines could be ecologically costly to the plant by causing shorter visits by legitimate pollinators (bees and flies) and could also be energetically costly to produce, although this was not investigated. We found that legitimate pollinators spent 20% less time per visit on control flowers with spines compared to spineless flow-

ers. Although a reduction in pollination potentially could impose a cost to this highly outcrossing species, our measures of reproductive success suggest that the benefits of deterring illegitimate pollinators outweigh the cost of shortened visits by legitimate pollinators. Future studies on costs and benefits of plant defense should consider alternative costs and benefits of defensive traits to contribute to a more comprehensive understanding of the evolutionary ecology of plant defenses. By studying the consequences of having spines for yellow star thistle we hope to contribute to the broadening of plant-herbivore studies to include interactions between plants and their pollinators and other mutualists (e.g., Strauss et al., 1996; Gehring et al., 1997).

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