

BIOLOGICAL CONTROL OF MEALY PLUM APHID

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INTRODUCTION

Reduced use of dormant season sprays in prune production in the Central Valley, due to human health risks of fog bound sprays and environmental risks of runoff into Central Valley river systems, has caused the mealy plum aphid (MPA), *Hyalopterus pruni* to be a significant problem. This aphid develops large populations on the undersides of prune leaves in the spring that sap tree vigor, slow the growth of young trees, aggravate the splitting of fruit in years with high June temperatures, and occasionally lower fruit sugar content. MPA populations generally migrate from older orchards to their summer host plant (cattails and *Phragmites*) around mid June, but in young orchards that have vigorous growth through the summer aphid populations will remain in the orchard all through the year.

Alternatives to dormant season sprays include fall applications targeted against the egg-laying generation (oviparae) of aphids that return to the orchards in November, or delayed dormant treatments targeted against the first generation of aphids that emerge from overwintering eggs in March. During the last three years we have characterized the timing of the fall migration and overwintering biology of both MPA and the leaf curl plum aphid (LCPA), *Brachycaudus helichrysi*. Fall migrations of both aphid species begin with the gynoparae that return to the orchards in September (LCPA) or October (MPA) to produce a generation of nymphs that develop into the egg-laying oviparae. Male aphids then return to the orchards to mate with the oviparae in either October (LCPA) or November (MPA). The beginning of the male migrations provides an indication of the last date by which fall treatments must be applied. Aphid eggs overwinter in diapause which must be broken by chilling before heat units can be accumulated toward egg hatch. For MPA diapause is broken around late January or early February depending on the year, and then egg hatch occurs approximately four weeks later. For LCPA diapause is broken at the same time as for MPA, but egg hatch is more rapid taking approximately 10 days. Thus treatments for the first generation of aphids could begin in late February for LCPA or mid March for MPA.

While substantial levels of parasitism by *Aphidius colemani* have led to a general reduction in the spread of LCPA aphid colonies both within and between trees in prune orchards in recent years, there remains a need to establish an effective population of *Aphidius transcaspicus* to provide successful biological control of MPA. We have made extensive surveys of mealy aphids and *A. transcaspicus* throughout the Mediterranean in recent years and have been able to document the occurrence of three different mealy aphid species with distinct host tree preferences. Each of these aphid species is attacked by what appears to be a single parasitoid species, *A. transcaspicus*, although there are genetically distinct subpopulations or strains of the parasitoid occurring in different geographic regions. We have now established that the invasive MPA in

California probably originated from southern Spain, and have focused collections of parasitoids on this region assuming that local populations of parasitoids would be better adapted to the MPA genotypes from this region. Attempts to establish parasitoid populations in prune orchards in early summer have not yet met with great success, but summer releases of parasitoids in *Phragmites* reed beds, one of the two summer host plants of MPA, led to substantial field parasitism in late 2006 and 2007. It is not yet clear whether it was the origin of the parasitoids or the focus on *Phragmites* that led to this success. However, these preliminary results were very promising, and suggested the need for additional releasing and monitoring both in prune orchards and *Phragmites* beds.

A dominant hypothesis from the biological control literature is that genetic matching, the selection of parasitoids from the same location as the source of an invasive pest, provides parasitoids that are better adapted to that particular genotype of the pest and that are best suited for use in biological control introductions. In addition, it has long been believed that climatic matching, the selection of parasitoids from a location that has the closest climatic match to that of the region that has been invaded by the pest, provides the best adapted parasitoids for biological control introductions. These two hypotheses may in some cases be complementary and lead us to focus attention on the same single location in the region of origin, but in other cases they may conflict and suggest two very different locations as the most suitable source populations of parasitoids for introduction. In the case of MPA, we know that southern Spain provides the best genetic match, but we currently do not know the location that provides the best climatic match.

latter case, in history of biological control no one has ever had the opportunity to test which of the two hypotheses may be the most important.

OBJECTIVES

1. To continue to field release and monitor different strains of *Aphidius transcaspicus* in prune orchards and *Phragmites* beds.
2. To compare the climatic match of two genetically distinct strains of *Aphidius transcaspicus* to the climate of California.

PROCEDURES, RESULTS, AND DISCUSSION

1. To continue to field release and monitor different strains of *Aphidius transcaspicus* in prune orchards and *Phragmites* beds.

Methods

In May 2007, parasitized mealy aphids were collected from *Prunus* trees from a number of locations in southern Spain. This region was selected as the most likely origin of the invasive populations of MPA in California as determined by genetic fingerprinting. Mummies were collected from 7 locations and were sent or hand carried to the quarantine facility at UC Berkeley for parasitoid rearing. Parasitoid colonies from populations collected in southern Spain and Morocco (strain K1 - western Mediterranean) and from Cyprus and Israel (strain K3 - eastern Mediterranean) were maintained in the Insectary and Quarantine Facility at UC Berkeley through 2007. This was reduced to southern Spain (K1) and Israel (K3) only through 2008, due to the failure of the Cyprus and Morocco colonies to survive the intervening winter.

Field releases of the different parasitoid strains were carried out in prune orchards in the Sacramento Valley from early May to mid June 2007 and 2008 and in *Phragmites* reed beds from August to October 2007. All parasitoids that were field released were reared on MPA on potted *Phragmites* rather than from black bean aphids on potted beans, as used in previous years. Although it is more difficult to produce parasitoids continuously on MPA it avoids the problem encountered in 2004, when parasitoids reared on black bean aphids began to reject MPA as suitable hosts. Some of the parasitoids were used for sleeve cage experiments to compare the performance of the different parasitoid strains, while others were released openly into the orchards or reed beds to allow the parasitoids greater freedom of movement in selecting trees and resting sites.

For the sleeve cage tests of the performance of the different parasitoid strains (K1 and K3), sets of 20 parasitoid individuals (10 males and 10 females) were collected from the colonies whenever a new generation of wasps began to emerge, and were enclosed on prune branches inside organandy sleeve cages that measured 0.75 inches in diameter and 3ft in length. The branches were selected to have young developing colonies of aphids, and to be in the lower canopy of the north side of the prune trees to provide maximum shade from direct sun. In 2007 the same sleeve cages were also used to enclose *Phragmites* reeds with newly developing aphid colonies. It is unknown exactly how many days the parasitoid adults survive in the sleeve cages, but it is assumed to be limited to 2-3 days. As it takes approximately 7-8 days for parasitoids to develop from egg to pupation (when mummification of the host aphid occurs) the sleeves were removed after 14 days to allow for all parasitized aphids to have reached the mummy stage. The number of mummies produced in each sleeve was noted, and the number of mummies per female parasitoid released into the sleeve was used as the measure of field performance.

Results and Discussion

In 2007, we were able to field release many more parasitoids than in previous years, due to the better production of parasitoids from aphids on *Phragmites*, and the sustained availability of aphids in the field both in prunes and *Phragmites*. While releases in prunes in 2008 were comparable to 2007, no releases were made in *Phragmites* due to the total absence of suitable aphid populations this year. A total of 3,575 parasitoids were field released in 2007, but only 1,900 in 2008 (Table 1). In both years the majority of the parasitoid released were the eastern Mediterranean strain (K3), but for the sleeve cages the numbers of replicate releases were more evenly matched between the eastern (K3) and western (K1) Mediterranean strains. With greater

emphasis on the open release of parasitoids from the eastern Mediterranean (K3) in prunes this year a significant number of mummies were found on trees surrounding release points in the field. Their abundance was not sufficient to impact the aphid populations, but this was a notable change from previous years where greater emphasis had been placed on open releases of parasitoids from the western Mediterranean (K1).

The decision to make releases into *Phragmites* reed beds in the Sacramento Delta was made in late summer 2006. Both that year and in 2007 these reed beds had heavy populations of aphids and open release of parasitoids was seen to result in the production of a number of mummies, indicating the potential of establishing parasitoids on these summer host plants. Early juvenile stages of the parasitoid are carried in the bodies of migrant aphids as they return from their summer host plants to prune orchards in the Fall. Thus the establishment of parasitoids on summer hosts would not only reduce populations that are able to make the return migration, but would also provide an effective means of naturally introducing parasitoids in prune orchards for overwintering and subsequent impact on aphids the following spring. The total absence of aphids from our field sites in the Sacramento Delta in 2008 is puzzling, but may in some way be associated with the herbicide treatments that are underway to remove this plant from the area.

From the perspective of genetic matching of parasitoids to aphids, the field performance of the eastern Mediterranean strain (K3) of *A. transcaspicus* showed consistently greater field performance than the western Mediterranean strain (K1) in sleeve cages (Fig. 1). For releases in prune orchards, the number of mummies produced per parasitoid released was 7-8 time greater in both 2007 and 2008. In both 2007 and 2008 K1 parasitoids were from Southern Spain, while K3 parasitoids were from both Cyprus and Israel in 2007, but from Israel alone in 2008 (Table 1). The comparative results from the two years are remarkably consistent in the magnitude of the difference in field performance of the two parasitoid strains, and in both years the difference was statistical significant.

Although the direction of field performance was the same for the sleeve cages used in *Phragmites* reed beds in 2007, the differences observed were not significant (Fig. 1). In this case K1 parasitoids came from Morocco as well as from southern Spain, which may have played some role, but it could also be that in contrast to prunes both parasitoids strains perform equally well in *Phragmites* beds. While mealy aphids in the genus *Hyalopterus* use distinct host plants for overwintering and early season population development, namely *H. amygdalus* on almond, *H. persikonus* on peach, and *H. pruni* on plums, these aphids all share a common host plant *Phragmites australis* for population development through the rest of the summer season. Thus all *A. transcaspicus* strains must have good capability of using mealy aphids on *Phragmites* and may differentiate little in terms of preference or performance on this summer host plant, in contrast to their performance on orchard trees.

2. To compare the climatic match of two genetically distinct strains of *Aphidius transcaspicus* to the climate of California.

Methods

An earlier attempt was made to compare the climates of the Mediterranean region with those of both the Sacramento and San Joaquin Valleys (see Fig. 5 of my report for 2004). In the absence of good comparative climatic data, this attempt consisted of comparing the mean annual rainfall, the mean January temperature and the mean July temperature for 21 locations around the Mediterranean with those of Marysville and Modesto. Since then with increasing interest in climate change around the world, several much more detailed climatic databases have been developed. In addition, several ecological niche models have appeared with the capability of comparing climates over different time periods for the same region, or for different regions over the same time period.

Here we use Maxent, an ecological niche model that has shown excellent performance for climate comparison. Maxent uses presence-only sampling data (i.e. the locations around the Mediterranean where we have found *A. transcaspicus*) and climatic data for those specific locations to generate a probability distribution of species occurrence over a given area using the principle of maximum entropy. For climatic data we used the WorldClim database which consists of 19 bioclimatic variables collected from globally distributed weather stations that represent annual trends, seasonality, and extreme or limiting climatic factors. To avoid over-parameterizing the Maxent model and reduce the potential for correlation among bioclimatic variables we first ran a Pearson correlation matrix for all 19 bioclimatic variables for 1000 randomly selected locations across the Mediterranean. For all variables with a correlation $r > 0.80$, we retained only one of the two variables and selecting for seasonality or extremes over annual means. This reduced the initial 19 variables to just 10: BIO2 – mean diurnal temperature range [(mean of monthly (maximum – minimum temperature)]; BIO3 – isothermality [(mean monthly temperature range/ annual temperature range) \times 100]; BIO4 - temperature seasonality (SD \times 100); BIO5 – maximum temperature of the warmest month; BIO6 – minimum temperature of the coldest month; BIO8 – mean temperature of the wettest quarter; BIO9 – mean temperature of the driest quarter; BIO13 – precipitation of the wettest month; BIO14 – precipitation of the driest month; and BIO15 – precipitation seasonality (coefficient of variation). We used logistic output from the Maxent model which generates a probability of occurrence (range 0-1) within each grid cell of a map, and during initial runs we randomly selected 70% of sampling localities as training data and 30% as test data. Separate Maxent models were developed for the localities where the K1 (western Mediterranean) and K3 (eastern Mediterranean) strains of *A. transcaspicus* had been found, and both models were mapped onto California to indicate the favorability of the Central Valley climate for each parasitoid strain.

Results and Discussion

Using 10 bioclimatic variables and known locations for the two strains of *A. transcaspicus* in the Mediterranean generated effective Maxent models for the natural distribution of these parasitoids in their area of origin. The average area under the receiver operating characteristic curve (AUC) was 0.96 for the training data and 0.94 for the test data. As values for AUC vary from 0 to 1, a value of greater than 0.9 is generally considered representative of a model that has an excellent fit to the data.

Projecting the two Maxent models onto the California landscape generated surprising differences in the suitability of our climate for these two strains of *A. transcaspicus* (Fig. 2). For the western Mediterranean strain (K1), the overall suitability of the California climate was lower than for the eastern Mediterranean strain (K3). However, there were three main regions of suitability (Fig. 2a). The most northerly region extends from central Tehama county down through Butte, Yuba and western Placer into Sacramento county. A second region spans eastern Fresno and Tulare counties, and a third region covers eastern Santa Barbara county eastward across the border between Kern and Ventura and Los Angeles counties. For the eastern Mediterranean strain (K3) there is greater suitability of the climate, with two main regions (Fig. 2b). One extends throughout the coastal counties of the state, with a small break at the border of Monterey and San Luis Obispo counties, and extending into inland counties such as Solano and Yolo around the San Francisco Bay area. The other runs from Shasta county down through Tehama, Butte and Yuba counties extending on down the eastern side of the valley into Tuolumne county. One important outcome from these projections is that they suggest that neither of the two parasitoid strains is well matched to the climate of the prune production counties of the San Joaquin Valley. For the Sacramento Valley, both strains show some suitability, with the eastern Mediterranean (K3) strain having a broader area of suitability through the prune production region than the western Mediterranean strain (K1).

These projections therefore suggest that the eastern Mediterranean strain (K3) of *A. transcaspicus* is climatically better matched to the prune orchards of California than the western Mediterranean (K1) strain. This information also provides an explanation for why the eastern Mediterranean strain (K3) performed better than the genetically matched eastern Mediterranean strain (K1) in sleeve cages in the Sacramento Valley. The observation of a significant number of mummies from open releases of the eastern Mediterranean strain (K3) can also be explained by the better climatic match of this strain.

CONCLUSIONS

The need to develop alternatives to dormant oil sprays for the control of aphids in prunes has generated interest in establishing parasitoids that will provide long term control of both leaf curl aphid and mealy plum aphid in prune orchards. *Aphidius colemani* has been successfully established against leaf curl plum aphid and populations of this aphid have diminished in recent years as a result. Early spring populations can still occur in orchards in the north and the west side of the Sacramento Valley, but these populations are generally confined to individual branches, they remain small, and seldom develop to spread between branches and trees within orchards. The establishment of a strain of *Aphidius transcaspicus* that could achieve the same impact on mealy plum aphids is an important goal that would be of tremendous value to the prune industry as the most cost-effective way to manage these aphids in prunes.

Attempts to establish *A. transcaspicus* in prune orchards have not yet been successful. We do see mummies when parasitoid adults are confined in sleeve cages, but until this year open releases of parasitoids rarely led to the development of any mummies in prune orchards although we did see mummies resulting from open releases in Phragmites beds in both 2006 and 2007.

However, as we were unable to find aphids in *Phragmites* in 2008 this summer host plant obviously does not provide consistent opportunities for parasitoid establishment, and can not be relied upon in the future as colonization sites for this parasitoid.

Our release efforts over the last three years have focused very much on open releases of the genetically most closely matched parasitoid strain (K1). The development of ecological niche models to compare the climatic match of the two key parasitoid strains from the Mediterranean region revealed for the first time that unfortunately the K1 strain appears less well suited to the climate of the prune production areas of the Sacramento Valley. In contrast the climatic match of the K3 strain to these areas is much better, and this year we focused our open releases on this strain which led to observations of significant numbers of mummies in the prune orchards. Thus, the use of the ecological niche models and the identification of the better climatically matched strain now offer a new strategy for securing the establishment of *A. transcaspicus* in California, the open field release of the eastern Mediterranean strain (K3) of the parasitoid directly into prune orchards in spring and early summer.

Parasitoid strain	Prunes				Phragmites		Total
	2007		2008		2007		
	Open	Slv	Open	Slv	Open	Slv	
Cyprus (K3)	0	90	0	0	1,225	240	1,555
Israel (K3)	300	90	1,000	520	700	75	2,685
Morocco (K1)	0	0	0	0	400	50	450
Spain (K1)	100	230	0	380	0	75	785

Table 1. A summary of the releases of *Aphidius transcaspicus* in prune orchards and *Phragmites* reed beds in 2007, indicating the geographic origin of the subpopulations of parasitoids used and whether releases were into sleeve cages or open.

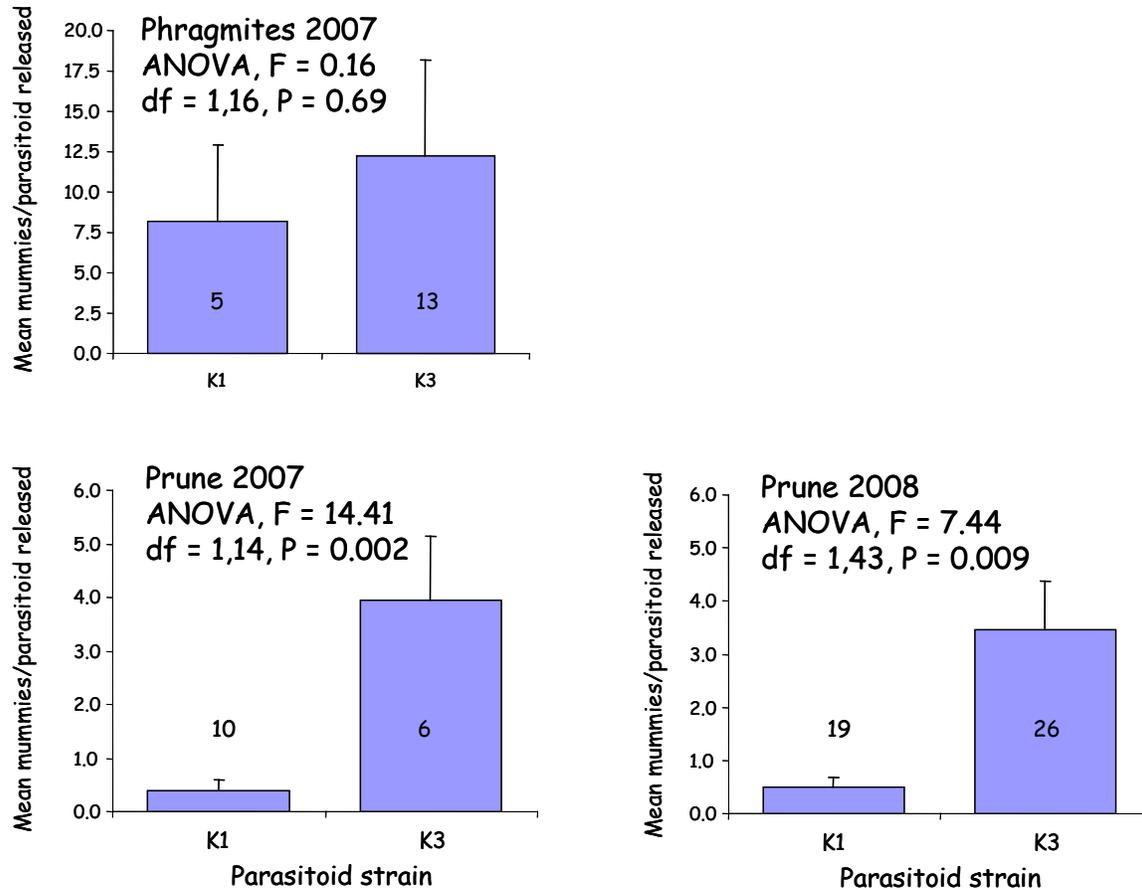


Fig. 1. The field performance of the western Mediterranean (K1) and eastern Mediterranean (K3) strains of *Aphidius transcaspicus* in sleeve cages in prune orchards in 2007 and 2008, and in *Phragmites* reed beds in 2007. The numbers inset in the bars are the number of replicates on each parasitoid strain.

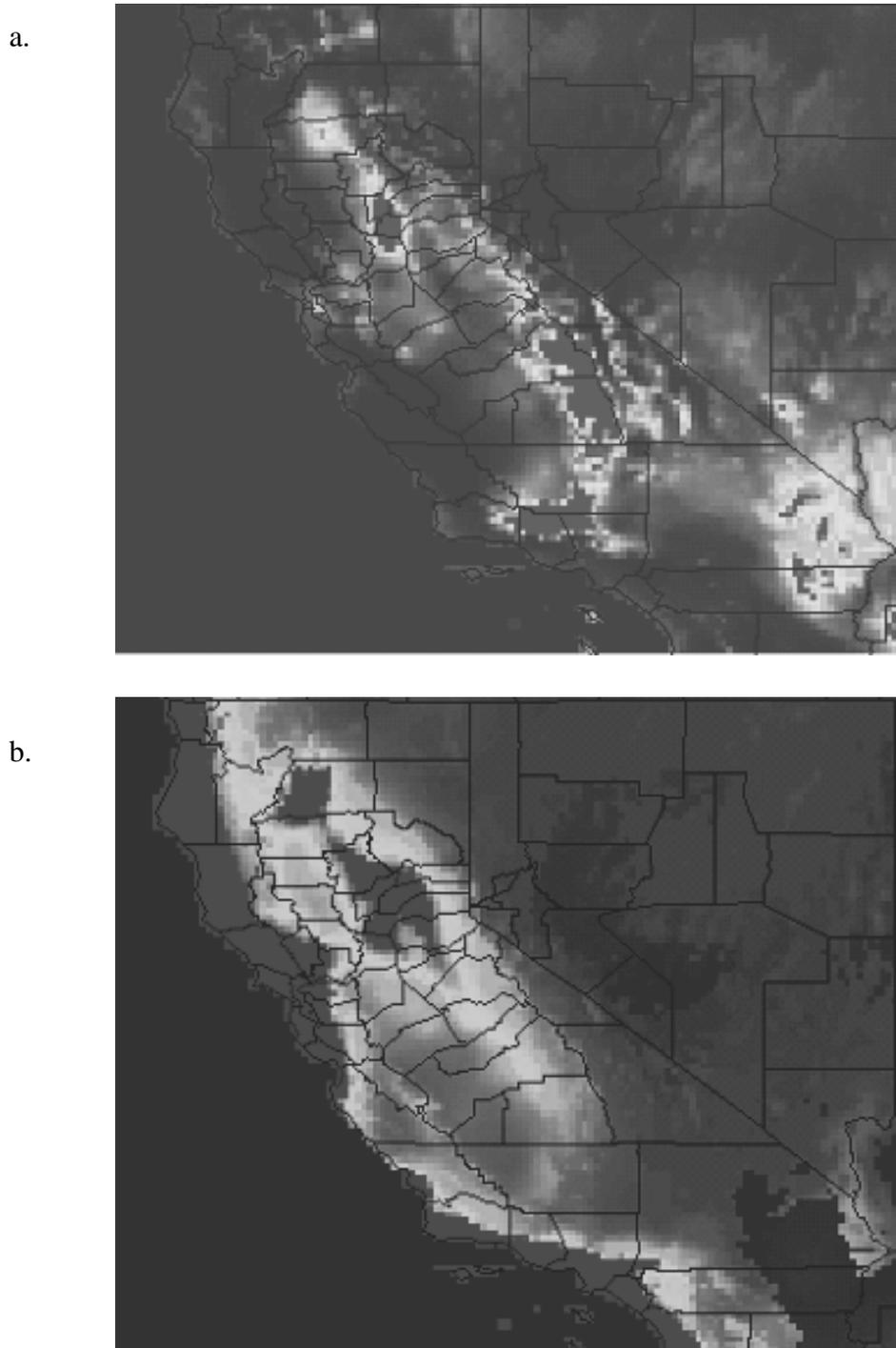


Fig. 2. Climate matching for (a) western Mediterranean (K1) and (b) eastern Mediterranean (K3) strains of *Aphidius transcaspicus*. The suitability of the climate in California for the two strains is represented by a color scale that varies from dark blue (suitability index of 0) to red (suitability index of 1).