# The effects of seed feeding insects on seed production of yellow starthistle (*Centaurea solstitialis* L.) in Adana province in southern Turkey

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# Adana (Türkiye) ilindeki Sarı peygamber dikeni (*Centaurea solstitialis* L.)'nin tohum üretimi üzerine tohumla beslenen böceklerin etkinliği

Özet: Sarı Peygamber Dikeni (Centaurea solstitialis L., Asteraceae), Türkiye'nin de içerisinde bulunduğu Avrasya'ya özgün bir bitki olup, Amerika Birleşik Devletlerinin batısında da istilacı bir yabancı ottur. Bu bitki Amerika Birleşik Devletleri'nde kapitula (cicek başcıkları) böcekleri ile biyolojik mücadelede hedef yabancı ot olup, Türkiye'deki böceklerin durumları hakkında çok az şey bilinmektedir. Bu çalışmada iki yıl boyunca Türkiye'nin güneyindeki Adana ilinin üç farklı bölgesini temsil eden yedi örnekleme noktasından kapitulalar (çiçek başcıkları) toplanmıştır. Bulaşık olmayan her bir kapituladaki ortalama sağlıklı tohum sayısı, kapitula çapı ile ilişkili olup, 10 ile 56 arasında değişmektedir. Genel olarak, böcek bulaşıklığı, kapituladaki sağlıklı tohum sayısı azaltmaktadır. Bulaşık olmayan bir kapituladaki sağlıklı ortalama tohum sayışı 34,7 iken bulaşık bir kapituladaki ortalama sağlıklı tohum sayısı 16,8 olup, % 52'lik azalma söz konusudur. Ancak azalış aynı zamanda kapitula büyüklüğüne de bağlıdır. Kapitulalar böcekler tarafından % 5-55 oranında bulaşmaktadır. Bu oran Kaliforniya'da uzun zamandır gözlenen üç bölgenin değerleri ile kıyaslandığında daha azdır (% 49 ila 89 arası). Çalışma alanlarında parazitoidler % 25 oranında gal oluşturan böcekleri, % 4 oranında gal oluşturmayan böcekleri tercih etmekte iken Kaliforniya'da hemen hemen parazitizm yoktur. En yaygın böcekler tanılanmamış bir mordellid ve iki adet gal oluşturan türdür. Bu iki tür; Urophora sirunaseva (Hering) (Diptera: Tephritidae) ve Isocolus scabiosae (Giraud) (Hymnenoptera: Cynipidae)'dir. Diğer tohum böcekleri daha az yaygın olan Oxycarenus pallens (Herrich-Schäffer) (Hemiptera: Oxycarenidae), Bangasternus orientalis (Capiomont), Eustenopus villosus (Boheman) (Coleoptera: Curculionidae), ve Terellia uncinata White (Diptera: Tephritidae)' dir. Yoğun olarak bulunan herbivorların bazıları, Amerika Birlesik Devletlerinde olası doğal düsman olarak değerlendirilebileceği gibi, Türkiye'de de kitle üretimi yöntemi veya koruma yöntemi kullanılarak Sarı Peygamber Dikeninin biyolojik mücadelesinde düşünülebilir.

Anahtar sözcükler: Sarı peygamber dikeni, Asteraceae, kapitula, tohum böcekleri, biyolojik mücadele, yabancı ot

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Abstract: Yellow starthistle (*Centaurea solstitialis* L., Asteraceae) is native to Eurasia, including Turkey, but is an invasive rangeland weed in the western United States. Although this plant has been targeted for biological control in the USA using insects that attack capitula (flower heads), little is known about attack rates in Turkey. We sampled capitula at seven sites representing three different regions in Adana Province in southern Turkey during two years. The mean number of viable seeds per uninfested capitulum was related to capitulum diameter and ranged from 10 to 56. Overall, insect infestation reduced mean per capitulum production of viable seeds from 34.7 in uninfested capitula to 16.8 in infested capitula (52% reduction), but the reduction per capitulum was also a function of capitulum size. The proportion of capitula infested by insects ranged from 5 to 55%, which was lower than rates of infestation previously observed at three long-term study sites in California (49 to 89%). Parasitoids attacked 25% of galls and 4% of non-galling insects, whereas almost no parasitism has been observed in California. The most common insects included an unidentified mordellid and two gall-forming species: Urophora sirunaseva (Hering) (Diptera: Tephritidae) and Isocolus scabiosae (Giraud) (Hymenoptera: Cynipidae). Other seed-feeding species that were less widely distributed included Oxycarenus pallens (Herrich-Schäffer) (Hemiptera: Oxycarenidae), Bangasternus orientalis (Capiomont), Eustenopus villosus (Boheman) (Coleoptera: Curculionidae), and Terellia uncinata White (Diptera: Tephritidae). Some of the more abundant herbivorous species should be evaluated as prospective biological control agents for the United States, whereas augmentation or conservation of some species should be considered to improve control of yellow starthistle in Turkey.

Key words: Yellow starthistle, Asteraceae, capitulum, seed feeder, biological control, weed

# Introduction

Yellow starthistle (*Centaurea solstitialis* L., Asteraceae) is native to Eurasia, including Turkey, and occurs throughout the Mediterranean Basin (Wagenitz 1975; Dostál 1976; De Bolos & Vigo 1995). It is an invasive alien weed in rangeland in the western United States where it was accidentally introduced over 150 years ago by importation of contaminated alfalfa seed (Maddox & Mayfield 1985; Di Tomaso et al. 2006). The plant is toxic to horses, its sharp spines deter grazing and it displaces desirable vegetation. Yellow starthistle is also considered a weed in Turkey in cereals (Kurcman 1993), pastures (Uygun et al. 1996), and field margins (Uygur 1997). In the pastures of the eastern Mediterranean region of Turkey, the weed comprises between 1 and 10% of vegetation coverage (Uygun et al. 1996); however, densities of mature plants are about 4% of that in California (Uygur et al. 2004). Higher densities of the plant in invaded regions may be caused by the absence of coevolved natural enemies (Hierro et al. 2006; Van Driesche et al. 2008).

Three subspecies of *C. solstitialis* are known to occur in Turkey: *C. solstitialis* subsp. *solstitialis* (with yellow petals and long straw-colored spines), *C. solstitialis* subsp. *pyracantha* (with yellow petals and short reddish spines) and *C. solstitialis* subsp. *carneola* (with pink petals and straw-colored spines) (Wagenitz 1975). These are all winter annuals, with seeds germinating after the first fall rains in the Mediterranean region of Anatolia. The cotyledon stage first appears in

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October/November in the eastern Mediterranean region of Anatolia, and a second group comes in March/April. The plant begins to flower in June/July and matures during July and August (Uygur et al. 2007).

In Turkey, C. solstitialis is generally controlled by herbicides in croplands. In field margins and pastures, only early stages of yellow starthistle are eaten by livestock (sheep and goats). However, when plants mature, sharp spines on the flower heads (capitula) can deter grazing. Several management strategies have been developed to control yellow starthistle in California (DiTomaso et al. 2006). Among them, classical biological control is advantageous because it has the possibility of permanently controlling the weed over large regions, economically and with little environmental impact. Other methods, such as herbicides, mowing, managed grazing or controlled burning are expensive, logistically difficult or dangerous. For biological control of yellow starthistle, seven exotic insect species (including one accidental introduction), which attack flower heads and destroy developing seeds (achenes), and a rust fungus have been introduced to the western U.S.A. (Turner et al. 1995; Woods et al. 2010). Although two species of introduced insects are widespread and abundant: Eustenopus villosus (Boheman) (Coleoptera: Curculionidae) and *Chaetorellia succinea* (Costa) (Diptera: Tephritidae) (Pitcairn et al. 2008), they apparently do not provide sufficient control to prevent the weed from spreading (Pitcairn et al. 2006). So, research is being conducted to find additional agents to introduce (Cristofaro et al. 2006; Smith 2007).

In Turkey, previous surveys have found 49 species of herbivores associated with *C. solstitialis* (Rosenthal et al. 1994; Cristofaro et al. 2002; 2006; Uygur 2004). Of these, 30 insect species attack capitula of yellow starthistle. Although there is some information on the impact of *C. succinea* and the weevils *Bangasternus orientalis* Capiomont, *Larinus curtus* Hochhut and *E. villosus* in Greece (Fornasari & Sobhian 1993; Sobhian and Pittara 1988; Sobhian et al. 1992; Sobhian and Fornasari 1994), there is no specific information on the impact of capitula insects in Turkey. Previous field observations in Turkey suggest that the presence and abundance of insect species varies among regions, but this has not been well documented. Because there is interest to increase the level of control of *C. solstitialis* in Turkey, it would be useful to know the level of infestation of the most abundant insects, and how it varies among sites and years.

The purpose of this study was to learn the identity, abundance and impact of seed feeding insects on yellow starthistle in Adana Province, Turkey, where it is presumed native, and to compare the results with observations from California, where it is exotic. This knowledge should help improve our ability to manage the weed in native and non-native areas.

# Material and methods

# **Study sites**

Adana province is located in the Mediterranean Region of Turkey. It is in the middle of the Cukurova Plain, bounded by Taurus Mountains to the north and the Mediterranean Sea to the south. The area has a Mediterranean climate with hot dry summers and cool rainy winters. According to long-term data, average annual precipitation is 625 mm, and the average relative humidity is 66%, but sometimes reaches 90% in summer. The average annual temperature is 18.7 °C. The coldest month is January, the hottest month is August (Anonymous 2010).

Because ecological parameters such as elevation, temperature, and soil properties may affect plant growth, seed production and activities of natural enemies, we chose three locations in Adana province, which represent different elevation, climate and soil properties. Two to three "sites" within a "location" were sampled. All sample sites were uncultivated areas near roadsides and field/orchard margins. All the sites had a good infestation of yellow starthistle when capitula were collected in 2004 and 2005.

# Location 1: Karataş

Two sites were sampled. The first called site A: elevation 6 m, latitude N  $36^{\circ} 35' 58.1''$ , longitude E  $35^{\circ} 23' 20.9''$ , the second called site B: elevation 8 m, latitude N  $36^{\circ} 35' 27.5''$ , longitude E  $35^{\circ} 25' 02.3''$ ). The sites were next to the shore of the Mediterranean Sea and had sandy soil. The location was occupied by *Centaurea solstitialis* subsp. *solstitialis*.

#### **Location 2: Çatalan**

Three sites were sampled (site A: elevation 71 m, latitude N 37° 06' 0.8", longitude E 35° 22' 21.5"; site B: elevation 112 m, latitude N 37° 07' 52.5", longitude E 35° 20' 16.9"; site C: elevation 121 m, latitude N 37° 11' 09.5", longitude E 35° 17' 23.4". All sites in this location were occupied by *Centaurea solstitialis* subsp. *carneola*, which has pink flowers and is endemic to this region. The soil was stony clay.

# **Location 3: Pozantı**

This area has colder winter and is the only one of the three locations, which is snowy in winter. Two sites were sampled (site A: elevation 1215 m, latitude N 37° 30' 55.8", longitude E 34° 56' 21.5"; site B: elevation 1344 m, latitude N 37° 44' 13.7", longitude E 35° 00' 54.0" in Pozanti. The soil was stony clay. The location was occupied by *Centaurea solstitialis* subsp. *solstitialis*.

Climatic data for these locations were obtained from the Turkish State Meteorological Service for years 2004 and 2005 from the closest meteorological stations to the collection locations. For Karataş, data were from Karataş station no. 17981 (10-15 km distance from collection sites); for Çatalan Adana station no. 17351 (20-40 km); and for Pozantı, Nigde station no. 17250 (45-60 km).

# **Collection of capitula**

At least 100 mature capitula were collected at each site on each sample date (total of 1400). The nearest plant was collected every two paces walking outside the perimeter of permanent plots used for another study until at least 100 capitula were collected. In 2004, all locations were sampled on 27 August. In 2005, Pozanti was sampled on 28 July, and Karataş and Çatalan on 7 August. In both years, plants had senesced at all sites by the time that they were sampled. Capitula from each sample site were sealed in a paper bag and were held for emergence of adult insects. After collection, the bags were placed in a cold and dark room at 4 °C for a month to facilitate completion of diapause. Then the bags were held at room temperature and ambient light until dissection.

# **Dissection of capitula**

Capitula were dissected at least 9 months after collection. The diameter of each capitulum was measured to the nearest mm using a caliper, and the presence of florets, signs of oviposition by *Bangasternus* sp. (black eggs on stem or phyllaries) or *Eustenopus* sp. (covered hole in capitulum), and numbers of galls were recorded (Sobhian & Zwölfer 1985). Seeds were classified as viable, damaged or undeveloped, and by presence or absence of pappus; however, here we report only data on the combined (pappus and nonpappus) seeds. Adult and immature insects were counted and classified as well as possible. Specimens were sent to the Systematic Entomology Laboratory, United States Department of Agriculture, Agricultural Research Center, Beltsville, MD for determination, and representative specimens were deposited. For capitula from which insects had already emerged, we classified them as infested by a "galling insect" if a gall was present, otherwise as "nongalling" if there was obvious chewing damage. Lepidoptera were not identified because of poor condition.

#### Climate

Weather data from the three collection locations are presented in Figure 1. Karataş and Çatalan had the same mean annual temperature (18.9 and 19.3 °C during Sept. 2003-Aug. 2004 and Sept. 2004-Aug. 2005, respectively) and were warmer than Pozantı (11.7 and 12.0 °C). August maximum temperatures were higher at Çatalan (33.3 and 34.4 °C) than at Karataş (30.7 and 31.4 °C and Pozantı (30.0 and 31.2 °C). Mean minimum temperatures remained above 5.8 °C at Karataş and 5.2 °C at Çatalan, but fell to -4.0 °C at Pozantı, which had mean minimum temperatures below 0 °C for 3 months each winter. Annual precipitation was similar at Karataş and Çatalan (700 and 556 mm vs. 640 and 505 mm), but was much lower at Pozantı (289 and 264 mm). Thus, Karataş and Çatalan were most similar, although the latter had less precipitation and hotter summers. Pozantı had much colder winters and less than half the precipitation of the other two locations. Although temperatures did not change very much from year to year during our study, precipitation was 21% lower during the second year at Karataş and Çatalan and, 8% lower at Pozantı.

#### **Statistical analysis**

Mean values for number of seeds per capitulum were analyzed by ANOVA to determine the effects of site, location, year, capitulum size, infestation and all the possible interactions (Abacus Concepts 1998). Daniel's test for trend was used to evaluate the effect of capitulum diameter on the proportional reduction of viable seeds per capitulum caused by insect infestation (Conover 1980). Confidence intervals for proportion data were calculated using the binomial distribution (Conover 1980). The Shannon index  $(H' = -\Sigma[p_i * \log(p_i)])$ , which represents species richness, and the Simpson index  $(\lambda = \Sigma[p_i])$ , which represents community dominance, where  $p_i$  is the proportion of individuals of species *i*, were calculated for each location (Pielou 1975).

# **Results and discussion**

## Size of capitula and seed production

Data for uninfested capitula were analyzed separately to determine the effects of year, site and capitulum size on seed production independent of the effects of insect infestation. Both the number of total seeds (undeveloped, damaged and viable) and the number of viable seeds per capitulum were linearly related to the capitulum diameter (Fig. 2, total seeds =  $-1.00 (\pm 1.92 \text{ SE}) + 6.87 (\pm 0.30)$ \*diameter,  $R^2 = 0.36$ ,  $F_{(1, 947)} = 538.8$ , P = 0.0001; viable seeds =  $-16.83 (\pm 2.3 \text{ SE}) + 8.12 (\pm 0.36)$ \*diameter,  $R^2 = 0.35$ ,  $F_{(1, 947)} = 510.7$ , P = 0.0001).

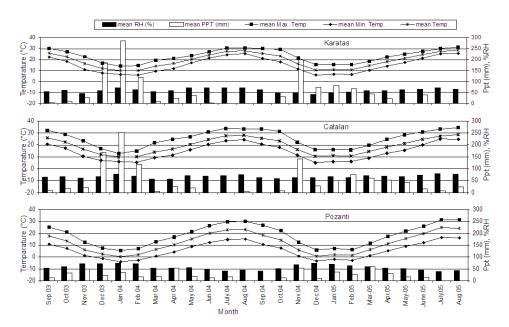
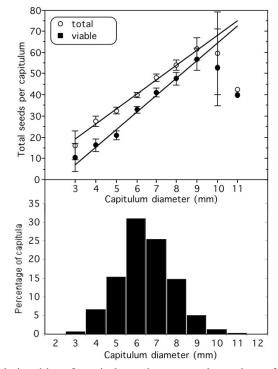


Figure 1. Mean climatic conditions for the three locations in Adana Province, Turkey during September 2003 to August 2005.

The number of viable seeds that we observed was less than the number that were observed in northern Greece by Sobhian & Zwölfer (1985), who reported a much steeper slope and lower y-intercept (seeds = -125.2 + 23.87\*diameter); however their graph suggests that their data may be based on a narrower range of capitula (range 6.5 to 10 mm). Woods et al. (2008) reported similar data for production of mature seeds by uninfested capitula at two sites during two years in California. Slope values ranged from 4.94 to 7.64 and the Y-intercept from -12.27 to -33.12.



**Figure 2.** Relationship of capitulum size to total number of seeds per capitulum for uninfested capitula (mean  $\pm$  95% CI), and frequency distribution of all capitula. Regression for total seeds: Y = 6.87[0.30 SE] \* X - 1.00[1.92], P < 0.0001, R<sup>2</sup> = 0.36; viable seeds: Y = 8.12[0.36 SE] \* X - 16.83[2.33], P < 0.0001, R<sup>2</sup> = 0.35.

In our study, the proportion of seed that was viable in uninfested capitula was 58 to 65% in small capitula (3-5 mm diameter), but it rose steadily from 82% in 6mm capitula to 92% in 9-mm capitula (data not shown). The frequency distribution of all capitula by size is presented in the bottom of Figure 2 (mean diameter  $6.4 \pm 1.3$  [sd] mm). The number of total seeds per uninfested capitulum and capitulum size varied among sites and between years at each site (Fig. 3); however, we do not know the specific causes of this variation. Among the three locations, the number of total seeds per uninfested capitulum size also varied (Fig. 4); however, this did not appear to be related to differences in elevation or subspecies of yellow starthistle.

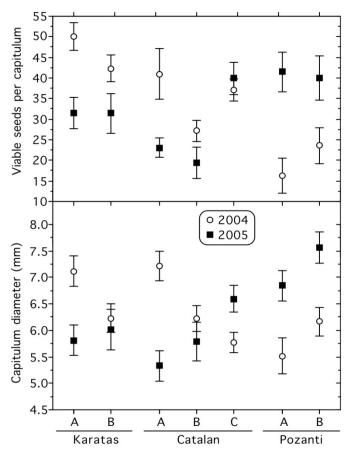


Figure 3. Variation in viable seeds per capitulum and capitulum size for uninfested capitula among sites and years (mean <u>+</u> 95% CI).

# **Capitulum insects**

The species of herbivorous insects reared from the capitula are listed in Table 1. The abundance of each species varied among sites and sample years (Fig. 5). The most common herbivore was *Urophora* sp. (Diptera: Tephritidae), which is probably mainly *U. sirunaseva*, based on identification of four adult specimens and results of other surveys (Sobhian & Zwölfer 1985, White & Clement 1987, Sobhian 1993, Rosenthal et al. 1994). This species occurred at all sites. It forms hard, thick-walled galls in the receptacle (Sobhian & Zwölfer 1985), which permitted identification of infested capitula even when they were parasitized or the adult had previously emerged. The average number of galls per infested capitulum was 1.2, with a maximum of 4. This species is known to develop only *C*.

*solstitialis* (Sobhian 1993) and was common in a more extensive survey of Turkey (Rosenthal et al. 1994).

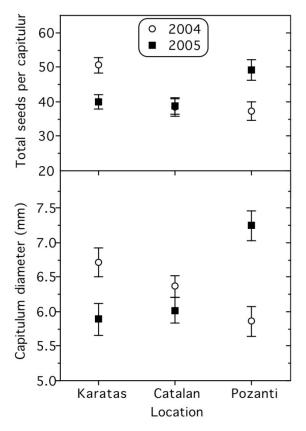


Figure 4. Variation in capitulum size and total seeds per capitulum for uninfested capitula among locations and years (mean <u>+</u> 95% CI).

The second most common species was a gall wasp identified as *Isocolus* scabiosae (Giraud) (Hymenoptera: Cynipidae), which was most abundant at Çatalan and Pozantı. It forms small galls from developing seeds, which facilitated identification of infested capitula (Sobhian & Zwölfer 1985). The average number of galls per infested capitulum was 1.6, with a maximum of 5. The species limits and host plant specificity of this gall wasp are questionable (M.L. Buffington pers. comm.); however, members of this genus mainly attack capitula of plants in the genus *Centaurea* (Baumann & Brandl 1993). A phylogenetic analysis indicates that *I. scabiosae* and *I. rogenhoferi* Wachtl, which were both associated with *C. scabiosa*, are very distinct from other *Isocolus* species associated with *Centaurea* species in the Jacea clade (*C. solstitialis*, *C. jacea* L. and *C. maculosa* Lam.). Other species names have been reported associated with *C. solstitialis* (Baumann & Brandl 1993). *Isocolus* sp. associated with *C. solstitialis* (Baumann & Brandl 1993). *Isocolus* sp. associated with *C. solstitialis* in France, Italy and the

Balkan Peninsula was considered by Sobhian & Zwölfer (1985) to be different than *Isocolus jaceae* (Sch.) associated with *C. scabiosa* L. However, in Serbia, a gall wasp reported as *I. jaceae* infested 39 and 66% of *C. solstitialis* capitula during two different years (Manojlovic & Tosevski 1990). These authors reared this insect from seven *Centaurea* and one *Carduus* species. Thus, there appears to be some confusion regarding the taxonomy and host associations of species in this genus. It will be important to do molecular genetic studies to help clarify the taxonomy of these gall wasps and to determine if a host-specific population exists that could be used as a classical biological control agent (Antonini et al. 2008).

An unidentified mordellid (Coleoptera) was very common at Karatas site B, but also occurred in low numbers at Çatalan and Pozantı sites. Such a species was not discussed by Sobhian & Zwölfer (1985), and Rosenthal et al. (1994) only reported a rare unidentified species on C. calcitrapa encountered during their Turkey survey. Oxycarenus pallens (Herrich-Schäffer) (Hemiptera: Oxycarenidae) was collected only at Pozanti. Rosenthal et al. (1994) reported this species as rare on C. calcitrapa and C. solstitialis during their Turkey survey. This species is widely distributed in Europe, northern Africa, and western and central Asia and has been reported to feed on Solanum and other herbaceous species (Costas et al. 1997, Krist & Kment 2006). However, we found little information on the specificity or behavior of this species. The remaining species were less widely distributed at our sites. Eustenopus villosus was collected only at Pozanti, and all but one specimen of B. orientalis were collected at Pozanti. All except one specimen of Terellia uncinata White (Diptera: Tephritidae) were collected at Karataş, and at only one site during one year. However, Eustenopus villosus and B. orientalis were reported to be common (present at >1/4 of 39 sites), and T. uncinata was widespread (at >1/2 of sites), in a more extensive survey of C. solstitialis in Turkey (Rosenthal et al. 1994). The geographic occurrence of E. villosus in Greece was reported to be patchy (present at 3 of 5 sites); and the authors thought that this may be caused by high parasitism rates (up to 97%) (Fornasari & Sobhian 1993). Although E. villosus is widely distributed in California (where there is almost no parasitism), it is most abundant in cooler habitats, such as the coastal hills and Sierra foothills.

The climate in these habitats is more similar to that of Pozanti than Çatalan or Karataş, which suggests that this species is more successful in cooler climates. Understanding the influence of climate and other environmental factors on the success of some of the principal herbivores may help reveal ways to improve their impact, especially within their native range. In any case, it appears likely that different species will play dominant roles in different habitats in Turkey, such as *E. villosus*, *O. pallens* and *B. orientalis* in cooler, higher elevation locations, such as Pozanti, and *T. uncinata* and the mordellid at lower elevation locations, such as Karataş and Çatalan. High abundance of *I. scabiosae* at the Pozanti and Çatalan locations suggests that this insect may have a wider climatic range, but it is not clear why abundance at the two locations differed so much between the two years. *Isocolus* sp. was reported as rare on *C. solstitialis* in a previous survey of Turkey (Rosenthal et al. 1994). It is conceivable that parasitism may cause temporal and

geographic fluctuation in some of these herbivore populations (Van Driesche & Bellows 1996).

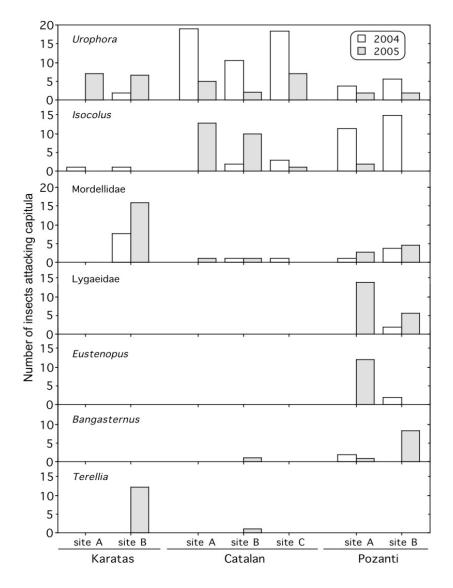


Figure 5. Number of insects attacking capitula.

#### **Infestation rates**

The proportion of capitula infested by insects ranged from 5 to 55% (Fig. 6 top). This is lower than rates of infestation observed at three long-term study sites in California, which generally ranged from 49 to 89% after introduced insects had become established (Pitcairn et al. 2005). Infestation rates of *U. sirunaseva* at sites

in Turkey (mean: 5%, range: 0 to 14%) were comparable to those observed at sites in Greece (mean: 5%, range: 0 to 56%; Sobhian 1993) and California (mean: 10%, range: 0 to 23%; Pitcairn et al. 2005).

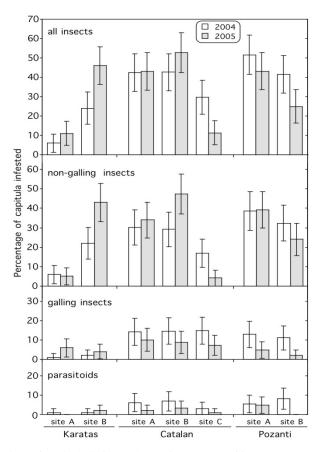


Figure 6. Proportion of capitula infested by various types of insects (mean  $\pm$  95% CI).

*Eustenopus villosus*, which was introduced from Greece (Turner & Fornasari 1995), is more abundant in California (mean: 51%, range: 5 to 82% of capitula infested), where it is very widespread (Pitcairn et al. 2005), especially at sites that have higher precipitation or elevation (Yacoub 2005). Mean infestation rates were 16% in Greece (range: 0 to 43%; Fornasari & Sobhian 1993) and 0.7% (range: 0 to 9%) in our study. *Chaetorellia succinea*, which was accidentally introduced from Greece (Balciunas & Villegas 1999), is also widespread in California (mean: 9%, range: 0 to 28%; Pitcairn et al. 2005), but was not recovered in Turkey during our study. This species is abundant at warmer sites in California, which have climate similar to Çatalan (Yacoub 2005).

Interspecific competition may affect the relative abundance of some capitula species. For example, *Urophora sirunaseva* attained infestation rates of up to 44% of capitula soon after establishment in the western USA (Turner et al. 1994).

**Table 1.** Herbivorous insects attacking capitula of yellow starthistle at three locations in Adana Province, Turkey, based on presence of adults or diagnostic signs of galls or immature stages.

	Number of insects				
Species	Çatalan	Karataş	Pozantı	Total	
Lasioderma sp. (Coleoptera: Anobiidae)	2	5	0	7	
Bruchidius sp. (Coleoptera: Bruchidae)	0	0	1	1	
Bangasternus orientalis (Capiomont)	1	0	12	13	
(Coleoptera: Curculionidae)					
Eustenopus villosus (Boheman)	0	0	15	15	
(Coleoptera: Curculionidae)					
Larinus curtus Hochhut	4	0	2	6	
(Coleoptera: Curculionidae)					
Unidentified species	4	25	13	42	
(Coleoptera: Mordellidae)					
Urophora sirunaseva (Hering)	64	16	14	94	
(Diptera: Tephritidae)					
Terellia uncinata White	1	13	0	14	
(Diptera: Tephritidae)					
Oxycarenus pallens (Herrich-Schäffer)	0	0	23	23	
(Hemiptera: Oxycarenidae)					
Isocolus scabiosae (Giraud)	29	2	30	61	
(Hymenoptera: Cynipidae)					
Unidentified species (Lepidoptera)	9	5	2	16	
Number of insects	114	66	112	292	
Number of capitula	617	414	430	1461	
Number of species <sup>a</sup>	8	б	9	11	
Shannon index (species richness, H') <sup>b</sup>	0.55	0.66	0.82	0.85	
Simpson index (dominance, $\lambda$ ) <sup>c</sup>	0.39	0.25	0.17	0.19	

<sup>a</sup> Assuming only one species of Lepidoptera.

<sup>b</sup> Higher values indicate more species and/or more individuals of each species.

<sup>c</sup> Higher values indicate more uneven abundance of the different species.

However, attack rates subsequently decreased in California (Pitcairn et al. 2005; Yacoub 2005; Woods et al. 2008), probably because of competition with *C. succinea* and *E. villosus*, which became the dominant capitulum species. In Turkey, the lowest incidence of U. *sirunaseva* occurred at Pozanti, where

*Bangasternus, Eustenopus* and *Oxycarenus* were most abundant. The fewest number of species (6) were collected at Karataş and the most at Pozanti (9), which suggests an increase in biodiversity going from the Mediterranean Sea to the Taurus Mountains (Table 1). *Urophora sirunaseva* dominated the herbivore community at Çatalan, which produced a high Simpson index and lowered the Shannon index relative to that at Karataş even though the latter had fewer species. Pozantı had the highest Shannon index and lowest Simpson index, reflecting both the greater number of species and more even abundance of individuals. Combining all three locations increased the number of herbivores to 11, but only slightly raised the Shannon index above that of Pozantı.

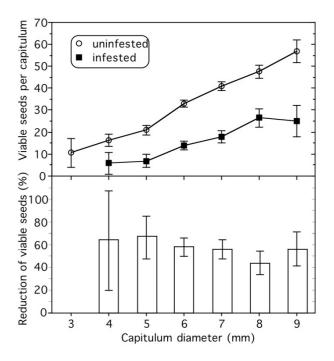


Figure 7. Effect of insect infestation and capitulum diameter on number of viable seeds per capitulum (mean  $\pm$  95% CI). Data for 10 and 11-mm capitula were excluded because of small sample sizes (9 and 1).

## Effects of capitulum feeders on seed production

Overall, insect infestation reduced mean per-capitulum production of viable seeds from  $34.7 \pm 0.6$  (SE) in uninfested capitula to  $16.8 \pm 0.8$  in infested capitula (52% reduction), but the number reduced per capitulum was a function of capitulum size (Fig. 7 top). The proportional impact tended to be higher for smaller capitula (Daniel's test for trend, P < 0.05; Fig. 7 bottom), except that none of the smallest capitula (3 mm diameter) were infested. Neither location or year, nor their interaction affected the relationship between infestation, capitulum size and number of viable seeds per capitulum. The mean number of viable seeds per

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capitulum was higher at Karataş ( $36.2 \pm 0.9$  SE) than at the Çatalan ( $27.0 \pm 0.7$ ) or Pozanti ( $24.3 \pm 1.1$ ), which is probably caused by the lower proportion of capitula infested by insects at Karataş (Fig. 6). Mean production of viable seed per capitulum was negatively correlated to the proportion of capitula infested at a site (Fig. 8). Extrapolation of the regression line suggests that viable seed production should decrease to zero as infestation rate approaches 84%. At one site in California, seed production and density of *C. solstitialis* plants decreased to zero at a site where overall annual infestation rates ranged between 52 to 82% during the previous 4 years (Pitcairn et al. 2005). However, in a two-year insecticide exclusion field experiment, an infestation rate of 79% reduced viable seed production per m<sup>2</sup> by 98%, and subsequent seedling density by 31%, but did not reduce the density of adult plants because of compensatory survivorship (Garren & Strauss 2009).

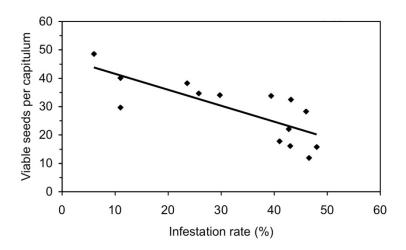


Figure 8. Effect of infestation rate on production of viable seed. Each point is one site during one year (Y = -0.56\*X + 47.23,  $R^2 = 0.60$ ).

# Parasitoids

Overall, 25% of capitula containing galls were parasitized and 4% of those with non-galling insects (Table 2). Sobhian and Zwölfer (1985) reported average parasitism of 42% of *Urophora* spp. galls in southern Italy and the Balkan peninsula and noted that parasitism of *Isocolus* spp. could be over 30%, but they did not report any site-specific data. Predation and parasitism caused 48% mortality of *U. sirunaseva* in Greece (Sobhian 1993), which is comparable to the rate we observed. However, 47% of *E. villosus* were parasitized in Greece (Fornasari and Sobhian 1993), which is much higher than the rate we observed on non-galling insects. Eggs of *B. orientalis* were found on 13% of capitula at Pozanti (sites and years combined, data not shown), but only 8% of these completed development to at least large larvae, suggesting high rates of predation or parasitism. Such mortality factors are likely to reduce the abundance of the

capitula insects, and thus their impact on seed production. Almost no parasitism of the introduced capitula insects has been observed in the western USA (Turner et al. 1990, M. Pitcairn pers. comm.), which is encouraging for their effectiveness when released outside their native range. However, there has been high mortality of eggs and larvae of *B. orientalis* in California (M. Pitcairn pers. comm.), similar to that in Greece (Maddox et al. 1991), so this species may be more susceptible to generalist parasitoids or predators.

Percentage of hosts parasitized							
Family	N total	N <sup>a</sup>	Gall	Non-gall			
Braconidae	12	10	0	2			
Eurytomidae	22	15	12	1			
Pteromalidae	13	7	6	1			
all parasitoids <sup>b</sup>	64	40	25	4			

Table 2. Host associations of parasitoids reared from yellow starthistle capitula.

<sup>a</sup> Number of specimens, excluding those not associated with a host.

<sup>b</sup> Includes unidentified specimens.

Because the capitula were bagged in groups rather than being individually enclosed, it was often not possible to associate individual parasitoids with their host. However, based on the specimens that were associated with a host, 93% of the eurytomids were associated with a gall; specifically: 67% with *Urophora* sp. and 20% with *Isocolus* sp. (and 6% uncertain host). It is possible that the one eurytomid specimen that was associated with a weevil may have moved into this capitulum after emerging from a gall. Two unidentified species of Braconidae were associated with non-galling insects. More pteromalids were associated with galls (80% of specimens) than with non-galling hosts (20%).

# Conclusions

The biology of several of the most abundant capitulum insects found in Adana province, including the cynipid *I. scabiosae*, the hemipteran *O. pallens* and the tephritid *T. uncinata* is poorly known. Given their occasionally high abundance observed in this study, it would be useful to know more about their host specificity, environmental requirements and impact on seed production. The geographically patchy distribution of these species, as well as of *B. orientalis* and *E. villosus*, may be related to their environmental preferences, but it is possible that they have been excluded from some areas because of historical events such as periodic destruction of the host plant and/or capitula, or the application of insecticides. In the latter case, it may be possible to improve management of the weed by augmentative biological control (Van Driesche et al. 2008) by redistributing these species, such

as applying insecticides or mowing capitula when the insects are developing inside (DiTomaso et al. 2006).

*Eustenopus villosus* and *Chaetorellia succinea* have a high impact as measured by seed reduction per capitulum infested (Sobhian & Zwölfer 1985). Densities of capitula per  $m^2$  (a measure of weed infestation) have decreased at several long-term study sites in California where insect infestation rates are high (Pitcairn et al. 2005, M. Pitcairn pers. comm.). The rarity of *E. villosus* and absence of *C. succinea* in Adana Province, which are the two most abundant agents in the western USA (Pitcairn et al. 2008), suggests that augmentation or introduction of these species may help reduce populations of *C. solstitialis* in this region. However, because there are endemic species of *Centaurea* in Turkey, additional host specificity tests should be conducted before making such releases. Other agents that attack the leaves, roots or stems may also be important for controlling the weed and should be studied (Smith et al. 2005; Cristofaro et al. 2006; Smith 2007).

The most abundant capitulum insects observed in Adana province, *U. sirunaseva, I. scabiosae, O. pallens* and *T. uncinata,* should be considered as prospective biological control agents to evaluate for use in USA. Of these species, only *U. sirunaseva.* has been introduced. *Urophora jaculata* Rondani was introduced to California from Italy; however, this species failed to establish because the California population of the weed was physiologically unsuitable (Clement 1994). *Urophora sirunaseva* was introduced to USA from Greece and has become widespread, but it is not very abundant (Sobhian 1993; Pitcairn et al. 2003). Preliminary host specificity studies should be conducted on the other three species.

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