# Developing Sampling Methods for Pre-Season Mite Detection and Implementing Management Decisions in Almonds

**Project No.:** **17-ENTO10-Rijal/Tollerup**

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**Objectives:**

1. Identify the new brown mites using morphological and molecular tools
2. Conduct studies to determine the seasonal phenology of the new brown mite species, and to develop sampling method(s).
3. Continue to explore methods for pre-season sampling for webspinning spider mites

**Interpretive Summary:**

The web-spinning group of the spider mites is a major pest problem in almond orchards in California. The three major mite species in this group are the Pacific spider mite (*Tetranychus pacificus*), twospotted spider mite (*T. urticae*), and strawberry spider mite (*T. turkestani*). The other group of the mites which do well under relatively cooler part of the year (spring) includes European red mites, *Panonychus ulmi* (Koch) and brown almond mite, *Bryobia rubrioculus.* In the past two years, our focus has been developing sampling methods to quantify the overwintering population of spider mites, and ultimately develop a pre-season mite sampling method for making monitoring and management decisions. We implemented soil sampling, ground vegetation sampling, and tree-band trapping methods to recover the overwintering mite population from the soil and tree. Based on the results of intensive sampling in five almond orchards covering both lower and upper San Joaquin Valleys, we found that these methods are not effective in estimating early-season mite population in almond orchards. Instead, we found that the tree-band trapping method is useful in recovering brown mites or similar species from tree trunks in almonds. Brown mites seem to be an increasing issue in almond orchards in recent years potentially due to the industry-wide reduction in a dormant spray which kills overwintering eggs of brown and European red mite eggs. There have been cases of elevated brown mite infestation that triggered the sprays to control brown mite both conventional and organic almond orchards. Although we did vegetation sampling and tree-band sampling for spider mites, the focus of 2017/18 study was to understand the phenology of brown mites in almonds and develop improved sampling strategy for brown mites. Below are some key findings and recommendations.

- Tree-band sampling (i.e., applying a layer of regular 2-inch duct tape on the tree trunk to recover moving mites) was used to capture brown mites effectively in almond orchards. However, this method was not effective in capturing overwintering spider mites.
- The use of the tree-band traps during the winter (February-April) should provide a general indication of in-season brown mite pressure in almonds. Studies will be conducted in 2019 season to validate this method for as a pre-season brown mite sampling method.
- We tested that the shoot sampling (i.e., jarring the twig/shoot of almonds to dislodge brown mites in a white paper, 8.5 in. x 11 in. size) can effectively be used to monitor in-season brown mite activity in almonds. Because of the sporadic feeding habit of the brown mites, the shoot sampling method is more reliable than the leaf sampling method for different levels of brown mite population.

**Materials and Methods:**

Objective 1: Identify the new brown mite using morphological and molecular tools
We are collecting brown mites from the leaves as well as from the tree-band traps in several times during the season. These all collected mite specimen will be sent to the Zalom lab at UC Davis Entomology, and to the California Department of Food and Agriculture (CDFA) for morphological identification. Since the PCR primer of the brown mite does not exist currently to compare, the molecular identification seems to be costly and time-taking, we will only pursue this approach if the morphological identification does not provide a confident result. This objective will be accomplished by the end of the year 2018.

Objective 2 & 3: Conduct studies to determine the seasonal phenology of the brown mite species, and to develop sampling method(s) for mite sampling
Four different methods were used to understand the phenology of mites in almond orchards;
1) Tree-band traps
2) Ground vegetation/soil sampling
3) Leaf
4) Shoot sampling

These techniques were used in three mature almond orchards located in Oakdale, Modesto, and Turlock of the Stanislaus County.

Use of tree-band traps**.** We have been using the tree-band traps to recover overwintering spider mites in the past two seasons. However, we were unable to detect any spider mites on these bands. Instead, we found the trap is useful in sampling brown mite species. In this method, a layer of regular duct tape (silver color, 2-in. wide) was used to create a band around the trunk of the tree to trap motile mites. The bands were applied at ~2-ft. high from the ground and left there for 2-4 weeks before replacing it with the fresh one. After removing the trap from the tree, a transparent plastic tape was placed over the sticky side of the duct tape throughout the length of the band and stored at 5 degrees Celsius for later evaluation. We evaluated 5-10 cm long portion of the tape and recorded the presence of the brown mites and webspinning spider mites.

Vegetation and soil sampling for spider mites. In past seasons, we took soil samples from 3 different distances covering multiple directions from the tree trunk, but we were unable to recover the overwintering spider mites from those samples. Because of that, we adopted a

slightly different method in which the collected sample comprised of the dried vegetation, plant debris, and a thin layer of surface soil underneath the tree. Eight samples for overwintering mite detection were taken from each of the ten random trees (within two consecutive border rows) from two sites, Oakdale and UCCE. Soil from each sample point was mixed properly and distributed into four transparent plastic cups (size: 8 oz.) (Walmart Inc.). The cups were then placed in the center of the sticky card (San Jose scale trap sticky card) so that mites after emergence from the soil crawled on the glued surface of the card and trapped there. After leaving outdoor under the shade for 3-4 weeks, the sticky cards were inspected for the spider mites using a microscope. All soil samples were taken during February.

Leaf and shoot sampling**.** Beginning late March, we started taking in-season leaf and shoot samples and record the brown mite population in almonds. The samples were taken from both interior and exterior portions of the tree. Since the distribution of mites is highly variable among the sampled leaves, we conducted both leaf and shoot sampling. The shoot is defined as the tuft of leaves developed from each bud on the twig (Summers and Baker 1952). The shoot sampling method was first described by Summers and Bakers (1952) to sample brown mites in almonds. We adopted that method with slight modification in our study. In this sampling, we used five shoots per tree and five trees per location (i.e., UCCE-Modesto, Oakdale, and Turlock). The shoot with a bunch of the leaves was jarred into the white copy paper (8.5 in. x 11 in.) using a small hand-held stick to dislodge brown mites into the paper, and the number of brown mites in all four quadrants of the paper were counted. For leaf sampling, we took leaf samples (15 leaves of each of the five trees) separately following the spider mite sampling protocol from the UCIPM Guidelines. The sampling was conducted bi-weekly from March through July.

**Results and Discussion:**

Tree-band traps for capturing mites in the trunk**.** The tree-band trap was effective in capturing brown mites in all three sites (Modesto, Oakdale, and Turlock). Brown mite adults were captured from the 2$^{nd}$ week of March through June with peak activities in April, and the 1$^{st}$ week of June (**Figure 1**). The June activity was much reduced in Turlock site potentially due the miticide included in the 'May spray.' Adult brown mites were not observed in February in any sites. Many brown mite nymphs were captured in February (>9 nymphs/cm trap) and March (>6.5 nymphs/cm trap) (**Figure 2**). The second peaks were observed in the 3$^{rd}$ week of May and 1$^{st}$ week of June in Oakdale and Modesto sites, respectively. As brown mites overwinter as egg stage, the early (February) high activity of nymphs was likely due to the hatching of those eggs during the delayed-dormant (Beers 2007). No overwintering spider mites were recovered from the traps, and this was true for studies conducted in earlier years as well.

The objective of the tree-band trap was to quantify the early-season population and determine its relationship to the seasonal mite population. In all three sites, we observed a trend that greater activity of brown mites in tree trunks earlier part of the season (Feb-April) contributed to the greater in-season brown mite infestation (**Figure 3**). This study will be continued for the 2019 season to validate the trend statistically.

Vegetation and soil sampling in detecting pre-season spider mite activity**.** Total 160 sub-samples (i.e., subsample size: 8 oz. soil volume) consisting of the orchard vegetation and surface soil were processed in 2018 season. Similar to the 2017 study results, no overwintering spider mite was recovered from any sampling location. Although there was a report of Pacific spider mites recovered using similar soil sampling method in Kern County (Zalom et al. 1995), our studies did not yield any overwintering mites in two consecutive years. There might be a couple of explanations for this. First, overall spider mite seasonal infestation tends to be considerably high in southern counties, and most likely that the overwintering spider mite population per unit area or per tree is high in heavily infested orchards and/or areas, resulting in successful mite recovery from the soil. Second, overwintering habit can be different for twospotted spider mite (*Tetranychus urticae*) which is predominant species in the northern portion of the central valley, then the Pacific mite (*Tetranychus pacificus*), the predominant spider mite species in the south. Third, the total surface area of the sampled portion (~1% of the total area) compared to the entire surface area under a tree might not have enough to detect overwintering mite population. Since spider mite population early in the season in the orchard is likely limited to clumped to the certain areas which act as a source of infestation for rest of the orchard later (Hoy et al. 1984), identifying those source trees for overwintering mite detection sampling is challenging.

Determining the phenology of brown mites using leaf and shoot sampling. There is a for conducting dormant spur sampling brown mites along with few other insect pests in almonds (UCIPM Guidelines, http://ipm.ucanr.edu/PMG/C003/m003dcdmtspursmpl.html). However, no guideline has been developed to detect and monitor in-season brown mite population. With increased activity of brown mites in almonds in recent years, it is important to have a sampling method that can be used to monitor brown mites, that potentially guide in deciding treatment decisions.

For leaf sampling, we followed the modified version of the UCIPM Guidelines for leaf sampling in which 15 leaves from each of the five trees were inspected for the brown mites as well as predators using a hand lens and recorded their numbers. For shoot sampling, five random shoots from each of the five sample trees were used to dislodge brown mites into a letter size paper and recorded the brown mites from the paper. We started the leaf and shoot samplings in mid-to-late April. We observed a similar trend in brown mite counts over time in both leaf and shoot samplings (see the trend line in the (**Figures 4-9**). The mite counts were in peak around late April and early May and slowly declined as the season progressed in the summer. Based on regression analysis combining all data points from three sites ($n$ =18), there was a strong linear relationship ($R^2$ = 0.44; $df$ = 16; $P$ = 0.0028) between mite counts using leaf and shoot sampling methods (**Figure 10**). Although brown mite counts using two methods were correlated, the leaf sampling method does not accurately represent the overall brown mite population of the tree (Summers and Baker 1952) due to the influence of the environmental factors such as sunlight and temperature in their feeding habit. Brown mites tend to switch between resting and feeding and move back and forth between the shoot and the leaf during these phases. These mites also have the tendency to avoid direct sunlight when the temperature is high (>80F). Because of this behavior, shoot sampling which covers both leaf and the wood provides a more accurate representation of the population and can be effective even under a low mite infestation as well. Since brown mite is still considered as a minor pest, the economic threshold for this pest is yet to be determined. The effective sampling method for

brown mites is also critically important to assess its population. We plan to continue these aspects of the sampling scheme in the future.

## Figures

**Figure 1**. Average brown mite adults per cm of the tree-band trap in almonds across three sites

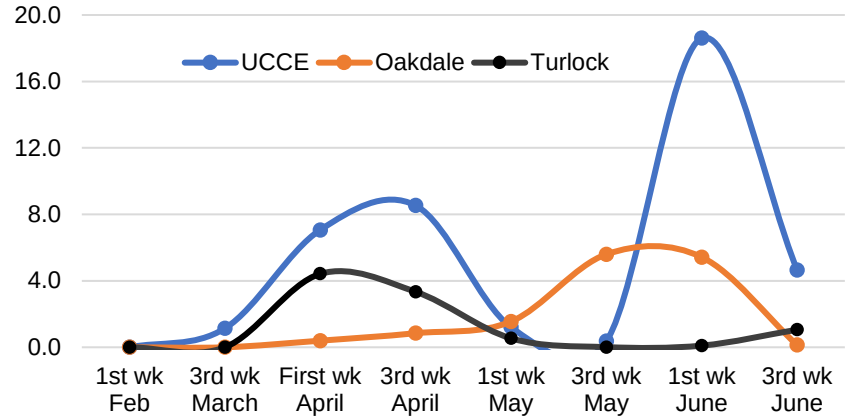



**Figure 2**. Average brown mite nymphs per cm of the tree-band trap in almonds across three sites

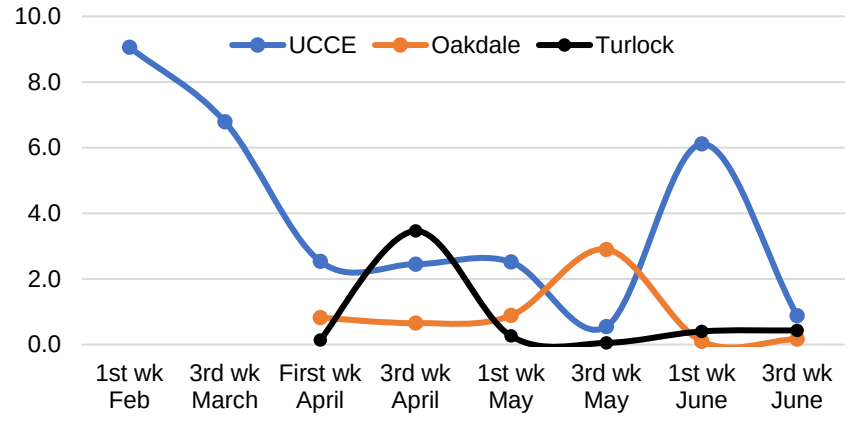


**Figure 3**. Overall trend comparison between mites captured in tree-band traps (mites/cm; Feb-April) and in shoot sampling (mites/shoot; seasonal)

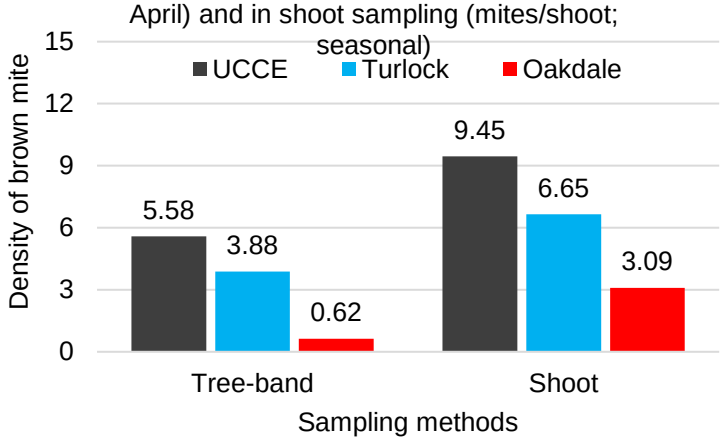



**Figure 4**. Average number of brown mites per 15 leaves, UCCE site

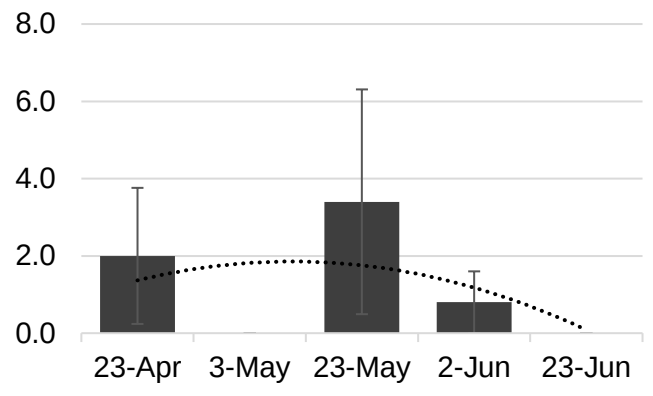



**Figure 5**. Average number of brown mites per 5 shoots, UCCE site

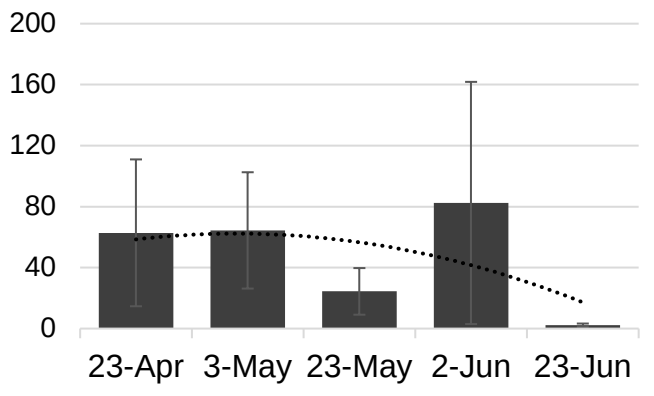



**Figure 6**. Average number of brown mites per 15 leaves, Oakdale site

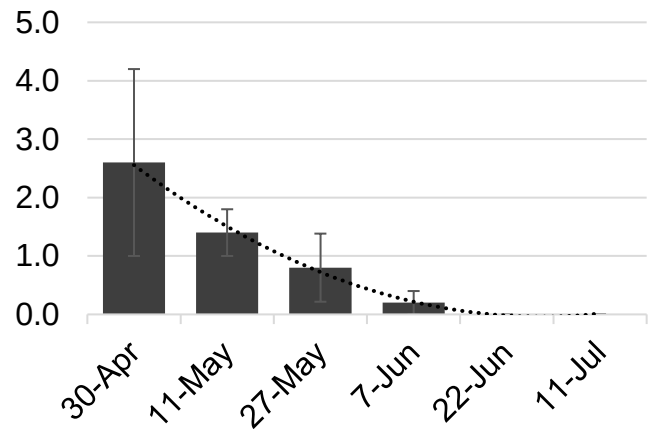



**Figure 7**. Average number of brown mites per 5 shoots, Oakdale site

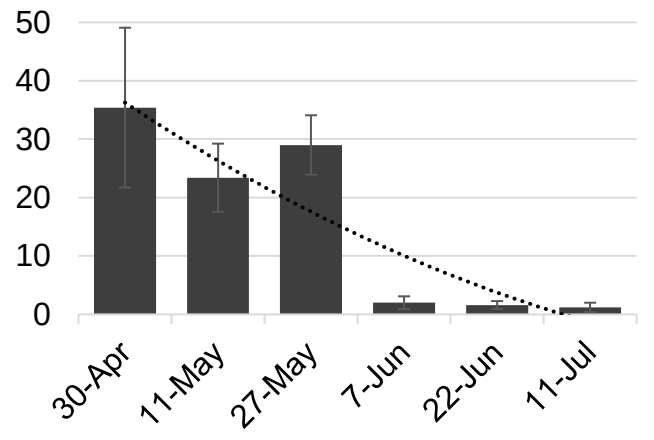


**Figure 8**. Average number of brown mites per 15 leaves, Turlock site

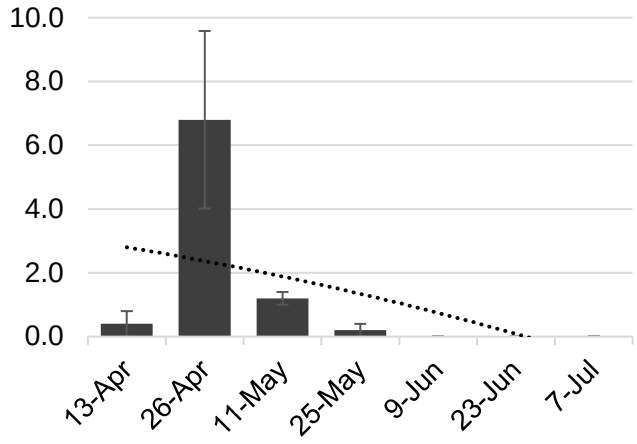



**Figure 9**. Average number of brown mites per 5 shoots, Turlock site

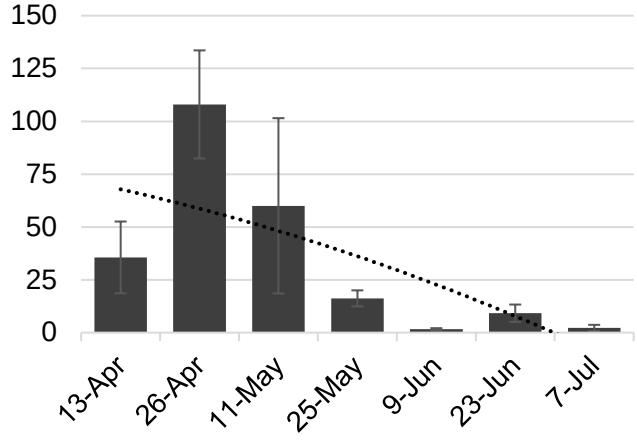
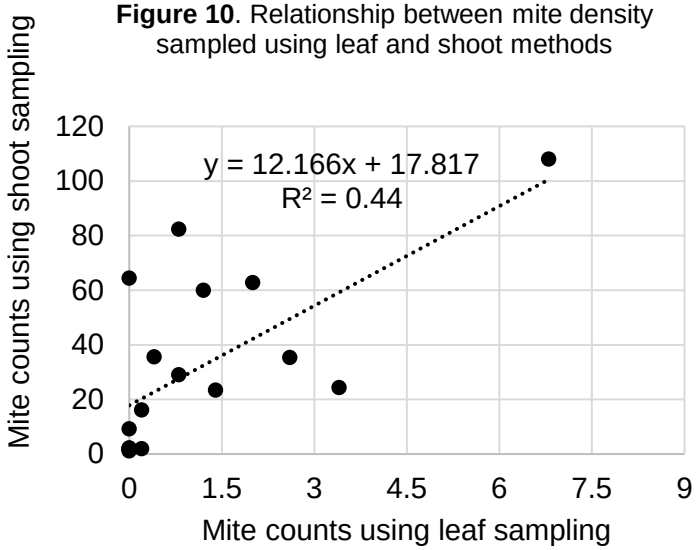



**Figure 10**. Relationship between mite density sampled using leaf and shoot methods




## Research Effort Recent Publications:


Rijal, J.P., and K. Tollerup. 2017. Developing sampling methods for pre-season mite detection in almonds. Report submitted to the Almond Board of California.

Rijal, J. P. (2016). Spider mites in almonds: monitoring and management. Field Notes Newsletter (May Issue).

Rijal, J. P. (2016). IPM of Spider mites in almonds: exploring the new sampling techniques. CAPCA Adviser Vol. 19, No. 4 (August).

Rijal, J. P. (2016). Research to better understand spider mite migration timing. Nuts 'n' Bolts section, Pacific Nut Producer Newsletter Vol. 22, No. 6 (June).

Rijal, J.P., and K. Tollerup. 2017. Poster: Developing sampling methods for pre-season mite detection in almonds. Almond Board of California Annual Conference, 5-7 December, Sacramento, CA.

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Summers, F. M. and G. A. Baker. 1952. Procedure for determining relative densities of brown almond mite populations on almond trees. Hilgardia. 21 (13): 369-382.

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