SPCB RESEARCH TRACKING

RESEARCHER	TRACKING	CONTRACT BALANCE
Dr. Michael Rust University of California, Riverside Agreement No. 26732 "Development and Evaluation of Baiting Strategies for Control of Pest Yellowjackets in California" Original Term Dates: 10/23/18 - 12/31/20 Amended Term: 10/23/18 – 12/31/21 **Total Contract: $280,017.00**	10/23/18 – UC Riverside notified of contract approval effective 10/23/18. 1/11/19 – received invoice #80108-001 for $141.99 4/18/19 – received April 2019 Progress Report 5/11/19 – received invoice #80108-002 for $6,093.28 7/17/19 – received invoice #80108-003 for $21,870.43 10/16/19 -received invoice #80108-004 for $12,361.04 1/14/20 – received invoice #80108-005 for $18,431.65 4/6/20 – received invoice #80108-006 for $20,484.70 7/17/2 – received invoice #801808-007 for $16,767.87 9/5/20 – requested progress report, progress report extended to 10/4/20 to allow a more informative report. 10/14/20 – received progress report 11/5/20 – received invoice #80108-008 for $28,328.52 11/20/20 - *Pending no cost extension from December 31, 2020 to December 31, 2021. 2/4/21 – received invoice #80108-009 for $32,369.60 2/25/21 – No-Cost extension approved by BSO. Term extended to 12/31/21 4/29/21 – Received invoice #80108-011 for $12,107.51 7/16/21 – Received invoice #80108-012 for $67,365.25 10/19/21 – Received invoice #80108-013 for $13,363.49 1/31/22 – received Final Report **Total Expenditures: $249,688.33**	**$30,328.67**
Dr. Niamh Quinn University of California, Agriculture and Natural Resources Agreement Number: 26727 "Investigation of Rodenticide Pathways in an Urban System Through the Use of Isotopically Labelled Bait" Original Term Dates: 10/16/18 – 12/31/20 Amendment #1 Term: 10/16/18 – 6/30/22 Amendment #2 Term: 10/16/18 – 6/30/23 **Total Contract: $329,749.00**	10/16/18 – UCANR notified of contract approval effective 10/16/18. 4/30/19 – Received April 2019 Progress Report 1/27/20 – received invoice #56318501 for $11,947.50 7/28/20 – received invoice 76c59-02 for $0.00 9/15/20 – received progress report 11/20/20 – received invoice #59174298 for $27,877.50 1/27/21 – No-cost extension approved to change term date from 12/31/20 to 06/30/22. 6/2/21 – No-cost extension requested to change term from 6/30/22 to 6/30/23 1/25/22 – Received fully executed amendment #2 2/2/22 – Received invoice #63702836 for $20,660.79 **Total Expenditures: $60,485.79**	**$269,264.21**

Neil Tsutsui University of California, Berkeley Agreement Number: 26735 "Diet and Colony Structure of Two Emerging Invasive Pest Ants" Original Term Dates: 10/18/18 - 08/31/21 Proposed Amended Term: 10/18/18 – 6/30/22 **Total Contract: $146,325.00**	10/18/18 – UC Berkeley notified of contract approval effective 10/18/18. 1/3/19 – received invoice #GM00159910 for $6,079.05 1/29/19 – received invoice #GM00162310 for $7,011.98 2/25/19 – received invoice #GM00166580 for $2,000.00 4/7/19 – received April 2019 Progress Report 5/29/19 – received invoice #GM00175634 for $681.23 7/2/19 – received invoice #GM00178838 for $1,220.99 8/9/19 – received invoice #GM00184114 for $22,099.22 8/19/19 - received invoice #GM00186274 for $764.23 9/19/19 – received invoice #GM00188490 for $10,290.87 10/19/19 – received invoice #GM00190757 for $517.02 11/19/19 – received invoice #GM00193312 for $827.24 12/19/19 – received invoice #GM00196412 for $2,849.02 1/20/20 – received invoice #GM00197182 for $1,259.45 2/19/20 – received invoice #GM00200261 for $174.19 3/19/20 – received invoice #GM00204264 for $239.20 4/20/20 – received invoice #GM00208324 for $2,696.44 5/19/20 – received invoice #GM00212124 for $7,394.14 6/19/20 – received invoice #GM00215027 for $16,451.16 8/6/20 – received invoice #GM00218961 for $6,644.52 8/19/20 – received invoice #GM00221330 for $6,499.04 10/12/20 – received October 2020 Progress Report 10/29/20 – received invoice #GM00228610 for $11,816.46 5/4/20 – received invoice #GM00248340 for $665.71 5/28/21 – received invoice #GM00251772 for $1,158.01 6/4/21 – Pending no-cost extension to change term date to 6/30/22 and change personnel. Pending at BSO for processing. 7/2/21 – received invoice #GM00254509 for $1,035.23 8/19/21 – received invoice #GM00262008 for $3,776.59 9/20/21 – received invoice #GM00265799 for $1,949.57 10/19/21 – received invoice #GM00269199 for $3,990.90 11/19/21 – received invoice #GM00272585 for $5,510.81 12/20/21 – received invoice #GM00276627 for $9,949.86 1/20/22 – received invoice #GM00279831 for $3,949.83 2/19/22 – received invoice #GM00283223 for $4,137.42 **Total Expenditures: $143,639.37**	**$2,685.63**

Dr. Andrew Sutherland University of California, Agriculture and Natural Resources Agreement Number: 26730 "Evaluation of bait station system efficacy for reduced-risk subterranean termite management in California" Original Term Dates: 10/10/18-08/31/21 Amended Term Date: 10/18/18 – 8/31/22 **Total Contract: $190,425.00**	10/10/18 – UCANR notified of contract approval effective 10/10/18. 12/11/18 – received invoice #51140867 for $270.67 12/19/18 – received invoice #51464298 for $1,075.53 3/4/14 – received invoice #52326394 for $3, 671.22 4/2/19 – received invoice #52526107 for $2,617.68 4/26/19 – received April 2019 Progress Report 5/1/19 – received invoice #52892570 for $4,179.03 5/30/19 – received invoice #5330024 for $3,220.42 7/26/19 – received invoice #54113894 for $4,040.68 10/3/19 – received invoice #54886547 for $272.95 11/13/19 – no cost extension approved by BSO to extend contract term from August 31, 2021 to August 31, 2022. 1/21/20 – received invoice #56314886 for $1,475.42 3/26/20 – received invoice #57095974 for $12,702.80 5/4/20 – received invoice #57413857 for $6,097.63 5/14/20 – received invoice #57647938 for $2,383.03 6/19/20 – received invoice #57984215 for $22,324.44 7/23/20 – received invoice #58296943 for $4,581.79 9/5/20 – requested progress report 9/14/20 – received September progress report 10/21/20 – received invoice #59172744 for $6,091.16 11/30/20 – received invoice #59515731 for $6,893.52 2/4/21 – received invoice #59990730 for $17,611.97 2/25/21 – received invoice #60260692 for $1,881.22 3/23/21 – received invoice #60542078 for $3,141.40 5/28/21 – received invoice #61217379 for $5,277.38 6/17/21 – received invoice #61535348 for $5,148.25 7/23/21 – received invoice #61886116 for $2,382.03 8/19/21 – received invoice #62226637 for $2,777.03 9/28/21 – received invoice #62526387 for $7,371.99 11/5/21 – received invoice #62841155 for $2,821.88 12/7/21 – received invoice #63174937 for $3,754.81 12/16/21 – received invoice #63453845 for $2,640.82 1/28/22 – received invoice #63701704 for $1,866.86 3/8/22 – received invoice #64020822 for $2,583.05 **Total Expenditures: $141,156.66**	**$49,268.34**

Dr. Dong-Hwan Choe University of California, Riverside Agreement No. 26710 "Improving Urban Pest Ants Management by Low-Impact IPM Strategies" Original Term Dates: 10/22/18 - 12/31/19 Amended Term Dates: 10/22/18 – 8/31/20 **Total Contract: $77,309.00**	10/23/18 – UC Riverside notified of contract approval effective 10/22/18. 1/28/19 – received invoice #80105-001 for $689.61 4/30/19 – Received April 2019 Progress Report 5/11/19 – received invoice #80105-002 for $2,645.77 7/17/19 – received invoice #80105-003 for $3,468.85 10/17/19 – received invoice #80105-004 for $29,042.96 1/24/20 – received invoice #80105-005 for $17,532.01 **Pending no cost extension. Extends current contract from December 31, 2019 to August 31, 2020. 4/3/20 – Contract amended to reflect new extension date 4/28/20 – received invoice #80105-006R for $16,748.06 7/17/20 – received invoice #80105-007 for $6,713.11 9/4/20 – emailed Dr. Choe requesting final report due beginning of December 2020. Asked Dr. Choe to prepare a presentation for March 2021 board meeting. 10/21/20 – received final report 11/4/20 – received invoice #80105-008 for $468.63 March 2020 – Presented final report to Board Members at the March 2020 Board Meeting. **Total Expenditures: $77,309.00**	**$0.00**

3/9/2022

Development and Evaluation of Bait Strategies for Control of Pest Yellowjackets in California
October 22, 2018 – December 31, 2021

This final report is divided into sections, covering research conducted in one of our study regions: the San Francisco Bay, the Lake Tahoe area, and southern California (five sites). Conditions varied at each location depending upon human use patterns (recreational park, RV park, wild animal park, etc.), weather, and local factors (elevation, bears, etc.). Consequently, modifications in the monitoring and baiting protocols were sometimes necessary to accommodate these differences.

This project was initiated in August 2018 (prior to the release of funding) so that some initial trapping and baiting studies could be conducted before the end of the 2018 yellowjacket season. Sites were selected in southern California, the San Francisco Bay Area, and the Lake Tahoe area based on historical yellowjacket pest problems. The COVID pandemic impacted the 2020 and 2021 testing protocols. Travel limitations and additional technical assistance at the various field sites resulted in a considerable savings of research funds. A no-cost extension to include the summer of 2021 was approved.

Two different monitoring threshold protocols have been proposed prior to baiting for yellowjackets. Grant et al. (1968) proposed that baiting should commence when 7 yellowjackets/trap/day (YJ/T/D) were attained. Rust et al. (2010) adopted a slightly higher threshold of 10 YJ/T/D. We have chosen to use the higher threshold. There were occasions when baiting trials were conducted when the trap counts were not above the threshold. Requests by local cooperators had to be considered.

Methods and Materials

Monitoring

Placer-Style Trap – The Placer-style yellowjacket trap is a durable, reusable design. A 969-ml plastic screw-top jar (32 oz., Carolina Biological Supply Company, Burlington, NC) with an opening of 120 mm diam. was modified by cutting a hole in the center of the lid and inserting the plastic funnel portion of a Rescue fly trap (Sterling International Inc., Spokane, WA) from which the plastic bag had been cut away. The funnel was secured to the jar lid with rivets. On the inside of the clear plastic jar were two vials (12 dram and 9 dram, Bioquip Products, Rancho Dominguez, CA), with the outer vial secured to the jar with hot-glue and acting as a sleeve to secure an inner vial containing attractant (heptyl butyrate or minced chicken). The volatile chemical lure heptyl butyrate is highly attractive to western yellowjacket workers and queens (Simmons 1991, Landolt et al. 2003). Placer-style traps were hung in trees to prevent bears and other animals from disturbing them (Fig. 1). These traps were utilized at the Lake Tahoe sites.

Fig. 1. A Placer-style yellowjacket monitoring trap in the field. These traps were baited with minced canned chicken or heptyl butyrate.

UCR-Style Trap – The foraging activity of yellowjackets was also monitored using a UCR-style trap constructed from a 946 ml (32oz.) plastic bleach bottle. A hole was drilled in the bottle, and a 9- or 13-ml glass vial was screwed into the hole. The glass vial contained a 7.6-cm piece of dental wick and 8 or 13 ml of heptyl butyrate (Reierson and Wagner 1975, Reierson et al. 2008). Wasps that entered the trap through 5 side ports were funneled into a bottom jar containing a solution of antifreeze coolant diluted with water (propylene glycol 70:30 vol:vol, Sierra® Antifreeze/Coolant, Old World Industries, Inc., Northbrook, IL, Fig. 2). The traps were hung from a piece of wire under trees and bushes about 0.5-1.5 m off the ground. The traps were inexpensive and re-useable. The heptyl butyrate vials were changed as needed.

The UCR-style traps were used at the Richmond Field Station, Irvine Regional Park, Ronald W. Caspers Wilderness Park, Tree of Life Nursery, UCR campus, and Silent Valley RV Park.

Fig. 2. The UCR-style trap with re-useable and interchangeable components.

Rescue Disposable Yellowjacket Traps – The Rescue Disposable Yellowjacket Traps (Model #YJTD-W, Sterling International Inc., Spokane WA) were used at the San Diego Zoo Safari Park (Fig. 3). The trap cost about $8.00, but it was very effective in trapping yellowjackets. The traps were hung under trees and bushes and from Shepard's hooks about 0.5-1.5 m off the ground. The bag contained hundreds of beads with heptyl butyrate. Instead of adding pure tap water to the trap's collection bag, a solution of propylene glycol coolant was made with water (1:2) and added to the collection bag. The solution was effective in killing and preserving the insects. The contents of the bag were removed, and the excess fluid drained. The contents were placed into 3.7-liter plastic zip lock bags and shipped to UC Riverside, where the number and species of yellowjackets were counted.

Fig. 3. Disposable Rescue yellowjacket trap used at the San Diego Zoo Safari Park.

Bait Stations

UCR Wooden Bait Stations – The UCR-style bait stations were constructed from two pieces of pine board about 18 x 18 cm and 1.8 cm thick and a piece of 2.54 cm hardware cloth (72 x 14 cm, Fig. 4). The hardware cloth was stapled to the edges of the boards to construct a cage (18 x 18 x 14 cm). The hardware cloth on one side of the cage was not stapled to the wood allowing for bait cups to be placed inside the cage. The opening was held closed with a twist tie. The bait stations were hung from a piece of wire and a Perky-Pet® ANT GUARD® (Woodstream Corp., Lititz, PA) to prevent ants from feeding on the baits.

These bait stations were used at the Richmond Field Station, Irvine Regional Park, Ronald W. Caspers Wilderness Park, UCR campus, and Silent Valley RV Park. In 2018, UCR stations were used at Lake Tahoe sites.

Fig. 4. A wooden bait station hung from tree to prevent bears and wild animals from disturbing them.

UCR Plastic Circular Bait Stations – The stations were constructed from plexiglass disks (29.2 diam, 5 mm thick, 2.54 cm PVC pipe and hardware cloth (1.27 by 2.54 cm mesh, Fig. 5). A ring of hardware cloth (8.9 by 91.7 cm) separated the disks and allowed yellowjackets to enter the bait station. The bait stations were hung from a bush, tree, or Shepard's hook with a wire. A Perky-Pet® ANT GUARD® (Woodstream Corp., Lititz, PA) prevented ants from feeding on the baits.

These bait stations were used at the San Diego Zoo Safari Park and Ronald W. Caspers Wilderness Park.

Fig. 5. The plastic circular bait stations used at the San Diego Zoo Safari Park.

Placer Bait Stations – The bait stations were constructed from black two-gallon buckets with four 13 cm by 13 cm openings cut into the sides (Fig. 6). The openings were covered with flexible plastic mesh poultry fencing material with 2 x 2-cm openings in the mesh to allow yellowjackets to enter and exit. Bait stations contained 3 cups filled with ≈ 25 g bait each and were hung along the trap line in an area of high yellowjacket activity. The station was suspended from a wire and a Perky-Pet® ANT GUARD® (Woodstream Corp., Lititz, PA) to prevent ants feeding on the baits.

The Placer-bait stations were only used at the Lake Tahoe sites.

Fig. 6. The Placer bait station hung from a tree to prevent bears and wild animals from damaging the stations.

Evaporation Controls –The above bait stations were modified to determine the amount of water loss from baits and food attractants during the baiting. The openings were covered by with window-screen (1 mm mesh) to exclude yellowjackets (Fig. 7). The evaporation stations were hung near the bait stations during the test.

Fig. 7. The openings in the bait stations were covered with window screening to prevent yellowjackets from foraging on the baits or food attractants.

To adjust for the water that evaporated from the baits during the exposure period, bait or food materials were placed in salsa cups in the above stations covered with window screen. The salsa cups and lids were weighed and ≈ 30 g of bait or food material was put in the cup. The entire cup (cup + lid + bait) was weighed. After the exposure period, the cups were sealed with the lids, returned to the laboratory, and weighed. The ratio of water loss was determined as [Evaporative Initial Bait weight (EIBw)-Evaporative Final Bait weight (EFBw)/Evaporative Initial Bait weight (EIBw)] for the cups placed in the evaporation control stations. The average ratio of the EIBw/EFBw was also calculated.

The following calculation was used to determine the amount of bait or food material taken by the yellowjackets with corrections for the water loss of the bait remaining at the end of the exposure. The amount of bait taken = Initial Bait weight – [Avg. EIBw/EFBw x (Final Bait weight)].

UC Berkeley Richmond Field Station, 2018

The UC Berkeley Richmond Field Station is approximately 9.7 km northwest of the main UC Berkeley campus (37° 54'47.57" N, 122° 20'02.93" W, elev. 28 m). It consists of 68.8 ha of which 40.5 ha are uplands and the remaining acreage is marsh or bay lands. This area is a natural coastal grasslands environment (Fig. 8).

Fig. 8. UC Berkeley Richmond Field Station coastal grasslands and marsh habitat.

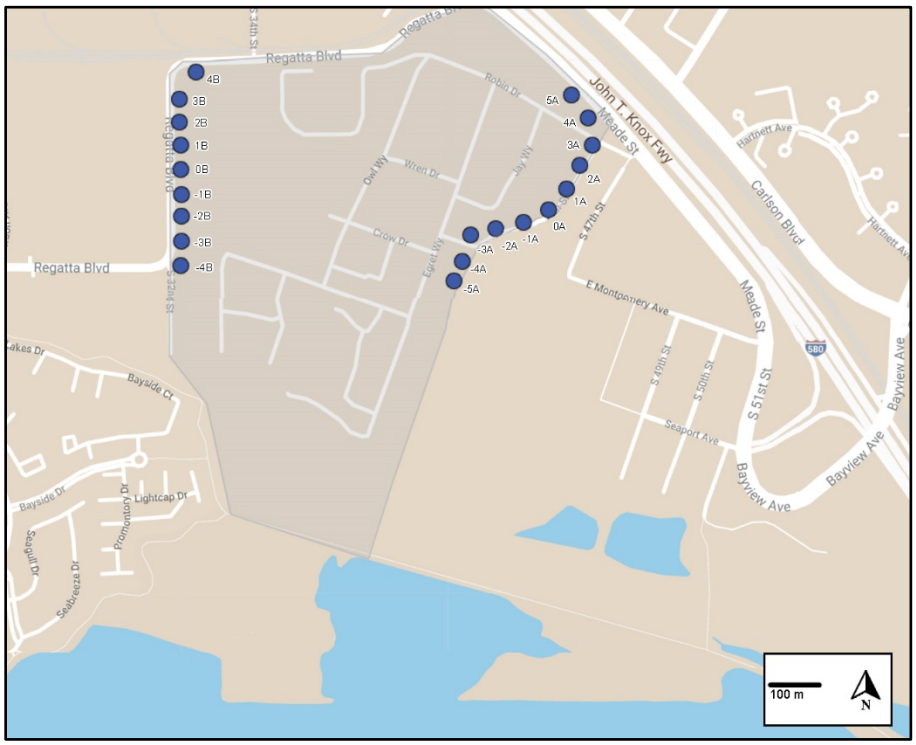

Fig. 9. The 2018 map of the UC Richmond Field Station with West and East Transects. UC Field station property outlined in grey.

Methods and Materials

Monitoring

The foraging activity of *V. pensylvanica* yellowjackets was monitored using UCR-style traps. Two monitoring transects (East and West) were maintained during the period 8/29/2018 to 10/2/2018, with three monitoring events (Fig. 9). The traps were hung on fences about 1.5 m off the ground (Fig. 10).

Choice Baiting Trial

The liquid contents from cans of Swanson's White Premium Chunk Chicken (Campbell Soup Co., Camden, NJ) were strained through cheesecloth. The chicken juice was then diluted with water (1:2) to make a 600 ml suspension to which 40 g of polyacrylamide crystals (PAA, Watering Storing Crystals, Miracle-Gro Lawn Products, Inc., Marysville, OH) were added, resulting in a PAA hydrogel matrix. A 0.1% aqueous solution of dinotefuran (Alpine 40WSG, BASF Corp., Research Triangle Park, NC) was prepared and added to the PAA hydrogels to make bait formulations containing 0.0025, 0.001 and 0.00075% active ingredient (AI).

A choice trial, evaluating yellowjacket preference for these formulations, was conducted on 9/5/2018 along the East transect. Salsa cups (59.1 ml) and lids were weighed, and 30 g of bait was added to each cup. The cups were weighed again. All three bait formulations were deployed in each of three UCR-style wooden bait stations, retrieved after 24 hours, returned to the laboratory, and weighed. To determine the amount of water lost to evaporation during deployment from the hydrogels, pre-weighed cups of each formulation were placed in one UCR-style bait cage covered with fine window screen (Fig. 8). These bait cups were also retrieved, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The yellowjacket trap catches before and after baiting were compared using a Wilcoxon signed-rank test. The amounts of each bait formulation that were removed in the choice test were compared with a one-way Anova.

Fig. 10. Bait station (yellow) and the evaporation control station.

Results

Monitoring

The only species collected in the traps was the western yellowjacket, *V. pensylvanica*. During the three monitoring events, 8,727 yellowjackets were trapped. The average trap counts in the East transect exceeded the threshold of 10 YJ/T/D at the first monitoring on 9/4/2018. The numbers of yellowjackets trapped in the West transect (untreated) peaked on 9/24/2018 (Fig.11).

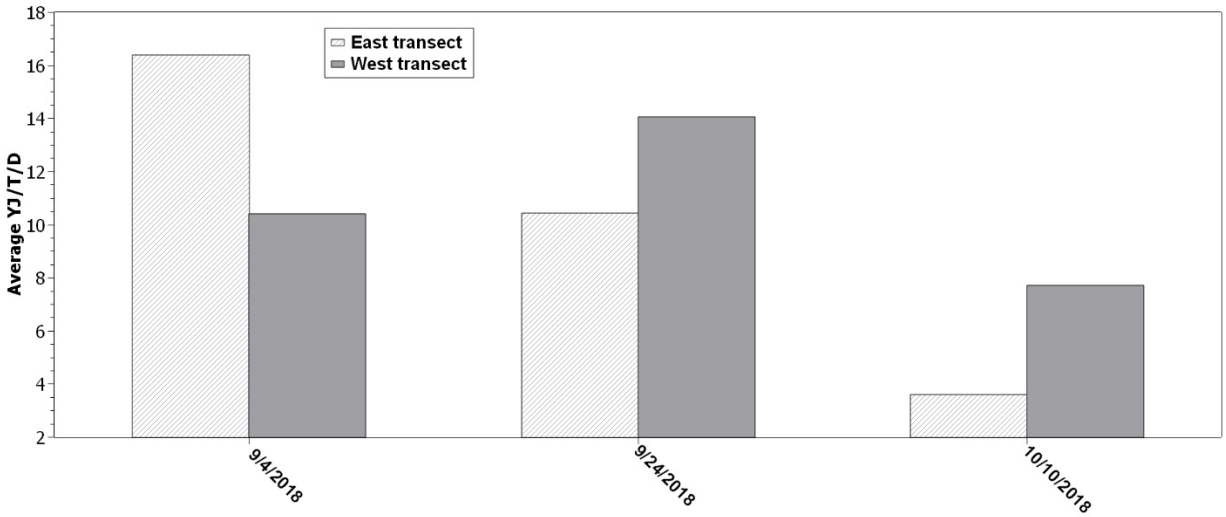

Fig. 11. The average number of yellowjackets/trap/day (YJ/T/D) before and after the choice bait test.

Choice Trial- Evaporation of water from the hydrogels was calculated to be about 17.6% of initial hydrogel mass during the 24-hour deployment period. Accounting for this loss of water, 32.9 g of dinotefuran bait was removed from the three different bait formulations: 0.0025% (10.2 g), 0.001% (9.5 g), and 0.00075% (13.2 g). There was no significant difference in the amount of each concentration of bait removed ($F = 2.62$; $df = 2,6$; $\mathbf{P} = 0.152$).

The yellowjacket trap counts along the East transect declined by 36.4% 18 days after the choice test, a statistically significant difference ($W = 46$, $Z = 2.02$, $\mathbf{P} = 0.043$). Trap counts were further reduced 26 days after baiting, with 65.3% fewer wasps trapped as compared with counts prior to baiting ($W = -66$, $Z = 2.91$, $\mathbf{P} = 0.001$). Along the West transect (considered as untreated control), there were no significant differences in trap counts before baiting and at days 18 ($W = -5$, $Z = -0.33$, $\mathbf{P} = 0.745$) and 26 ($W = 23$, $Z = 1.33$, $\mathbf{P} = 0.183$) after the choice test.

Discussion

The trap catches declined significantly in the East transect during both sampling periods after the choice test. The West transect (approximately 457 m from the East transect) was kept untreated to serve as the seasonal check (control). Trap catches in this untreated transect did not significantly decrease over our three sampling periods. In total, trap catches decreased by 78% in the East transect while trap catches in the West transect decreased by only 26%, perhaps in response to decreasing daily temperatures and photoperiods. Not enough of the dinotefuran bait was removed to reduce the number of yellowjackets trapped.

UC Richmond Field Station, 2019

Three transects were utilized at the Richmond Field Station during 2019, with 11 weekly monitoring events, beginning 5/6/2019 and ending 10/16/2019 (Fig. 12).

Fig. 12. Map of the UC Berkeley Richmond Field Station showing the locations of the three transects used during the 2019 trapping season.

Methods and Materials

Monitoring

In addition to the two transects used during 2018 (West = B, East = A), a third transect (transect C), with seven yellowjacket traps, was added along the southern boundary of the field station. Transect B was also shifted to the north to increase distance from the new transect C.

Bait Matrix Preference Trial

On 8/27/2019, two hydrogel matrices, the polyacrylamide crystals (PAA) and a novel seaweed alginate hydrogel (ALG), were deployed in tandem, without a toxicant, at two locations alongside evaporation controls (Fig. 8). The PAA gels were prepared by mixing 200 ml of chicken juice (as above), 600 ml of deionized water and 40 g of PAA crystals providing a 1:3 dilution ratio of chicken juice:water. The biodegradable ALG hydrogels were produced by the method described by Tay et al. (2017) with minor modifications. The Na-Alg solution (1%) was slowly dispensed dropwise through a modified 8-inch shower head nozzle (1.6 mm diameter). The droplets were immediately collected in a plastic container with 0.5% $CaCl_2$ crosslinker solution. The alginate hydrogel beads were crosslinked in the $CaCl_2$ solution for 2 minutes. The resulting hydrogel beads were filtered. The bait matrices were refrigerated and shipped overnight in a Styrofoam cooler on ice packs.

Salsa cups and lids were weighed and ≈ 30 g of PAA and ALG hydrogels were added to the cups. The entire cup (cup + lid + bait) was weighed again. Two salsa cups each of PAA and ALG baits were then deployed in the field at two locations along transect A within UCR wooden bait stations (Fig. 2). Two salsa cups each of PAA and ALG hydrogels were also deployed

within a screened evaporation cage (as above). One salsa cup each of PAA and ALG bait was removed from the bait station and the evaporation cage after 2 hours and 4 hours. These cups were then sealed, returned to the laboratory, and weighed.

After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6). The amount of each matrix removed was analyzed with a two-way Anova.

Efficacy Trial 1

A 0.001% dinotefuran bait was prepared by mixing 0.01 g dinotefuran, 100 ml of chicken juice, 300 ml of water, and 20 g of the PAA crystals (as above). This PAA bait formulation was then placed in the refrigerator and conditioned overnight for at least 16 hours. The baits were then packed in a cooler with an ice pack and shipped overnight to field sites for deployment.

The PAA bait containing 0.001% dinotefuran was deployed for 24 hours along transect A, centered at trap 2A on 9/4/2019 (Fig. 10). Three plastic salsa cups filled with bait (mean mass = 32.7 g, n = 9) were placed in each of three UCR bait stations that were hung about 1.5 m high and about 5 m apart. An evaporation check station, also with three cups of bait and screened to prevent wasp access, was hung alongside the central bait station.

After 24 hours, the bait cups were removed, covered, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets trapped before and after treatment was analyzed with a Wilcoxon signed-ranks test.

Efficacy Trial 2

A 0.0025% dinotefuran bait was prepared by mixing 0.025 g dinotefuran, 100 ml of chicken juice, 300 ml of water, and 20 g of the PAA crystals. The PAA mixture was placed in the refrigerator and conditioned overnight for 16 hours. The baits were then packed in a cooler with an ice pack and shipped overnight to be tested.

On 9/18/2021, PAA baits containing 0.0025% dinotefuran were deployed for 24 hours along transect A, centered at trap -3A. As with Efficacy Trial 1, three plastic salsa cups filled with bait (mean mass = 33.1 g, n = 9) were placed in each of three bait stations and hung alongside one evaporation check station.

After 24 hours, the bait cups were removed, covered, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets trapped before and after treatment was analyzed with a Wilcoxon signed-ranks test.

Results

Monitoring

The only species trapped was *V. pensylvanica*. A total of 25 queens and 15,077 workers were trapped during 2019.

Bait Matrix Preference Trial

The ALG beads lost 4% of their weight at 2 hours and 8% at 8 hours in the evaporation control. The PAA gels lost 2.8% of their weight at 2 hours and 6.2% at 8 hours (Table 1). After adjusting for water lost from each bait, yellowjackets removed significantly more of the PAA gels than the ALG beads ($F= 8.37$; $df = 1,7$; $\mathbf{P} = 0.034$).

Table 1. Choice tests with PAA and ALG hydrogels conditioned in chicken juice.

Bait matrix type	deployment (hours)	Average bait (g) removed (± SD)
polyacrylamide hydrogel (PAA)	2	1.44 ± 0.009
	4	6.85 ± 1.867
seaweed alginate hydrogel (ALG)	2	0.41 ± 0.142
	4	0.78 ± 0.219

Efficacy Trial 1

The bait cups in the evaporative controls lost about 20% of their initial mass during the 24-hour deployment period. Accounting for this evaporation, yellowjackets removed only 30.0 g of 0.001% dinotefuran bait, representing about 11.4% of the bait that was placed out on 9/4/2019.

The 0.001% dinotefuran bait provided significant reductions in the average number of yellowjackets trapped at day 7 ($W = 21$, $n = 7$, $\mathbf{P} = 0.03$) and 14 ($W = 24$, $n = 7$, $\mathbf{P} = 0.04$) after baiting (Table 2). The numbers of yellowjackets trapped at the untreated transects remained steady over the 14-day period.

Table 2. The average number of yellowjackets/trap/day (YJ/T/D) before and after deployment of 0.001% dinotefuran hydrogel bait.

	No. Traps	Average YJ/T/D (% reduction)		
		Pre-baiting	Day 7	Day 14
Transect C (treated with 0.001% dinotefuran)	7	16.69	9.33 (44.1%)	9.94 (40.5%)
Transect A (untreated)	9	15.25	8.70 (43.0%)	13.52 (0.0%)
Transect B (untreated)	7	10.18	8.37 (17.8%)	15.73 (0.0%)

Efficacy Trial 2

The bait cups in the evaporation control lost about 16.9% of their initial weights during the 24-hour deployment period. Accounting for this evaporation, yellowjackets only removed 26.3 g of 0.0025% dinotefuran bait from cups in the bait stations. This amount represented about 9.9% of the total bait deployed.

The 0.0025% dinotefuran bait failed to provide significant reductions and the number of yellowjackets trapped per day per trap was unchanged (Table 3). Along transect B (untreated) the average YJ/T/D significantly declined 28 days after the baiting ($W = 28$, $n = 9$, $\mathbf{P} = 0.016$), perhaps due to decreasing temperatures and or photoperiods.

Table 3. Yellowjacket wasp trap densities before and after deployment of 0.0025% dinotefuran hydrogel bait.

	No. traps	Average YJ/T/D (% reduction) days after baiting				
		Pre-baiting	Day 7	Day 14	Day 21	Day 28
Transect A (treated with 0.0025% dinotefuran)	7	7.73	7.89 (0.0%)	12.79 (0.0%)	5.72 (26.0%)	7.21 (6.8%)
Transect B (untreated)	7	12.24	10.29 (16.0%)	8.27 (32.4%)	11.93 (2.6%)	3.68 (69.9%)

Discussion

Yellowjacket trap density increased slowly during 2019, only surpassing the pest threshold of 10 YJ/T/D after 8/12/2019. The novel formulation of ALG bait was less attractive than the PAA hydrogel. The observations by the team members suggest that additional processing (maceration) of alginate hydrogel beads may be necessary to produce pieces of bait more easily handled by yellowjacket foragers.

The western transect (B) at the Richmond Field Station was left untreated as a seasonal density check for efficacy comparisons, while bait trials took place along the eastern (A) and southern (C) transects. A significant decline in wasp density was recorded between trapping events 10/8/2019 and 10/16/2019 along the untreated transect, probably due to seasonal phenology related to decreasing temperatures and photoperiods.

UC Richmond Field Station, 2020

Methods and Materials

Monitoring

Three transects were utilized at the UC Richmond Field Station during 2020, with 11 weekly trapping periods, beginning 5/12/2020 and ending 10/14/2020 (Fig. 13). The East transect (A) was baited during 8/27/20 and 8/28/2020 at location -1A, and again during 10/01/20 and 10/02/2020, at location -4A. The West transect (B) and the South transect C were both left untreated and considered as seasonal phenology checks (controls). Transect B was approximately 700 m from transect A and approximately 500 m from transect C.

Fig. 13. Map of the UC Berkeley Richmond Field Station showing the locations of the three monitoring transects used during the 2020 trapping season. Baits were applied along transect A while transects B and C were untreated and considered as seasonal checks (controls).

Efficacy Trial 1

Bait was prepared by mixing 420 g of minced chicken, 60 ml of chicken juice, and 0.4 ml fluralaner (Bravecto® 250 mg/ml, Intervet Inc., Madison, WI). The final concentration of fluralaner in the bait was 0.025%. The mixture was refrigerated overnight and shipped overnight on ice packs to northern California (as above).

The 0.025% fluralaner bait was deployed during daylight hours on two consecutive days along transect A, centered at trap -1A. Five plastic cups filled with bait (mean total mass ≈ 28 g, mean mass bait ≈ 25 g) were placed in each of three bait stations that were then hung about 1.5 m high and about 20 m apart. An evaporation check station with four cups of bait was hung alongside the central bait station. Bait stations were set out in the morning (1000 hours on 8/27/2020 and 0900 hours on 8/28/2020) and removed before sundown (1900 hours on both days). Bait was stored in a refrigerator overnight between these two baiting events. After day 2, the bait cups were removed, covered, and weighed. After adjusting for water loss, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

Efficacy Trial 2

A second bait trial, using similar methods as above and centered along transect A at trap -4A, was conducted with 0.025% fluralaner in minced chicken. This trial took place during a late-season heatwave and daytime temperatures were much warmer than is typical for this site (high temperatures 31.7° C and 33.3° C). Despite being at the end of the same transect associated with Efficacy Trial 1, trap -4A was used as the baiting location because wasp counts along transect A remained above the threshold of 10 YJ/T/D and were highest at -4A.

After day 2, the bait cups were removed, covered, and weighed. After adjusting for water loss, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

DNA Extraction and Microsatellite Genotyping

DNA was extracted from the thorax of *V. pensylvanica* workers using the DNeasy Blood and Tissue Kit (Qiagen, Hilden, Germany) following the manufacturer's instructions and stored at –20°C until used. Five workers from each trap in transect A of Richmond Field Station were scored at eight microsatellite loci: RUFA5, RUFA19, LIST2004, LIST2014, LIST2017, LIST2019, LIST2020, VMA6 (Daly et al. 2002; Hasegawa and Takahashi 2002; Thoren et al. 1995). In some traps with sample sizes smaller than five, fewer than five individuals were scored. PCR mixtures contained 1–2 µL of template DNA, 0.2 µM of each primer, 7.5 µL PCR Master Mix (Cat# K0171, Thermo Scientific, MA, USA), and ddH_2O (15 µL reactions volume in total). Forward primers were labeled with 5'-fluorescent tags (6-FAM or HEX; Integrated DNA Technologies, Iowa, USA) for genotyping. PCR conditions consisting of an initial denaturation of 3 min at 95°C, followed by 15 cycles of 30 sec at 95°C, 30 sec at an annealing temperature beginning at 60°C and decreasing 1°C each cycle, 30 sec at 72°C, then 25 cycles of 30 sec at 95°C, 30 sec at 50°C, 30 sec at 72°C, followed by a final 7-min extension at 72°C. The resulting PCR products were analyzed on an ABI-3730 Genetic Analyzer (Applied Biosystems) at the University of Arizona Genomic Analysis and Technology Core Facility (GATC). Microsatellite Analysis Software (available on Thermo Fisher Cloud) was used to visualize and score alleles.

The degree of relatedness among individual workers was estimated using the maximum likelihood sibship reconstruction method in COLONY ver. 2.0.6.6 (Jones and Wang 2010). This allowed us to group workers into colonies and to estimate the minimum number of colonies that had produced the workers present at the study site. The analysis was carried out with the following settings: female polygamous and male monogamous, outbreeding, dioecious haplodiploid organisms, and genotyping error rates ranged between 0–2.5% per locus. Colony analysis was run five times, using a different random number seed each time, to give a maximum likelihood reconstruction of full sibships overall runs.

Results

Monitoring

All the yellowjackets trapped were *V. pensylvanica.* The number of wasps trapped first surpassed the pest threshold of 10 YJ/T/D on 7/8/2020 at one trap (-1A). Baiting was not initiated until mid-August, when three traps in transect A exceeded 10 YJ/T/D. Transects B and C consistently yielded fewer wasps and were left untreated to serve as seasonal checks. A total of 33 queens and 9,266 workers were trapped during 2020.

Efficacy Trial 1

The baits in the evaporation cage lost an average of 12.4% of their initial weight during the 2-day exposure period. After compensating for this water loss, 172.1 g of bait (≈ 45.8 % of the bait applied) were removed by yellowjackets from 19 bait cups. Considering that the evaporation observed was less than in previous years and could not be explained by differences in temperature or humidity, we concluded that collecting the baits and storing them overnight reduced the amount of water lost and helped maintain their attractiveness.

After baiting, nine traps along transect A were monitored weekly to assess yellowjacket foraging populations. When considering only the trap at the baiting site (-1A) and the two traps nearest to the baiting site (0A and -2A), there were 94.3, 83.7, and 93.3% reductions in the average number of YJ/T/D at days 20, 27, and 34 post baiting, respectively (Table 4). When all nine traps were considered, there were 84.9 % (W =29, Z = 1.69, **P** = 0.098), 72.2% (W = 35, Z = 2.04, **P** = 0.041), and 75.3% (W= 35, Z = 2.04, **P** = 0.041) reductions in the average number of YJ/T/D at day 20, 27, and 34 post baiting, respectively. The untreated controls remained unchanged until 34 days after baiting, when there was a significant reduction in the number of yellowjackets trapped along transect B (W = 28, n = 7, **P** = 0.016).

Table 4. Percent reduction in the average number of YJ/T/D at Site A after baiting with 0.025% fluralaner in minced chicken. Baits applied on 8/26/2020 and 8/27/2020.

Traps	Average YJ/T/D (% reduction) days after baiting			
	Pre-baiting	Day 20	Day 27	Day 34
0A, -1A, -2A (Three traps nearest to bait application)	26.9	1.5 (94.3%)	3.4 (83.7%)	1.4 (93.3%)
Transect A (baited with 0.025% fluralaner, all 9 traps considered)	20.2	3.0 (84.9%)	5.6 (72.2%)	5.0 (75.3%)
Transect B (untreated)	6.1	4.9 (19.1%)	11.9 (0.0)	2.3 (62.4%)
Transect C (untreated)	4.4	16.8 (16.8%)	9.2 (0.0)	34.0 (8.9%)

Efficacy Trial 2

The baits in the evaporation cage lost an average of 12.4% of their initial weight during the 2-day exposure period. After compensating for this water loss, we calculated that a total of 83.7 g of bait (≈ 22.3 % of the bait applied) was removed by yellowjackets from 19 bait cups.

The second baiting reduced the trap counts at -2A, -3A, and -4A by 96.3 and 84.2% on days 11 and 20, respectively (Table 5). When considering the entire transect A (nine traps), there were 95.9% (W = 21, n = 7, **P** = 0.031) and 93.9% reductions (W= 34, Z = 2.35, **P** = 0.019) observed at days 11 and 20 after baiting, respectively. Wasp colonies along the untreated control transect B increased by 4 colonies and decreased by 1 colony along transect C 20 days after baiting, when there was a statistically significant reduction observed (W = 28, n = 7, **P** = 0.016).

Table 5. Percent reduction in the average number of YJ/T/D at Site A after a second baiting with 0.025% fluralaner in minced chicken. Baits were applied during 10/01/2020 and 10/02/2020.

Traps	Average YJ/T/D (% reduction) days after baiting		
	Pre-baiting	Day 11	Day 20
-2A, -3A, -4A (Three traps nearest to bait application)	27.4	0.5 (96.3%)	1.3 (84.2%)
Transect A (baited with 0.025% fluralaner, all 9 traps considered)	5.0	0.1 (95.9%)	0.2 (93.9%)

Transect B (untreated)	2.3	0.4 (67.9%)	0.2 (88.4%)
Transect C (untreated)	4.0	0.8 (59.5%)	0.4 (86.7%)

DNA Extraction and Microsatellite Genotyping

Before baiting, large numbers of samples were collected, but because of operational limitations, only 4% were genotyped (Table 6). A minimum of 27 colonies was identified along transect A. This number declined to 19 when examined 20 days after baiting. Of the 27 colonies detected before baiting, 9 of the same colonies were detected after the first baiting, and only 3 colonies were detected after the second baiting (Fig. 14). Sixteen new colonies were detected after the first baiting, and five new colonies were detected after the second baiting.

Ten different colonies were initially detected along transect B (untreated), and this number increased to 14 by the end of the study. Along transect C (untreated), 12 different colonies were detected at beginning, and 11 different colonies were detected at the end of the study.

Table 6. Analysis of yellowjackets at transects A, B, and C with an estimate of the minimum number of different colonies.

Transect		No. samples collected	No. samples genotyped	Proportion of genotyped individuals	Minimum No. colonies detected
A	Pre-baiting	1273	45	0.04	27
	20 d after 1st baiting	630	37	0.06	19
	34 d after 1st baiting	314	33	0.11	15
	11 d after 2nd baiting	13	10	0.77	8
	20 d after 2nd baiting	19	11	0.58	7
B	Pre-Baiting	298	34	0.11	10
	20 days after 1st baiting	792	30	0.04	9
	11 days after 2nd baiting	36	29	0.81	14
C	Pre-Baiting	214	26	0.12	12
	20 days after 1st baiting	585	30	0.05	11
	11 days after 2nd baiting	79	24	0.30	11

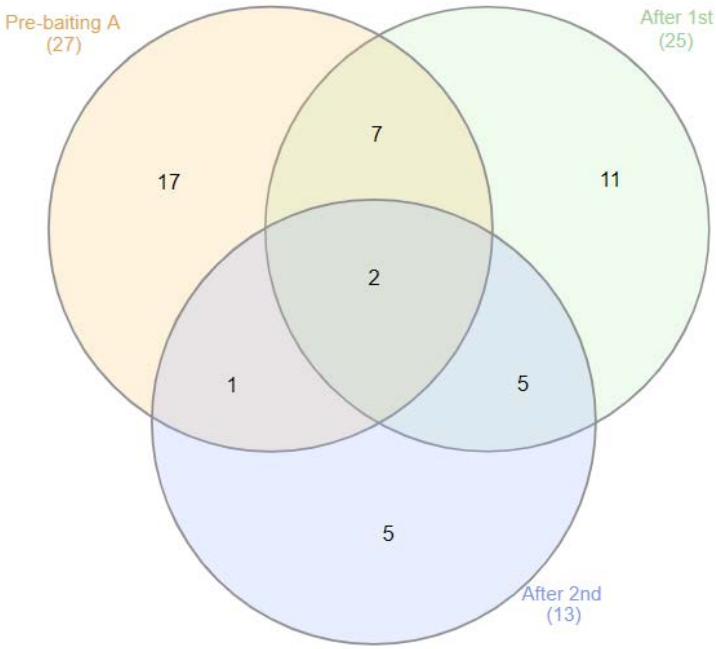

Fig. 14. Venn diagram of the number of colonies at transect A before and after baiting.

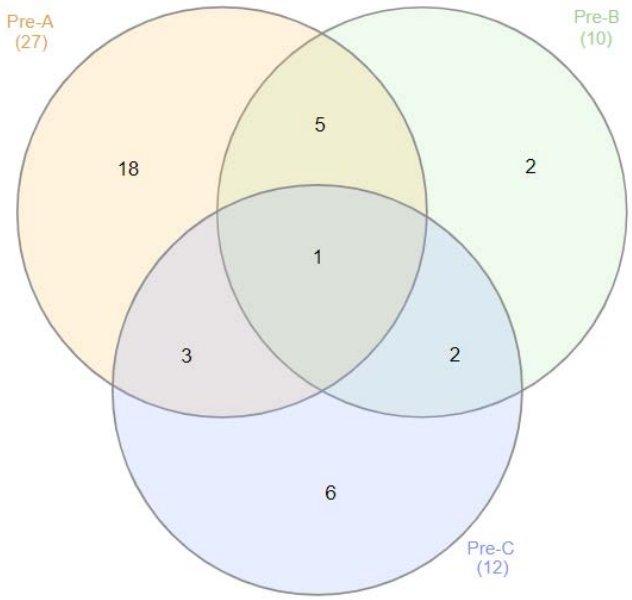

Fig. 15. Venn diagram of the number of colonies at transect A, B, and C before baiting.

The Venn diagram (Fig. 15) shows that individuals from 11 colonies foraged in more than one transect. For example, the foraging areas of 5 colonies contain both transect A and B, 3 colonies contain both transect A and C, and 1 colony foraged in all transects. In total, 31% (14 out of 45) of the individuals collected from transect A before baiting belonged to colonies which workers were also found in the two untreated transects, B and C.

Discussion

The yellowjackets actively foraged on and removed the 0.025% fluralaner in minced chicken. This bait removal resulted in significant reductions in the number of yellowjackets trapped after the first baiting. This decrease was observed during September when the number of foragers increased in the untreated control transects. The second baiting event significantly reduced the numbers of yellowjackets trapped 11 days after baiting. A significant reduction in number of yellowjackets trapped at one of the untreated transects 20 days after the second baiting indicated that the seasonal declines had begun well after the baiting.

Application of the fluralaner bait resulted in a decline in the number of colonies detected after the first baiting as based on the molecular analyses. Some colonies were eliminated and were not detected after the first and second baiting, while other new colonies appeared. The number of colonies detected at the untreated transect B increased by 4 and the at the untreated transect C decreased by 1.

We conclude that the addition of foragers from new colonies may have affected the wasp trapping data and associated analyses of wasp densities. Since new colonies were detected after baiting, the overall percent reduction in wasp density may have been underestimated due to an influx of new foragers, representing these new colonies. Thus, these trapping data might provide a conservative underestimate of the overall reductions in yellowjacket density.

The foraging range of colonies may be greater than 400 m. Some colonies were captured in more than one transect suggesting that the distances between the transects were not far enough to prevent the migration of workers among different transects.

UC Richmond Field Station, 2021

Previous comparison of genetic similarity among samples collected from the three transects indicated that 31% (14 out of 45) of the individuals collected from Transect A belonged to colonies whose workers also visited transects, B and C. This result suggested that the distances among transects were not enough to prevent the migration of workers between different transects in previous years. In 2021, an additional site (transect X) was established to provide greater distance from baited transects and therefore, serve as an additional seasonal check. The site was located about 500 m south of transect A and 800 m southeast of transect C (Fig. 16).

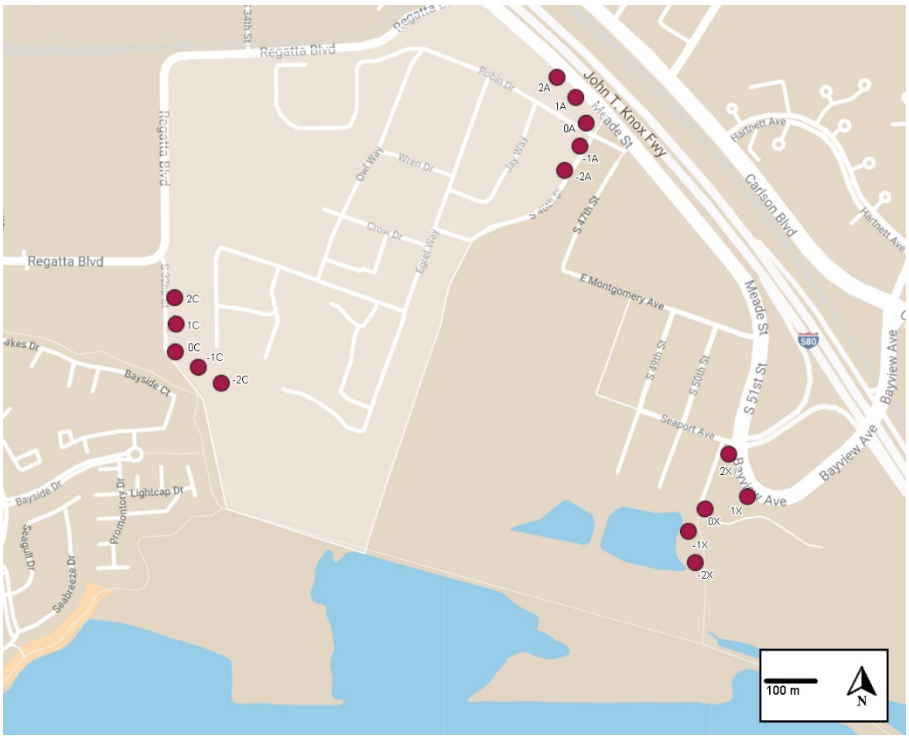

Fig. 16. The three transects monitored during 2021 at the Richmond Field Station.

Methods and Materials

Monitoring

As in previous years, monitoring traps were installed along transects A and C on 6/1/2021. Traps along transect X were installed on 6/8/2021. Transect B was eliminated for the 2021 season (Fig. 13). Monitoring along transects A, C, and X continued until 11/8/2021.

Efficacy Trial #1

A 0.025% fluralaner bait was prepared by mixing 250 ml of chicken juice, 250 ml water, 33.3 g of PAA crystals, and 0.125 g fluralaner (0.5 tube of Bravecto). The mixture was refrigerated and conditioned overnight. The bait was then shipped overnight on ice packs to Richmond.

Salas cups and lids were weighed and ≈ 30 g of conditioned bait was placed in each cup. The entire cup (cup + lid+ bait) was weighed again. Three UCR-style bait stations were then each provisioned with 4 bait cups and hung along transect A on 8/18/2021. An evaporation cage (as above) was also provisioned with 4 bait cups and hung alongside the central bait station to serve as the evaporative control.

After day 2, the bait cups were removed, covered, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets trapped before and after baiting was compared with a Wilcoxon signed-ranks test.

DNA Extraction and Microsatellite Genotyping

Specimens collected along transects A, C, and X were analyzed as described above in 2020.

Results

Monitoring

All the specimens collected were *V. pensylvanica.* A total of 10,532 workers and 2 queens were trapped during 2021.

Efficacy Trial #1

The hydrogels in the evaporative control lost 10.0% of their initial weight during the 48-hour baiting period. After accounting for this water loss, we estimated that the yellowjackets removed a total of 56.6 g of bait (14.3% of the amount deployed).

There was no significant reduction detected in the numbers of yellowjackets trapped after baiting (Table 7). The average numbers of yellowjackets trapped in the two untreated transects varied over the 35 days, but these differences were not significant.

Table 7. The efficacy of 0.025% fluralaner PAA bait against western yellowjackets[a].

Treatment	Average YJ/T/D (% reduction) days after baiting					
	Pre-bait	Day 7	Day 14	Day 21	Day 28	Day 35
0.025% fluralaner	14.40	9.74 (32.3%)	7.83 (45.7%)	12.83 (10.9%)	10.31 (28.4 %)	20.8 (0.0%)
Transect C Untreated	2.40	5.86 (0.0%)	4.08 (0.0%)	13.5 (0.0%)	8.49 (0.0%)	20.80 (0.0%)
Transect X Untreated	0.57	0.23 (60.0%)	0.43 (25.0%)	0.54 (5.0%)	0.51 (10.0%)	0.57 (0.0%)

[a] Baited 8/20/2021 to 8/22/2021.

DNA Extraction and Microsatellite Genotyping

Before baiting, large numbers of samples were collected, but because of operational limitations, only 25 specimens were genotyped from transect A and a minimum of 8 colonies was identified on 8/18/2021. This number increased to 18 when examined 10 days after baiting. Of the 8 colonies detected before baiting, 6 of the same colonies were detected after the first baiting. Colony #1 which represented 48% of the pre-baiting specimens was not detected on 8/30/2021.

Twenty-one specimens were genotyped on transect C (untreated) on 8/18/2020. A minimum of 12 colonies were detected. Three of the same colonies (# 1, 3, and 5) were present in transects A and C pre-baiting, but none of them were present in the 10-day post-baiting sample. Six new colonies were present 10 days after baiting on 8/30/2021.

Discussion

Removal of the 0.025% fluralaner in PAA bait failed to reduce the numbers of yellowjackets trapped over the 35-day post baiting period. Only 14.3% of the bait deployed was removed by the yellowjackets. The wasp trap counts in the nearby untreated transect C remained high during and after the baiting period. From these findings we conclude that the experimental bait formulation comprised of 0.025% fluralaner in PAA was not effective at reducing yellowjacket numbers.

The number of colonies sampled in the traps increased to 18 colonies 10 days after baiting on transect A. Two colonies (# 1 and 6) were no longer detected after baiting. Colony # 1 represented 48% of the specimens genotyped prior to baiting. Even though the overall effect of the baiting with 0.025% fluralaner failed to significantly reduce the YJ/T/D along transect A, colony #1 appeared to be eliminated.

Tahoe-area Bait Trials, 2018
Placer Mosquito and Vector Control District

Seven different sites from the Lake Tahoe region were monitored for yellowjacket activity in September and October 2018. Trapping was initiated on 9/12/2018 and terminated on 10/2/2018. Monitoring traps were set up about every 30 m at each site. Two sites (North Star Village and Serene Lakes) were baited.

Methods and Materials

Attractant Choice Test

To determine which yellowjacket species responded to the heptyl butyrate and the minced chicken, two Placer-style traps baited with minced chicken and 2 traps baited with heptyl butyrate were placed at each of seven sites. Two traps were provisioned with a glass vial containing 5-7 ml of heptyl butyrate and a piece of dental wick. The other traps had a vial with 25 g of minced chicken. The trap contents were returned to the laboratory and the species and the number of yellowjackets was recorded.

The data were analyzed with a Wilcoxon signed-rank test.

Bait Preparation

The liquid contents from cans of chicken were strained through cheesecloth. The juice was diluted with water (1:1) to make a 400 ml suspension to which 40 g of PAA crystals were added. Another PAA gel was prepared by mixing 200 ml of chicken juice and 400 ml of deionized water (1:2) and 40 g of PAA crystals. A 0.1% aqueous solution of dinotefuran was added to 100 g of the PAA crystals to make baits containing 0.0025, 0.001, and 0.00075% active ingredient (AI). The mixture was placed in the refrigerator and the hydrogel gels were allowed to condition for 48 hours.

Salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. The entire cup (cup + lid+ bait) was weighed again. The baits were packed in a Styrofoam cooler with an ice pack and shipped overnight to be tested.

Choice Trial 1 – North Star Village

North Star Village is located about 6.1 km from Lake Tahoe (39°16'29.68" N, 120°07'16.35" W, elev. 1,945 m). The site is located within the Tahoe National Forest and is covered with pine trees and native shrubs. The monitoring sites were along a wooded border of a shopping and recreation area.

Ten monitoring traps were set up about every 30 m at the site. Monitoring began on 9/12/2018 and the traps were collected 9/19/2018. Placer-style traps baited with heptyl butyrate were hung in trees. Yellowjackets were removed from the traps and placed in containers to be identified to species.

On 9/19/2018, three UCR-style bait cages each with three bait cups (≈ 30 g bait per cup), one replicate (cup) each of three concentrations of dinotefuran (0.00075%, 0.001%, and 0.0025%) in a 1:2 chicken juice to water hydrogel bait were hung in trees. To control for water loss from the hydrogel baits, two evaporation control cages covered with fine window screen were also hung from trees. Five cups of the untreated hydrogel were placed in evaporation cages. The baits and the cups from the evaporation control were removed after 24 hours, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

On 9/21/2018, the monitoring traps were returned to their original sites in the field. The traps were collected and returned to the laboratory on 9/28/2018.

The trap catches before and after baiting were analyzed with a Wilcoxon signed-rank test. The amount of bait taken in the choice tests was analyzed with a Chi-square goodness of fit analysis.

Choice Trial 2 – Serene Lakes

Serene Lakes is a private lakeshore park and picnic area located in the Tahoe National Forest approximately 16 km west of Truckee, CA (39°17'56.62" N, 120°22'59.45" W, elev. 2,103 m). The forests are populated by tamarack, cedar, white fir, and lodge pole pines. Ten monitoring traps were set up about every 30 m at the site. Monitoring began on 9/20/2018 and the traps were collected 9/27/2018. Traps were hung in small trees bordering the lake. Placer-style traps baited with either minced chicken or heptyl butyrate were hung in trees.

On 9/27/2018, four UCR-style bait cages with three bait cups (≈ 30 g bait), one replicate (cup) each of three concentrations of dinotefuran (0.00075%, 0.001%, and 0.0025%) hydrogel bait was hung in trees. To determine the amount of water loss from the hydrogel baits, two

evaporation controls were also hung from trees. Five cups of the untreated hydrogel were placed in a bait cage covered with fine screen to prevent yellowjackets from feeding on the control baits. The baits and cups in the evaporation cages were removed after 48 hours. The bait cups were returned to the laboratory and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

On 9/28/2018, the monitoring traps were placed in their original position. The traps were collected on 10/5/2018 and returned to the laboratory.

The trap catches before and after baiting were analyzed with a Wilcoxon signed-rank test. The amount of bait taken in the choice tests was analyzed with a Chi-square goodness of fit analysis.

Results

Attractant Choice Test

Four species, *Vespula acadica, V. alascensis, V. atropilosa,* and *V. pensylvanica,* were collected over three trapping periods. The traps with minced chicken attracted *V. alascensis* (n = 235) and *V. pensylvanica* (n = 219). Heptyl butyrate attracted four species, *V. acadica* (n = 57), *V. alascensis* (n = 14), *V. atropilosa* (n = 30), and *V. pensylvanica* (n = 1885). The minced chicken caught 94.4% of the *V. alascensis* and only 10.4% of the *V. pensylvanica* caught at the seven different sites. The heptyl butyrate caught significantly more *V. pensylvanica* than did the minced chicken (W = 2.49, n = 6, **P** = 0.008).

Choice Trial 1 – North Star Village

The evaporation check baits lost 17.3% of their weight in 24 hours. When adjusted for the water loss, the yellowjackets removed 51.8 g of dinotefuran bait (Table 9). There was no significant difference in the amount of each concentration of dinotefuran bait removed by the yellowjackets (χ^2 = 0.018, df = 2, **P** > 0.05).

Prior to the choice tests there were 14.03 YJ/T/D. Seven days after baiting, there was a 68.9% reduction in the number of *V. pensylvanica* trapped, but it was not significantly different from the pre-baiting count (W = 37, n = 9, **P** = 0.065).

Choice Trial 2 – Serene Lakes

The evaporation checks lost 22.9% of their weight in 24 hours. After adjusting for the water loss, the yellowjackets removed 81.8 g of dinotefuran bait (Table 8). There was no significant difference in the amount of each concentration of dinotefuran bait taken (χ^2 = 0.18, df = 2, **P** > 0.05)

Table 8. Choice baiting study with 0.0025, 0.001, 0.00075% dinotefuran in PAA hydrogels at two sites near Lake Tahoe.

Site	traps	Mean (± SD) Bait removed (g)			Total Bait removed (g)
		0.0025%	0.001%	0.00075%	
North Star Village	3	5.7 ± 1.17	5.7 ± 0.85	5.9 ± 1.00	51.8
Serene Lakes	4	8.2 ± 1.37	6.7 ± 2.48	6.1 ± 1.56	81.8

Prior to baiting, there were 15.32 YJ/T/D (Table 9). Seven days after baiting, there was an 83.5% reduction in the number of *V. pensylvanica* trapped, but the difference was not significant (W = 37, n = 9, **P** = 0.065).

Table 9. The reduction in the number of yellowjackets after the choice tests with dinotefuran.

Site	traps	Average YJ/Trap/Day (% reduction)	
		Pre-baiting	7 days
North Star Village	10	14.03	4.36 (68.9%) n.s.
Serene Lakes	10	15.32	2.53 (83.50%) n.s.

Tahoe-area Bait Trials, 2019
Placer Mosquito and Vector Control District

Seven different sites from the Lake Tahoe region were monitored for yellowjacket activity in 2019 using a pair of Placer-style traps at each site, one with a heptyl butyrate attractant and one with minced chicken attractant. Two of these sites were selected for bait efficacy trials: Alpine Meadows Water District and North Star Village.

Methods and Materials

Bait Preparation

The contents from Swanson canned chicken were strained through cheesecloth. The juice (100 ml) was diluted with water (300 ml) and combined with 0.01 g dinotefuran. Twenty grams of the PAA were added to the mixture resulting in hydrogels containing 0.001% dinotefuran. The PAA mixture was placed in the refrigerator and conditioned overnight (minimum of 16 hours). The baits were packed in a cooler with an ice pack and shipped overnight to be tested.

Efficacy Trial 1 – North Star Village

Thirty monitoring traps were set up about every 25 m at the site (Fig. 17). Monitoring began on 8/10/2019 and the trap contents were collected weekly.

On 8/29/2019, three bait stations each containing three bait cups of ≈ 30 g of 0.001% dinotefuran bait were hung in trees. To control for water loss from the baits, one evaporation control bait station with three bait cups was hung. The bait and evaporation stations were removed after 24 hours. At the end of the baiting period, the cups were sealed, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

On 8/30/2019, monitoring traps were returned to their original sites in the field. Yellowjackets were collected and traps were collected at 7, 14, 21, and 28 days after baiting.

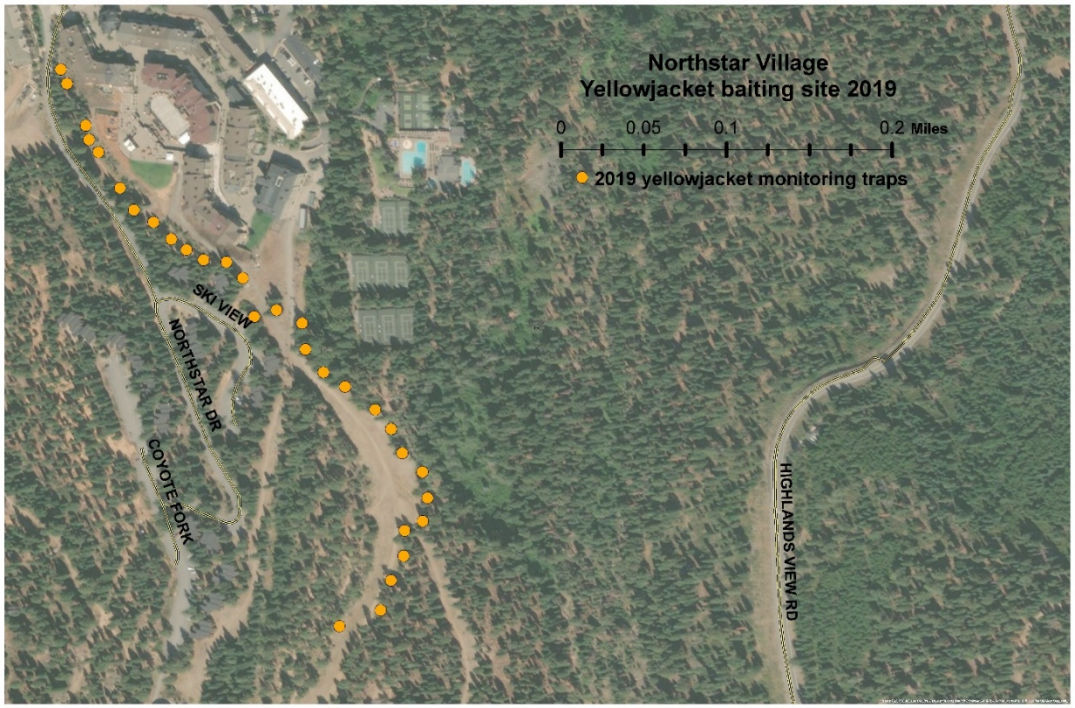

Fig. 17. North Star Village with monitoring traps indicated with yellow dots.

Efficacy Trial 2 – Alpine Meadows

The Alpine Meadows site (39°11'21.67" N, 120°11'55.62" W, elev. 1,975 m) is located about 5.6 km northwest of Lake Tahoe in a partly sloped and rocky area heavily forested with pine, fir, and incense cedar trees. The site is adjacent to the Alpine Meadows Water District offices and garbage collection area and is bordered to the south by a small, landscaped park and to the north by Bear Creek.

Thirty monitoring traps were set up about every 25 m at the site (Fig. 18). Monitoring began on 8/14/2019 and the traps were collected 8/21 and 8/28 (14 and 7 days prior to baiting). Placer-style traps baited with heptyl butyrate were hung in trees to prevent bears and other animals from disturbing them. Yellowjackets were removed from the traps and preserved in ethanol for later identification and counting.

On 9/10/2019, three bait stations each containing three cups of ≈ 30 g bait were hung in trees. To control for water loss from the baits, one evaporation control cage, also containing three bait cups, was hung. The bait stations were removed after 24 hours, and the bait cups were returned to the laboratory and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

On 9/11/2019, monitoring traps were placed in their original positions. Yellowjackets were collected and traps were collected at 7, 14, 21, and 28 days after baiting.

Fig. 18. The Alpine Meadows site with monitoring station indicated with yellow dots.

Results

Monitoring

Collections at North Star Village were predominantly *Vespula pensylvanica* (95%), but also included *V. alascensis, V. acadica, V. atropilosa, Dolichovespula maculata, and D. arenaria*. Yellowjackets captured at Alpine Meadows were predominantly *V. pensylvanica* (83%), followed by *V. acadica* (9%), *V. alascensis* (6%), *V. atropilosa, Dolichovespula maculata*, and *D. arenaria* (all < 2%).

The trap counts at all sites declined dramatically on 10/14/2019.

Baiting Trial 1 – North Star Village

The bait cups in the evaporation controls lost an average 9.2 g (10.1%) in 24 hours. A total of 130.1 g was removed by yellowjackets in 24 hours from 9 bait cups (Table 10). The numbers of yellowjackets trapped increased for the first 14 days after baiting. The decline in the number of yellowjackets trapped at 21 days preceded the declines at the other sites on 10/14/2019.

Baiting Trial 2 – Alpine Meadows

The bait cups in the evaporation controls lost an average of 9.2% of their weight in 24 hours. When adjusted for water loss, the total amount of bait removed was 15.8 g (Table 11).

The number of yellowjackets trapped increased 7 and 14 days after baiting and then sharply declined. However, the large decreases were also observed at this time at other unbaited sites as well (Fig. 19). The declines were likely due to drops in temperature. A number of the collection jars were frozen on October 2 (day 28).

Table 10. The amount of dinotefuran bait removed and the average number of yellowjackets/trap/day (YJ/T/D) before and after baiting.

Site Bait	Bait Taken (g)	Average YJ/T/D (% reductions)				
		Pre-baiting	Day 7	Day 14	Day 21	Day 28
North Star Village						
0.001% dinotefuran	130.1	47.28	70.87 (0.0%)	51.22 (0.0%)	27.03 (42.2%)	35.60 (24.7%)
Alpine Meadows						
0.0025% dinotefuran	15.82	18.06	25.31	22.24	0.91	Disc.

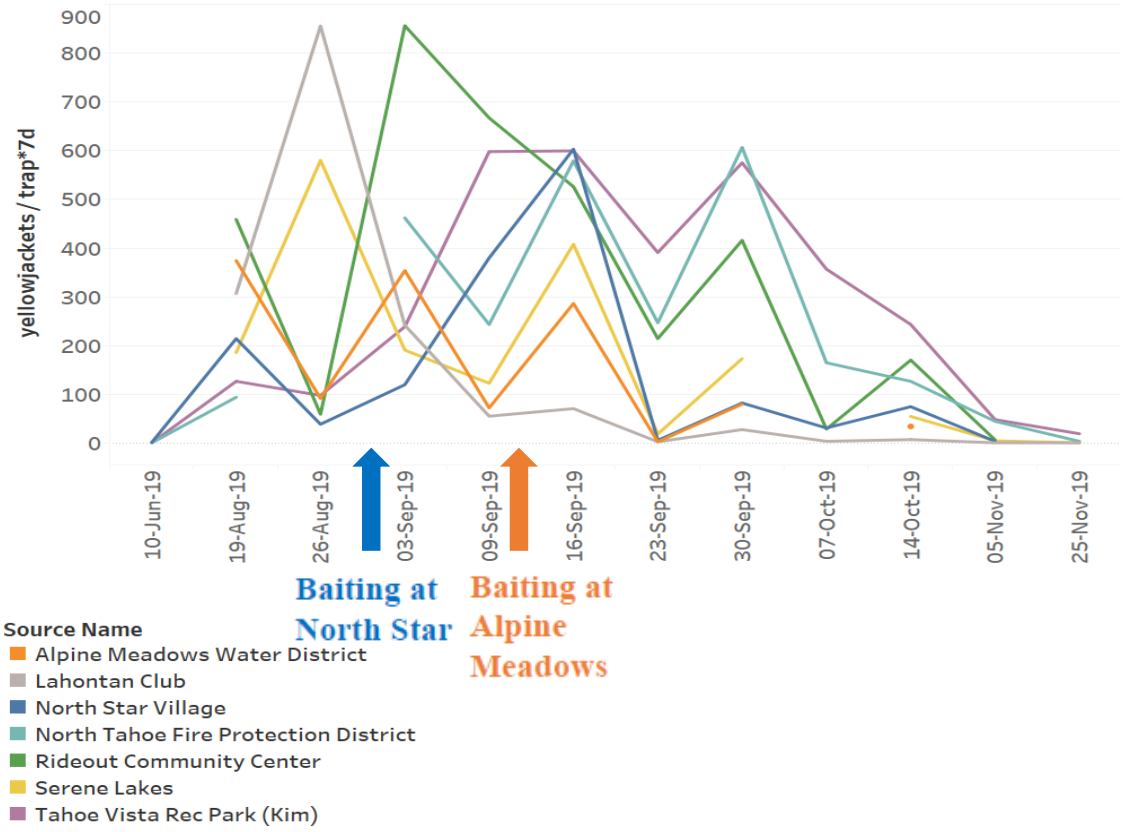

Fig. 19. Weekly monitoring data for all seven sites at Lake Tahoe (including the two baited sites). Only the yellowjackets from traps baited with heptyl butyrate are shown.

Tahoe-area Bait Trials, 2020
Placer Mosquito and Vector Control District

*S*even different sites from the Lake Tahoe region were monitored for yellowjacket activity in 2020 using a pair of Placer-style traps at each site, one with a heptyl butyrate attractant and one with chicken attractant. Two of these sites were selected for bait efficacy trials: Alpine Meadows Water District and North Tahoe Regional Park. Two additional sites, North Star Village and Tahoe City were used for bait preference trials.

Methods and Materials

Efficacy Trial 1 – Alpine Meadows

The Alpine Meadows site is located about 5.6 km northwest of Lake Tahoe in a partly sloped and rocky, naturally forested area. The site is adjacent to the Alpine Meadows Water District offices and garbage collection area. The site is bordered to the south by a small, landscaped park and to the north by Bear Creek (Fig. 18).

Bait was pre-mixed at UC Riverside using the selamectin + sarolaner (Revolution® Plus, Zoetis, Inc., Kalamazoo, MI). Four packets of Revolution Plus (240 mg selamectin + 40 mg sarolaner) were mixed with 80 ml of chicken juice and 420 g of minced canned chicken to make a 0.048% selamectin + 0.008% sarolaner bait. The bait was shipped via overnight mail in an insulated cooler.

Salsa cups and lids were weighed and ≈ 25 g bait was put into each salsa cup. The entire cup (cups + lid + bait) was weighed again. On 8/26/2020, four bait cages each containing four

bait cups of selamectin + sarolaner bait were hung in trees. One evaporation control bait station with three bait cups was also placed to control for water loss from the baits. The bait and evaporation cages were removed after 48 hours. The bait cups were placed in a cooler, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

On 8/28/2020, monitoring traps were returned to their original sites in the field. Yellowjackets were collected at 7 and 14 days after baiting. The trap catches before and after baiting were analyzed with a Wilcoxon signed-rank test.

Efficacy Trial 2 – North Tahoe Regional Park

The North Tahoe Regional Park site (39°14'59.69" N, 120°3'11.016" W, elev. 1,969 m) is in Tahoe Vista, about 1.2 km north of Lake Tahoe in a partly sloped and rocky, naturally forested area, heavily treed with pine, fir, and incense cedar. The 50.2 ha park includes picnic areas, sports fields, and other developed recreation areas including trails and natural areas.

Monitoring began on 8/18/2020 and the traps were collected seven days before baiting. Placer-style traps baited with heptyl butyrate were hung in trees to prevent bears and other animals from disturbing them. Yellowjackets were removed from the traps and preserved in alcohol for later identification to species.

Bait was pre-mixed at UC Riverside using technical fluralaner (98%, BOSCO Sciences, Inc., Shirley, NY) dissolved in 1 ml DMSO and 1 ml of water. The solution was added to 80 ml of chicken juice and 420 g of minced canned chicken to make a 0.05% bait. The bait was shipped via overnight on ice packs.

Salsa cups and lids were weighed and ≈ 25 g of bait was added to the cups. The entire cup (cup + lid + bait) was weighed again. Three cups of bait were put into three Placer-style bait stations and hung in trees to prevent bears and other animals from disturbing them. To control for water loss from the baits, one evaporation control cage containing four bait cups was also hung. The bait cups were removed after 48 hours, sealed, placed into coolers, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The trap catches before and after baiting were analyzed with a Wilcoxon signed-rank test.

Choice Preference Test – North Star Village

North Star Village is located about 6.1 km from Lake Tahoe (39°16'29.68" N, 120°07'16.35" W, elev. 1,945 m). The site is located within the Tahoe National Forest and is covered with pine trees and native shrubs. The trapping site is along a wooded border of a shopping and recreation area.

A choice test was prepared for different concentrations of clothianidin bait in either minced chicken or PAA crystals. Clothianidin cockroach bait (Maxforce Impact, 1% clothianidin, Bayer Environmental Sciences, Cary, NC) was mixed with 80 ml of chicken juice and 420 g of minced canned chicken so that the finish concentrations of bait were 0.05, 0.025, and 0.0125%.

Plastic salsa cups and lids were weighed and filled with ~25 g of bait each. The entire cup (cup + bait + lid) was weighed again. Cups were deployed on 9/23/2020 in bait cages as described above, with three cups of each matrix (PAA or minced chicken) placed in a screened cage to serve as controls for evaporation. Bait cups were collected and weighed after 24 hours.

After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The trap catches before and after baiting were analyzed with a Wilcoxon signed-rank test. The amount of bait taken in the choice tests was analyzed with a Chi-square goodness of fit analysis.

Choice Preference Test – Tahoe City

The Tahoe City site (39° 10' 3.954" N, -120° 8' 49.473" W, elevation 1,967.8 m) is a small greenbelt area 0.5 km west of Lake Tahoe containing pines, incense cedar and firs and bordered by a fire station, a golf course, a grocery store, and a retention basin (dry in summer). There is also a nearby drainage area creek that was still somewhat wet in summer 2020. Clothianidin baits were prepared for the North Star Village choice test above and deployed for 24 hours starting 9/30/2020. Bait cups were collected and weighed after 24 hours. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The amount of bait taken in the choice tests was analyzed with a Chi-square goodness of fit analysis.

Results

Efficacy Trial 1 – Alpine Meadows

The monitoring collections were predominantly *V. pensylvanica* (62.0%), but other species were collected in the heptyl butyrate traps including *V. acadica* (26.8%), *V. alascensis* (5.8%), *V. atropilosa* (5.4%), and *Dolichovespula maculata* (0.03%). Enough *V. alascensis* were trapped to analyze the data.

The bait cups in the evaporation checks lost an average of 28.8% of their weight during the 48-hour exposure. After correcting for evaporation, the yellowjackets removed 198 g of bait (about 48.4% of the bait) from 16 cups. The selamectin + sarolaner bait provided a significant 77.3 and 60.6% reductions in trap counts of *V. alascensis* after day 7 (W = 276, Z = 3.5, **P** = 0.0005) and 14 (W = 249, Z = 3.55, **P =** 0.0004). respectively (Table 11). The bait provided a significant 56.8 and 42.1% reduction of *V. pensylvanica* at days 7 (W = 258, Z = 2.78, **P** = 0.005) and 14 (W = 296, Z = 3.04, **P** = 0.002), respectively.

Table 11. The average number of yellowjackets/trap/day (YJ/T/D) and the percent reductions after baiting with 0.048% selamectin + 0.008% sarolaner in minced chicken.

	Average YJ/T/D (% reduction)		
Species	Pre-baiting	Day 7	Day 14
V. alascensis	0.75	0.22 (71.0%)	0.38 (49.5%)
V. pensylvanica	8.5	3.67 (56.8%)	4.86 (42.1%)

Efficacy Trial 2 – North Tahoe Regional Park

The predominant species collected in the traps was *V. pensylvanica* (89.8%) followed by *V. atropilosa* (5.1%), *V. alascensis* (2.9%), *V. acadica* (2.0%), and *Dolichovespula maculata* (0.07%). Enough *V. alascensis* were trapped to analyze the data.

The control baits lost 56.1% of their weight in the evaporation controls during the 48-hour exposure. After compensating for the water loss, yellowjackets removed 134.3 g of 0.05% fluralaner minced chicken bait from 9 bait cups.

The 0.05% fluralaner bait significantly reduced the number of *V. alascensis* trapped at days 7 (W = 371, Z = 4.45, **P** < 0.001) and 14 (W= 423, Z = 4.57, **P** < 0.001; Table 12). There were significant reductions in the number of yellowjackets trapped for *V. pensylvanica* at days 7 (W = 457, Z = 4.69, **P** < 0.001) and 14 (W = 266, Z = 2.73, **P** = 0.005).

Table 12. The average number of yellowjackets/trap/day (YJ/T/D) and the percent reductions after baiting with 0.05% fluralaner in minced chicken.

Species	Average YJ/T/D (% reduction)		
	Pre-baiting	Day 7	Day 14
V. alascensis	0.78	0.13 (83.4%)	0.14 (81.6%)
V. pensylvanica	17.14	2.39 (82.6%)	9.87 (42.4%)

Choice Preference Test – North Star Village

Choice tests with three concentrations of clothianidin in minced chicken and PAA crystals were conducted from 9/16/2020 to 9/17/2020 (Table 13). The minced chicken bait in the evaporation control lost 21.9% of its weight in 24 hours. After compensating for the weight loss, the yellowjackets removed 33.6 g of clothianidin in minced chicken and untreated chicken in 24 hours. The yellowjackets did not prefer any concentration or untreated chicken ($\chi^2 = 2.95$, df = 3, **P** > 0.05).

Table 13. Choice acceptance tests with 0.0125, 0.025, and 0.05% clothianidin in minced chicken and PAA crystals tested from 9/16/2020 to 9/17/2020.

	Average bait taken g (±SD)			
Bait Type	0.0%	0.0125%	0.025%	0.05%
Minced Chicken	25.5 (0.057)	26.3 (0.014)	23.1 (0.023)	25.1 (0.031)
PAA	23.6 (0.022)	29.1 (0.062)	21.3 (0.007)	22.5 (0.023)

The choice test with three concentrations of clothianidin in PAA gels was conducted 9/16/2020 (Table 14). The PAA crystals in the evaporation control lost 25.1% of their weight in 24 hours. After adjusting for the water loss, the amount of all bait removed was 32.1 g. The yellowjackets did not prefer any concentration or untreated chicken ($\chi^2 = 3.51$, df = 3, **P** > 0.05).

Choice Preference Test – Tahoe City

Choice tests with three concentrations of sodium selenate and sodium selenite were conducted from 9/23/2020 to 9/24/2020 (Table 14). The sodium selenate and sodium selenite lost 29.5 and 22.2% of their weight in the evaporative controls during the 24-hour exposure, respectively. After adjusting for the water loss, yellowjackets removed 304.0 g of sodium selenate and the untreated crystals and 303.6 g of sodium selenite and untreated crystals in 24 hours.

There were no significant differences in the amounts of sodium selenate bait taken between the concentrations ($\chi^2 = 0.003$, n = 3, **P** > 0.05). Similarly, there were no significant

differences in the amount of sodium selenite bait removed between the concentrations (χ^2 = 0.003, n = 3, **P** > 0.05).

Table 14. Choice preference tests with 0, 0.0125, 0.025, and 0.05% sodium selenate and sodium selenite in PAA crystals.

	Average bait taken g (±SD)			
Bait Type	0.0%	0.0125%	0.025%	0.05%
Na Selenate	25.5 ± 0.27	25.4 ± 0.28	25.3 ± 0.33	25.2 ± 0.14
Na Selenite	25.5 ± 0.06	25.2 ± 0.10	25.2 ± 0.11	25.3 ± 0.18

Discussion

The 0.05% fluralaner baits initially provided > 80% reductions in the number of yellowjackets trapped. Trap numbers increased by the end of day 14. The 0.048% selamectin + 0.008% sarolaner in minced chicken provided significant reductions in *V. alascensis* and *V. pensylvanica* over 14 days post-baiting. The 0.05% fluralaner (technical AI) also provided significant reductions of both species. When baited, the percent reductions of *V. alascensis* were consistently higher than *V. pensylvanica*.

The yellowjackets retrieved similar amounts of all three concentrations of clothianidin and the untreated control in the choice tests. Similarly, all concentrations of sodium selenate and sodium selenite were taken by yellowjackets. Higher concentrations of each of these potential toxicants should be tested.

Tahoe-area Bait Trials, 2021
Placer Mosquito and Vector Control District

Monitoring began at the Lake Tahoe sites on 7/21/2021. The wildfires began on 8/14/2021 and burned 896 km^2. The baiting and monitoring were discontinued because of the excessive smoke and fire hazard. The fire was not contained for nearly two months.

The North Lake Tahoe site is located within the North Tahoe Regional Park (39°15'02.42" N, 120°03'13.58" W, elev. 1,977 m). The Park consists of 50.2 ha mostly forested with pines and fir trees. It is a multi-use facility with outdoor activities including hiking, trail biking, picnicking, and baseball and soccer fields.

Methods and Materials

Efficacy Trial 1 – North Lake Tahoe

The bait was prepared by mixing 8 tubes of Revolution Plus (480 mg selamectin and 80 mg of sarolaner), 250 ml of chicken juice, 250 ml of water and 33.33 g PAA crystals. The final bait consisted of 0.096% selamectin + 0.016% sarolaner. The gels were conditioned for 48 hours in a refrigerator and then shipped overnight to Placer Mosquito and Vector Control District along with pre-weighed plastic salsa cups and lids.

About 22-25 g of bait was added to each pre-weighed cup. The entire cup (cup + lid + bait) was weighed again. Four bait stations with 4 cups of bait (377.3 g) and an evaporation check station with 4 cups were place hung in trees on 8/11/2021. After 24 hours, the bait stations

and cups were retrieved. The bait cups were covered, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets per trap before and after baiting was analyzed with a Wilcoxon signed-rank test.

Results

Efficacy Trial 1 – North Lake Tahoe

The 30 monitoring stations trapped 13.4 YJ/T/D prior to baiting. The evaporative checks lost 49.3% of their weight. After compensating for water loss, yellowjackets removed 333.8 g of the selamectin + sarolaner bait (88.4% of the total).

At day 7, there was a significant 50.3% reduction in the number of YJ/T/D (W = 286, n = 30, **P** = 0.001). Similarly, at day 14 there was a significant 35% reduction (W = 217, n = 30, **P** = 0.014). At day 21, the reduction in the number of yellowjackets trapped (26.5% reduction) was no longer significant.

Discussion

The 0.096% selamectin + 0.016% sarolaner bait was readily accepted suggesting that higher concentrations of active ingredients may be feasible. The reductions in the number of yellowjackets trapped was initially statistically significant, but still not enough to provide area-wide control.

The wildfires disrupted the monitoring and baiting in late August.

Irvine Regional Park 2018

Irvine Regional Park (IRP, 33°47'46.82" N, 117°45'19.82" W, elev. 180 m) is a multiple-use park (≈ 64.7 ha) surrounded by undeveloped wilderness areas composed primarily of a riparian, coastal sage scrub, and oak woodland plant community. The Park offers many activities, including picnics, concession stands, horse stables, shady turf areas, a zoo, and a small lake (Fig. 20). The Park is nestled in the foothills and provides an excellent foraging setting for *V. pensylvanica*.

In 2017, an extensive wildfire burned much of the surrounding native habitat, especially along the northern boundary (Sites 34- 44), but the park remained open.

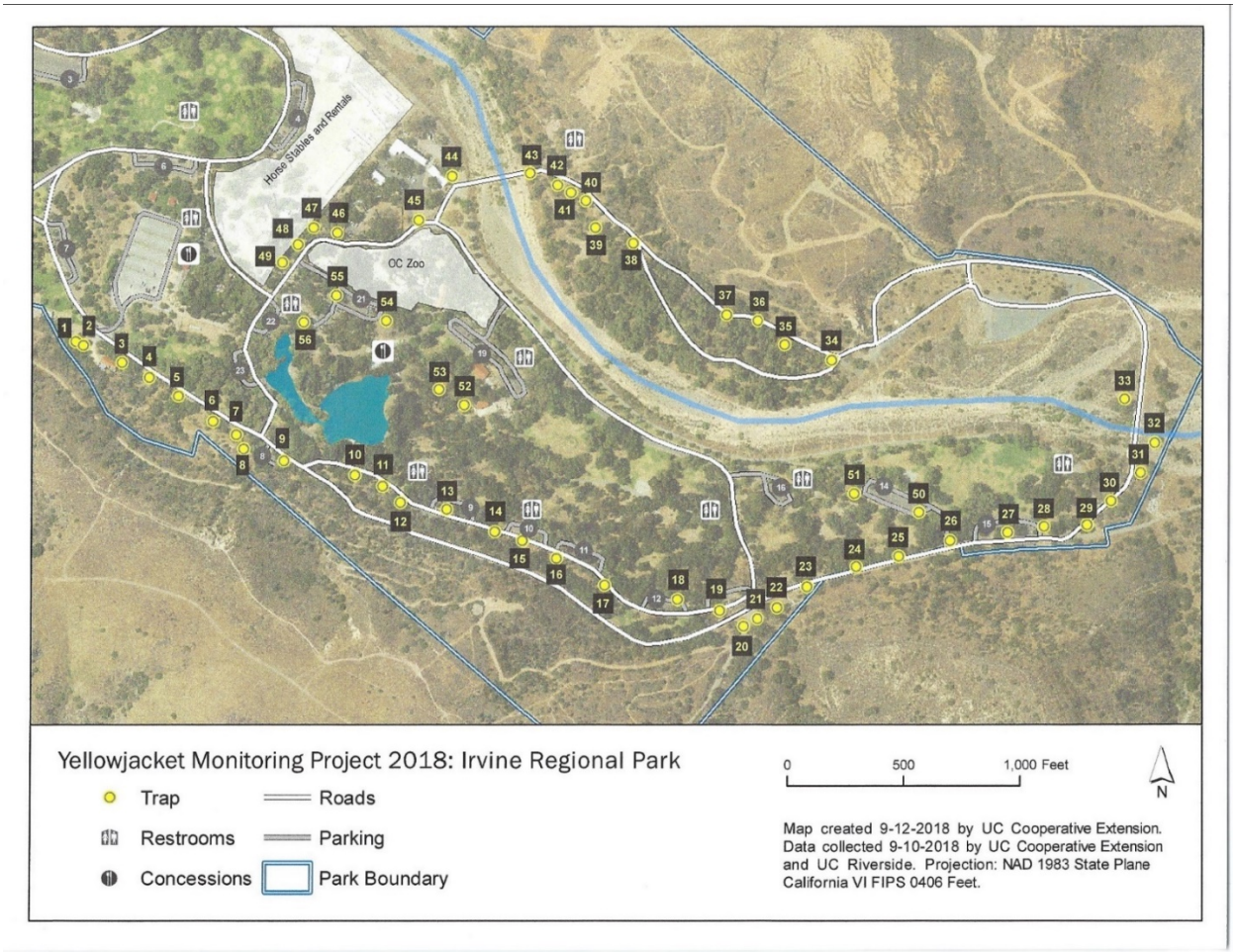

Fig. 20. Map of Irvine Regional Park and the yellowjacket monitoring sites in 2018.

Methods and Materials

Monitoring

The foraging activity of yellowjackets was measured using 56 UCR-style traps. Monitoring began on 8/27/2018 and ended on 10/29/2018. The collection jars were changed every 14 days. The heptyl butyrate vials were replaced as needed.

Results

Only workers of *V. pensylvanica* were trapped from 8/27/2018 until 10/29/2018. By 10/29/2018, nearly 100% of the 56 traps had caught at least one yellowjacket. However, only 1,242 yellowjackets were caught throughout September and October (Fig. 21). The highest trap catch was 0.67 yellowjackets/trap/day (YJ/T/D).

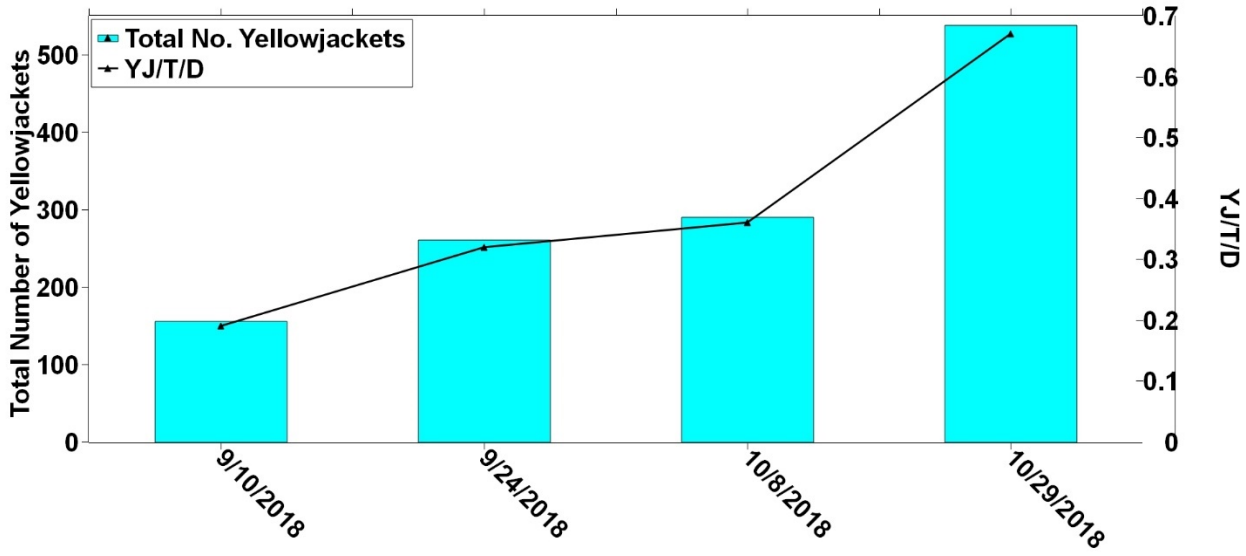

Fig. 21. Yellowjacket trapping data from Irvine Regional Park in 2018. Bars represent the total number of yellowjackets trapped. The straight line represents the average number of yellowjackets/trap/day (YJ/T/D) for all 56 traps.

Discussion

The average number of yellowjackets trapped remained below the 10 yellowjackets/trap/day (YJ/T/D) threshold to initiate a baiting trial. The wildfires in 2017 probably had a negative impact on the yellowjacket populations in 2018. Only a few yellowjackets were collected at monitoring trap locations 33 to 44, adjacent to the burn area. This area had historically high numbers of yellowjackets in the past (Rust et al. 2010).

Irvine Regional Park 2019

Methods and Materials

Monitoring

The monitoring began on 6/12/2019 and ended on 10/28/2019. UCR traps were hung under trees about 1 to 1.5 m off the ground and about 20–80 m apart. The park's perimeter was surrounded by 56 traps in the same configuration used in 2018 (Fig. 20).

Preparation of Food Baits

The Swanson White Premium Chunk Chicken and canned cat food (Friskies Flaked Ocean Whitefish Dinner, Nestle Purina Pet Care Co., St. Louis, MO) were selected because they were attractive and taken by foraging yellowjackets (Rust et al. 2010). To extract the juices from the canned meat and pet food, the contents of each can were poured into a large funnel lined with cheesecloth (Fig. 22). The liquid passed through the cheesecloth into the glass container. The cheesecloth containing the chicken or fish was squeezed over a bowl to collect the remaining juices. A 133-ml can of Swanson chicken (4.5 oz.) provided approximately 70-80 ml of fluid (from now on referred to as “chicken juice”). A can of whitefish dinner pet food (5.5 oz.)

provided approximately 47 ml of a very viscous liquid (from now on referred to as “fish juice”). Chicken broth was obtained by slowly cooking a whole chicken in water for 6 hours. The meat, bones, and skin were removed, and the liquid contents were poured through a strainer. The filtered liquid was refrigerated, and the fat hardened and solidified over the broth. The fat was removed, and only the liquid broth portion was used for preparing the PAA gels.

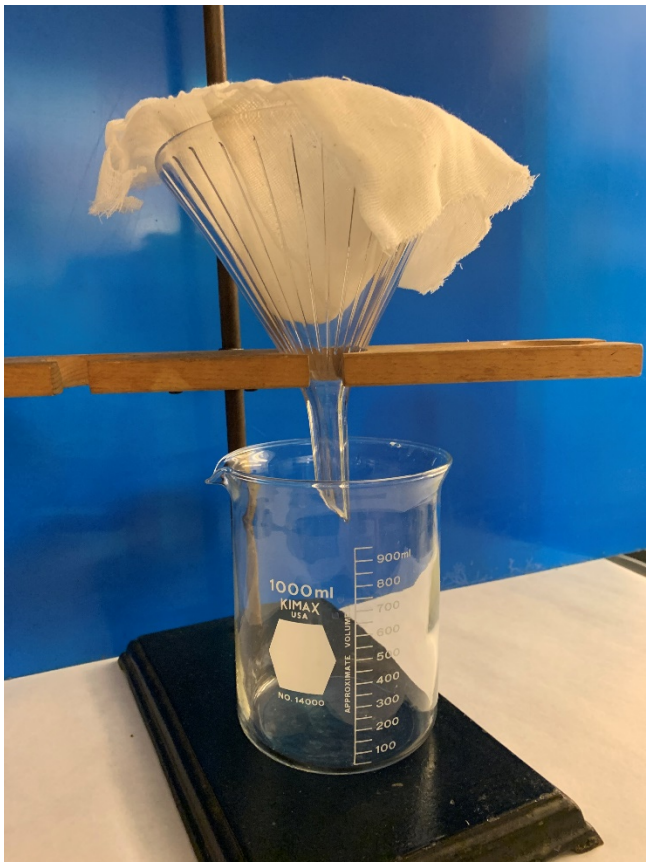

Fig. 22. Extract juices from canned minced chicken and whitefish pet food through cheesecloth.

The hydrogels were prepared with 20 g of PAA crystals for every 400 ml of liquid. The liquid consisted of either pure chicken broth, chicken juice, or fish juice diluted with water in a 1:1 or 1:3 ratio. The gels were prepared as follows:

> Step 1. Three types of baits (chicken juice, fish juice, and chicken broth) were prepared by extracting the juice from the canned meats or a cooked whole chicken into three separate 1-L glass containers.
>
> Step 2. All the juices and broth were collected, measured in a graduated cylinder, and poured into a 1-L glass beaker. Water was added to make 400 ml of conditioning liquid with the appropriate ratios (100% juice

or broth, 1:1, and 1:3 ratio). The conditioning liquid was stirred for approximately 2-3 minutes.

Step 3. The PAA crystals gels (20 g) were added to each mixture and liquid. The mixtures and the pure juice/broth were stirred using the stirring rod for an additional 2-3 minutes.

Step 4. The PAA gels were then conditioned by storing them inside the refrigerator for at least 16 hours (overnight).

Step 5. After the gels were conditioned, they were mixed with a stirring rod before being transferred to bait cups, weighed, and placed back into the refrigerator.

Preparation of hexane/aqueous fractions of chicken and fish juice

About 150 ml of either canned chicken or fish-based pet food juices were collected in a 250 ml Erlenmeyer flask with a glass stopper. About 100 ml of hexane was added to the flask. With the stopper securely closed, the flask was shaken vigorously. After shaking, the flask was left in a fume hood overnight until the two layers (bottom: aqueous fraction; top: hexane fraction) separated and settled. Each fraction was collected using a 5-ml glass pipette and transferred to a clean glass flask.

In some cases (especially for the fish juice), further fractionation and cleanup by centrifugation were necessary. The hexane and aqueous fractions were kept in the refrigerator, loosely covered with aluminum foil overnight. This process helped in further removing any additional hexane from the aqueous fractions.

For “water extract” treatment, 50 ml of the water fraction was used to hydrate 2.5 g of PAA. For “hexane extract” treatment, 50 ml of the hexane fraction were first placed in a glass jar (8 oz.), and hexane was evaporated under a gentle flow of N_2, leaving an oily residue at the bottom. PAA hydrogels fully hydrated in 0.9% NaCl isotonic solution (B. Braum Medical Inc. Irvine, CA; 2.5 g of PAA in 50 ml of 0.9% NaCl solution) were subsequently added to the glass jar and mixed with the oil residue using a spatula. For “both extracts together” treatment, PAA hydrogels hydrated with 50 ml of the aqueous fraction were subsequently treated with the oily residue from 50 ml of the hexane fraction by following the processes described above.

Choice Tests Polyacrylamide (PAA) Gels vs. Sodium Alginate (ALG) Gels

Choice tests were conducted with the hydrogels conditioned in chicken juice to determine if yellowjackets preferred either PAA or ALG hydrogels.

The PAA gels were prepared by mixing 200 ml of chicken juice, 600 ml of deionized water, and 40 g of PAA crystals, providing a 1:3 dilution ratio of chicken juice: water. The gels were allowed to condition in the refrigerator overnight. Excess liquid was drained from gels through a strainer resulting in about 840 g of conditioned gels.

The biodegradable ALG hydrogels were produced by the method described by Tay et al. (2017) with minor modifications. The Na-Alg solution (1%) was slowly dispensed dropwise through a modified 8-inch shower head nozzles (1.6 mm diam.). The droplets were immediately collected with 0.5% $CaCl_2$ crosslinker solution in a plastic container. The alginate hydrogel beads were crosslinked in the $CaCl_2$ solution for 2 minutes. The resulting hydrogel beads were passed

through a sieve. The resulting ALG hydrogels mainly were water, so to obtain an approximate 1:3 (chicken juice: water) ratio, 300 ml of chicken juice, 300 ml of deionized water, and 600 ml of the ALG gel were mixed. The ALG gels were placed in the refrigerator overnight to condition. After 24 hours, the excess liquid was drained from the gels through a strainer, resulting in about 750 g of conditioned gels.

Salsa cups and lids were weighed, and 30 g of gel were added to each cup. Then, the entire bait cup (cup+ gel + lid) was weighed again. The cups were kept in a refrigerator until used (within 1-2 days) and held on ice packs during transportation to the field site.

Two cups containing PAA and two cups containing ALG gel were placed in opposite corners in each bait station (Fig. 23). Six choice tests were set up (trap locations #1, 5, 22, 39, 42, and 44, Fig. 8). To estimate water loss from the hydrogel baits, four evaporation control stations with two ALG cups and two PAA gels cups were hung alongside a bait station (Fig. 24).

The choice test was initiated on 8/19/2019 at 1300 hours. At 1500 hours, one cup of PAA and 1 cup of ALG were removed from each station and the four evaporation control stations. The cups were covered, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

After 4 hours, all the remaining cups were empty, and the test was discontinued.

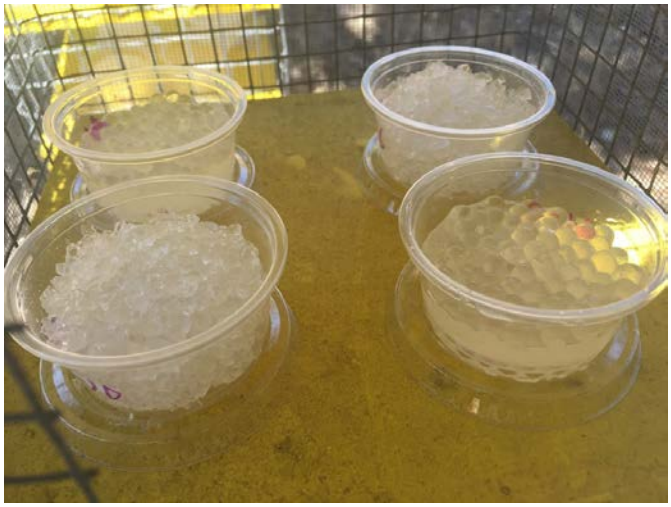

Fig. 23. Gel cups inside of a bait station. From the lower left, clockwise, cups contained PAA, ALG, PAA, and ALG hydrogels. The ALG hydrogels are spherical, and PAA hydrogels have an irregular shape.

Fig. 24. A bait station (left) and an evaporation control station (right). Note the fine metal screen on the evaporation check station to prevent yellowjackets from foraging on the hydrogels.

Choice Tests with Chicken Juice:Water PAA Gels

A choice test was conducted with PAA hydrogels prepared with different dilutions of chicken juice in water to determine the optimal amount of chicken juice to condition the hydrogels. The gels were prepared with pure chicken juice, chicken juice (diluted 1:1), and chicken juice (diluted 1:3) with deionized water (Table 15). The gels were conditioned overnight in the refrigerator and transported to the field on ice packs.

Table 15. Composition of PAA gels with various amounts of chicken juice and deionized water.

Bait name	Chicken Juice (ml)	Water (ml)	Total (ml)	PAA crystals (g)
Chicken juice gel (1:3 dilution)	30	90	120	6
Chicken juice gel (1:1 dilution)	60	60	120	6

Chicken juice gel (undiluted)	120	0	120	6

Salsa cups and lids were weighed, and 30 g of gel were added to each cup. The entire gel cup (cup + gel + lid) was weighed. Six choice tests with one cup of each concentration of chicken juice were set up in the field in the UCR bait stations. Evaporation control stations were hung alongside bait stations with 4 cups per concentration of chicken juice.

The choice test was initiated on 8/19/2019 at 1300 hours. After 3 hours, the cups were sealed and returned to the laboratory and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

Choice Test PAA Gels and Pet Food

Preliminary tests with PAA gels and whitefish pet food were conducted. The PAA gels were prepared with deionized water, chicken juice (1:1), and Swanson's chicken broth diluted with water 1:1. About 33.3 g of the PAA crystals were added to 400 ml of each fluid. The gels were conditioned in the refrigerator overnight.

Salsa cups and lids were weighed, and 30 g of gel or 30 g of whitefish pet food was placed in each cup. The entire bait cup (cup + gel + lid) was weighed and transported to the field on ice packs.

The test was initiated on 8/7/2019 and terminated on 8/8/2019. After 24 hours, the cups were covered, placed on ice, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

Choice Tests with Gels and Food Baits

A series of choice tests were conducted at several sites at Irvine Regional Park to determine which might be the most attractive. The choice tests conducted were as follows:

#1 - Swanson chicken, chicken juice, chicken juice (1:1 dilution), whitefish dinner, whitefish juice (1:1 dilution), whitefish juice (1:3 dilution).

#2 - Swanson chicken, chicken juice, chicken juice (1:1 dilution), chicken juice (1:3 dilution) (2 replicates).

#3 - Chicken juice, chicken broth, chicken broth (1:1 dilution) (3 replicates).

#4 - Fish juice hexane extract, fish juice water extract, and both extracts.

#5 - Chicken juice hexane extract, chicken juice water extract, and both extracts (3 replicates).

All dilutions were made with deionized water.

Salsa cups and lids were weighed, and 15 g of gel or solid food were added to each cup. The entire bait cup (cup + gel + lid) was weighed and transported to the field on ice packs.

The choice tests were conducted on 10/24/2019. Each choice station was placed out for 3 hours. The cups were covered, placed on ice packs, returned to the laboratory, and weighed. The

evaporation controls were conducted on 10/29/2019 on the UCR campus. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The amount of each fraction of chicken juice or fish juice taken was analyzed with a chi-square analysis.

Efficacy Trials

To determine the efficacy of dinotefuran baits against yellowjackets, baiting tests were conducted with low concentrations of dinotefuran, 0.00075%, 0.001%, and 0.0025% prepared in the PAA hydrogels. The baits consisted of 33.3 g of PAA crystals, 100 ml of chicken juice, and 300 ml of water (1:3 dilution). An aqueous stock solution of dinotefuran was added so that the final concentrations of the gels were 0.00075, 0.001, and 0.0025% (wt:vol). The hydrogels were allowed to condition in the refrigerator overnight.

Salsa cups and lids were weighed, and about 30 g of gel were added to each cup. Then the entire bait cup (cup+ gel + lid) was weighed. The baits were transported to the field on ice packets.

Three bait stations were placed at each site associated with a monitoring location with high yellowjacket trap counts. One station was placed next to a monitoring location, and the other bait stations were placed about 20 m away from the first one. The baits were placed out on 9/3/2019. Another site at the other end of the park greater than 400 m away from the bait sites was monitored and served as an untreated control.

After 24 hours, the bait cups were sealed and returned to the laboratory. The entire cup was weighed again. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The yellowjacket trapping data before and after baiting was analyzed with a paired *t*-test.

Results

Monitoring

The Park was monitored from 6/12/2019 until 10/28/2019. The number of workers trapped increased dramatically in early August, with 12 sites exceeding 10 YJ/T/D by mid-August (Fig. 25). The number of yellowjackets declined in early October. The total number of yellowjackets trapped during 2019 was 8,219.

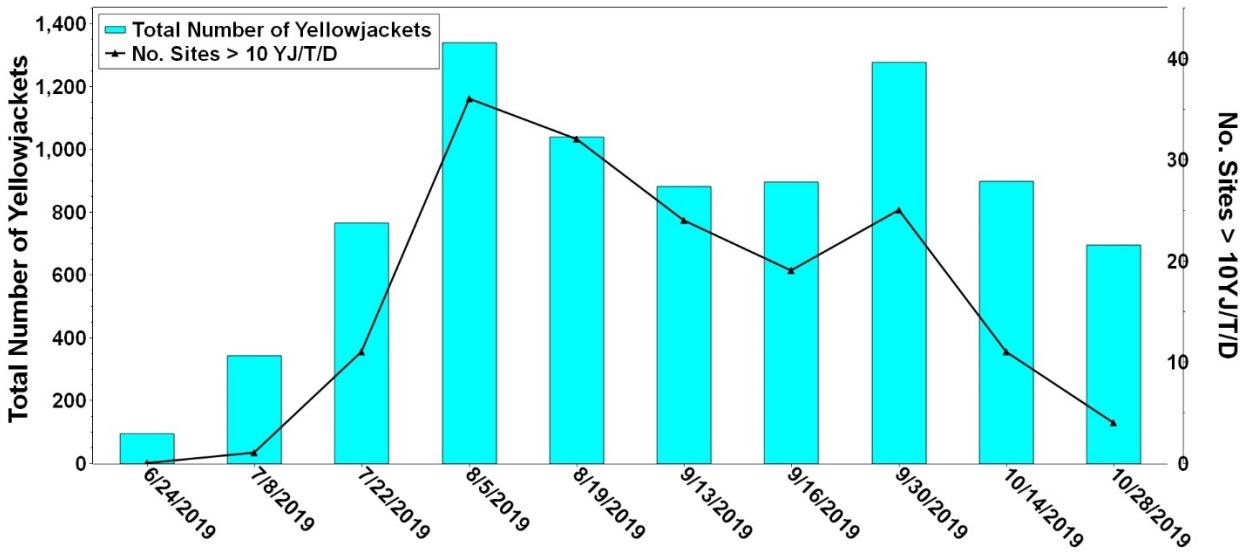

Fig. 25. The total number of yellowjackets trapped and the number of sites with > 10 yellowjackets/trap/day (YJ/T/D) at Irvine Regional Park during 2019.

Choice Test of Polyacrylamide (PAA) vs. Sodium Alginate (ALG) Hydrogels

All the PAA cups were empty or almost empty after 2-3 hours. At the sites where the PAA cups were empty, the ALG cups still had more than half of the bait left in them (Fig. 26). Based on data collected at 1500 hours, 19.0 ± 11 and 5.0 ± 3.4 g (mean ± SD, n = 6 cups each) of bait was removed by wasps from the cups. The PAA and ALG gels lost about 7.3% and 5.6% of their weight in 2 hours, respectively. When adjusted for water loss, the yellowjackets removed 122.8 g of PAA gels (about 68% of the total) at 2 hours. Only 32.7 g of the ALG gels (about 18% of the total) were taken at 2 hours. All the remaining bait cups were empty at 1750 hours.

Based on our site observation, the yellowjackets visited both gel baits. In contrast, wasps took a piece of PAA gel within 5 sec after landing on the bait cup, whereas wasps took more than 30 seconds longer to take off with a piece of ALG bead than with the PAA gel. The wasps spent more time handling the ALG beads before being able to take off with a small piece of the hydrogel.

Fig. 26. Choice test at monitoring location #1. The photo was taken about 2 hours after the initial setup. From the lower left, clockwise, cups contained PAA, ALG, PAA, and ALG hydrogels. Note both PAA cups were nearly empty while ALG cups still had most of the ALG hydrogel beads.

Choice Test with Chicken Juice:Water PAA Gels

The PAA gels conditioned in chicken juice were very attractive. All the gels from cups containing pure chicken juice or chicken juice and water (1:1) were removed within 2 hours. Very little of the diluted chicken juice (1:3) was removed at 2 hours, but it was removed entirely by the following morning (18 hours later).

Choice Tests with PAA Gels and Pet Foods

The yellowjackets preferred the Swanson chicken whenever it was a choice in the tests, with an average of 15.84 g (n=3) taken (Table 16). The PAA crystals conditioned with undiluted chicken juice were a second choice with an average of 11.1 g removed (n=6). The yellowjackets removed an average of 9.71 g (n=3) of PAA crystals conditioned in chicken broth. When the chicken juice was diluted with water (1:1), yellowjackets took an average of 4.66 g (n=3).

The hexane:water partition of the chicken juice was attractive. The yellowjackets removed an average of 17.6 g (n=3) of the three conditioned fractions. There was no significant difference in the amounts of each fraction retrieved by the yellowjackets ($\chi^2 = 1.67$, n = 2, **P** >0.05).

The canned whitefish dinner and the PAA gels conditioned in whitefish juice were not readily removed. The gels congealed together and were difficult to manipulate. Similarly, the

tests with the hexane and water partitions of the fish extract resulted in the lowest amount of bait removed in the studies.

Table 16. The removal of food materials and PAA gels ranked from various choice tests conducted at Irvine Regional Park.

Choice Test	Bait	Bait Taken (g)	% total	Rank
1	Swanson chicken	16.44	41.9	1
	Gels with chicken juice (undiluted)	14.91	38.0	2
	Gels with chicken juice (1:1)	4.17	10.6	3
	Whitefish pet food	1.58	4.0	4
	Gels with fish juice (1:1)	0.73	1.9	6
	Gels with fish juice (1:3)	1.33	3.6	5
	Total	39.30		
2	Swanson chicken	14.91	69.4	1
	Gels with chicken juice (undiluted)	1.58	7.4	3
	Gels with chicken juice (1:1)	4.24	19.8	2
	Gels with chicken juice (1:3)	0.73	3.4	4
	Total	21.46		
3	Swanson chicken	16.17	43.2	1
	Gels with chicken juice (undiluted)	10.96	29.3	2
	Gels with chicken juice (1:1)	5.57	14.9	3
	Gels with chicken juice (1:3)	4.74	12.7	4
	Total	35.64		
4	Gels with chicken juice (undiluted)	16.76	48.4	1
	Gels with chicken broth (undiluted)	11.18	32.3	2
	Gels with chicken broth (1:1)	6.66	19.3	3
	Total	34.60		
5	Gels with chicken juice (undiluted)	13.51	43.1	1
	Gels with chicken broth (undiluted)	10.03	32.0	2
	Gels with chicken broth (1:1)	7.82	24.9	3
	Total	31.36		
6	Gels with chicken juice (undiluted)	8.88	41.4	1
	Gels with chicken broth (undiluted)	7.93	36.9	2
	Gels with chicken broth (1:1)	4.66	21.7	3
	Total	21.47		
7	Gels with fish hexane	2.23	25.9	3
	Gels with fish water extract	3.04	35.3	2
	Gels with fish water + hexane extracts	3.35	38.8	1
	Total	8.63		
8	Gels with chicken hexane extract	2.76	13.5	3
	Gels with chicken water extract	9.67	47.2	1
	Gels with chicken water + hexane extracts	8.05	39.3	2
	Total	20.48		

9	Gels with chicken hexane extract	5.47	39.2	2
	Gels with chicken water extract	2.40	17.2	3
	Gels with chicken water + hexane extracts	6.09	43.6	1
	Total	13.97		
10	Gels with chicken hexane extract	5.11	36.6	3
	Gels with chicken water extract	6.78	48.5	1
	Gels with chicken water + hexane extracts	6.52	46.7	2
	Total	18.41		

Efficacy Trials

The yellowjackets removed about 20-25% of the dinotefuran bait placed out in stations within 24 hours. The 0.0025% dinotefuran bait provided a 40.6 and 37.3% reduction on days 14 and 28, but the YJ/T/D were still above the action threshold (Table 17). The trap counts increased with 0.001 and 0.00075% baits and the controls at day 28 (Table 18). On 10/15/2019 (42 days after baiting), yellowjacket traps started declining at all the sites at Irvine Regional Park. Even though there's a trend that shows a decline in the number of wasps associated with the treated locations starting as early as 14 days after the trial, no significant differences were found before 56 days, probably due to the limited number of monitoring stations and the high variability in trap catches at those sites. Significant reductions occurred at day 56 with 0.0075% dinotefuran bait ($T = 2.85$, $df = 4$, $\mathbf{P} = 0.05$) and 0.001% dinotefuran bait ($T = 5.10$, $df = 4$, $\mathbf{P} < 0.01$),

Table 17. Amount of bait removed at the bait stations containing 0.00075, 0.001, and 0.0025% dinotefuran PAA baits at Irvine Regional Park. The average number of yellowjackets/trap/day (YJ/T/D) and percent reduction at each location before baiting and 14, 28, 42, and 56 days after baiting.

Treatment	Bait taken (g)	Average YJ/T/D (% reduction)				
		Pre-baiting	14 days	28 days	42 days	56 days
Dinotefuran 0.00075%	69.1	17.6	18.5 (0.0 %)	19.8 (0.0 %)	13.3 (24.9 %)	9.2 (47.9 %)
Dinotefuran 0.001%	65.9	10.9	9.6 (11.2 %)	21.1 (0.0 %)	6.0 (45.0 %)	2.4 (78.3 %)
Dinotefuran 0.0025%	41.5	28.9	17.1 (40.6%)	18.1 (37.3 %)	10.0 (65.3 %)	6.0 (79.3 %)
Control		12.0	12.8 (0.0 %)	20.0 (0.0 %)	8.4 (29.9 %)	6.9 (42.6 %)

Conclusions

The findings were consistent with other San Diego Zoo Safari Park observations that chicken juice and water (1:3 dilution) were less preferred than pure chicken juice or chicken juice and water (1:1 dilution). To be competitive with food sources in the environment, the hydrogel baits need to be conditioned with higher concentrations of chicken juice.

Previous studies have shown that certain pet food containing whitefish and canned minced chicken were highly preferred and removed by yellowjackets (Rust et al. 2010). However, the fish pet food was not readily accepted by the foragers.

The presence of hexane-soluble oils on hydrogels' surface might initially attract the yellowjackets (unpublished data). Still, the presence of the oil was not sufficient to elicit the bait removal behavior of foraging yellowjackets. Our observation indicates that important phagostimulant(s) may be present in the aqueous fractions of the chicken juice.

The foragers readily accepted both the PAA and ALG gel baits conditioned in chicken juice, and all the offered bait was removed after 24 hours. However, the PAA gel was taken faster than the ALG. The wasps seem to spend more time handling the ALG beads, which explains the initial difference in the bait taken. ALG are larger and harder to handle, and some wasps were observed spending time “drinking” from the surface of gels in the cup. Since the goal of baiting is to have as much bait as possible removed to the colony before the foragers start to die under the effects of the insecticide, shorter handling time for the PAA gels might be essential to maximize the amount of bait taken. The handling time for ALG beads might be shortened if the hydrogel beads were cut into small/irregularly shaped pieces or the beads were made smaller. Also, the ALG beads lose less water than the PAA gels, which might extend the acceptability of the baits in the field. Based on our observation at the site, it was not clear if there is any inherent preference between PAA and ALG gel baits.

Relatively small amounts of the low concentrations of dinotefuran bait were taken, and there were no significant reductions in the number of yellowjackets trapped at day 28. This confirms an earlier study in which the dinotefuran baits were too toxic and killed workers too quickly (Rust et al. 2010), which results in reduced recruitment and bait take and, consequently, reduced effectiveness at reducing yellowjacket populations.

Irvine Regional Park 2020

Methods and Materials

Monitoring

The Park was surrounded by 56 traps initially installed on 6/29/2020 at the same locations as previous years (Fig. 20).

Choice Preference Tests

Choice tests were conducted to determine the acceptability of the potential toxicants such as the isoxazoline fluralaner and the neonicotinoid clothianidin, mixed in the minced chicken. The liquid juices were removed from the chicken and the chunks of chicken were chopped into fine pieces. About 80 ml of the chicken juice were added to 420 g of the minced chicken and thoroughly mixed.

Baits were prepared by mixing 1 tube of fluralaner (Bravecto® 250 mg) into the 80 ml of chicken juice and 420 g of finely minced chicken. The mixture was stirred and thoroughly mixed providing 0.05% fluralaner bait. This bait was diluted with untreated minced chicken to provide 0.025, 0.0125, and 0.00625% fluralaner bait. Clothianidin baits were prepared by mixing 25 g of clothianidin cockroach bait (Maxforce Impact, 1% clothianidin) into 420 g of minced chicken and 80 ml of chicken juice. This mixture was added to minced chicken to 0.1, 0.05, 0.025, and 0.0125% clothianidin yellowjacket bait.

Plastic salsa cups and lids were weighed, and ≈ 30 g of bait was added to each cup. Then the entire bait cup (cup + minced chicken + lid) was weighed again. The bait cups were refrigerated until tested.

Three bait stations were prepared for each series of bait. One cup of each concentration and an untreated cup of minced chicken were placed in a bait station for each toxicant. A bait station covered with a screen containing bait cups served as the evaporation control. The stations were placed at sites with high trap counts. The bait stations were deployed on 9/8/2020 and recovered on 9/9/2020.

After 24 hours, the cups were covered, placed on ice, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The data were analyzed with chi-square analysis.

Efficacy Trial on 10/12/2020

The number of yellowjacket foraging remained high at several sites in the park in September, and one late season baiting trial was conducted. Two sites were baited between 10/12/2020 to 10/13/2020 with 0.025% fluralaner in PAA gels (monitoring sites 35, 36, and 37) and 0.024% selamectin + 0.008% sarolaner gels (sites 41, 42, and 43). Monitoring traps were collected 7 days later.

After 24 hours, the cups were covered, placed on ice, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets trapped before and after baiting was analyzed with a Wilcoxon's signed-rank test.

Results

The total number of yellowjackets trapped at the 56 sites within the park is shown in Fig. 27. A total of 27,446 *V. pensylvanica* were trapped at Irvine Regional Park in 2020. The number of sites with > 10 YJ/T/D peaked on 9/8/2020.

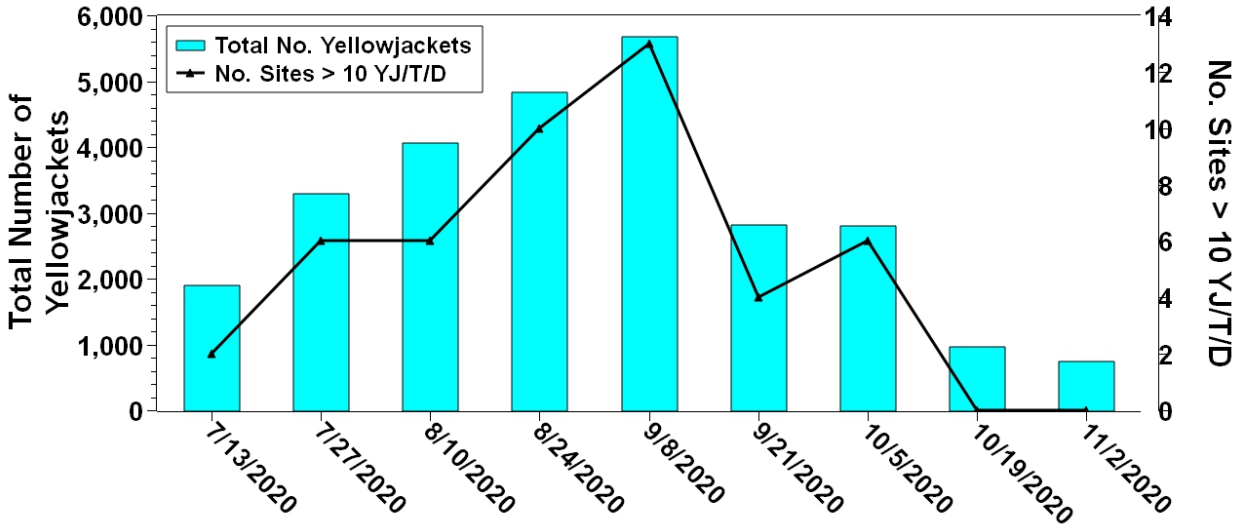

Fig. 27. The total number of yellowjackets trapped and the number of sites with >10 yellowjackets/trap/day (YJ/T/D).

Choice Preference Tests

There were no significant differences in the amount of the four concentrations of fluralaner bait removed by yellowjackets (χ^2 = 5.25, df = 4, **P** > 0.05, Table 18). The amount of treated and untreated bait removed by yellowjackets varied from 27 to 75%, depending upon the location of the bait station. About 49% of all the treated bait (194.5 g) was removed.

There were no significant differences in the amount of the four concentrations of clothianidin bait removed by the yellowjackets (χ2 = 0.561, df = 4, **P** > 0.05). About 16.2% of the treated bait was removed (63.8 g).

Table 18. Bait acceptance study with fluralaner and clothianidin mixed in finely minced chicken.[1]

Bait	Concn. (%)	Avg. (± SD) Amount Taken (g)	% of Total Taken
Fluralaner	0.05	17.3 ± 8.18	18.65
	0.025	14.7 ± 8.70	16.65
	0.0125	17.2 ± 11.05	19.65
	0.00625	15.6 ± 3.58	18.10
	0.0	21.7 ± 1.72	26.9
Clothianidin	0.1	4.2 ± 0.60	15.26
	0.05	5.8 ± 1.36	20.93
	0.025	5.1 ± 1.00	18.75
	0.0125	6.2 ± 0.61	25.70
	0.0	6.1 ± 0.14	22.36

[1] Choice tests conducted for 24 hours from 9/8/2020 to 9/9/2020.

The number of yellowjackets began declining after September 8 throughout the Park and the untreated control (Fig. 28). However, there was a stronger reduction in the number of yellowjackets trapped at the three choice baiting sites with the clothianidin bait (84.8%) compared to the control sites (45.5%) two weeks after the choice test.

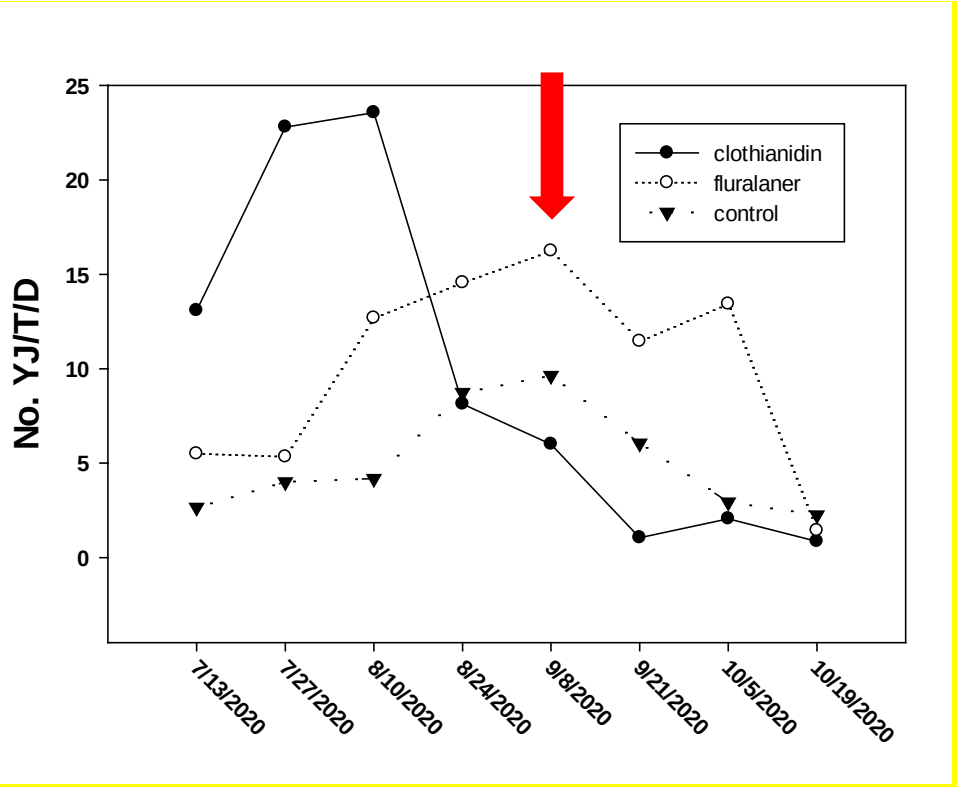

Fig. 28. The average number of yellowjackets/trap/day (YJ/T/D) at the two choice preference sites on September 8-9 and an untreated site on the opposite end of the park. The red arrow indicates when the choice tests were initiated.

Efficacy Trial 10/12/2020

The PAA gels in the evaporation control lost 22.2% of their weight in 24 hours. When adjusted for water loss, the yellowjackets removed 321.3 g of 0.025% fluralaner bait (61.2% of the total) and 412.6 g of 0.024% selamection + 0.004% sarolaner bait (77.9% of the total) in 24 hours (Table 19). The selamectin + sarolaner baits provided a significant reduction in the number of yellowjackets trapped after 7 days (W = 28, df = 6, **P** = 0.01) and 21 days (W = 21, df = 7, **P** = 0.03). The reductions with the 0.025% fluralaner were not significant. However, only 4 monitoring traps were close to those bait stations, and the variability in counts and the low number of traps limited the statistical analysis. The number of yellowjackets trapped in the untreated area remained unchanged during the test.

Table 19. The efficacy of 0.025% fluralaner and 0.024% selamectin + 0.004% sarolaner baits in a late-season baiting conducted on 10/12/2020 at Irvine Regional Park.

Baits	Bait Taken (g)	No. Monitoring Traps	Average YJ/T/D (% reduction)		
			Pre-baiting	7 days	21 days
0.025% fluralaner	321.4	4	6.4	0.6 (91.1%)	0.9 (85.5%)
0.025% selamectin + sarolaner	412.6	7	10.5	1.9 (82.1%)	1.0 (90.5%)
Untreated		8	2.24	2.2 (0.0%)	1.7 (25.6%)

Discussion

The yellowjackets removed all concentrations of fluralaner and clothianidin tested. During the 1-day test period, > 194 g of treated fluralaner bait were removed compared with only 61.2 g of the clothianidin. The fast-acting clothianidin likely decreased overall foraging. There was a significant decrease in the number of yellowjackets trapped in the monitoring traps near the fluralaner choice test. The data certainly warrants additional testing with 0.05% fluralaner and lower concentrations of the clothianidin bait.

The 0.024% selamectin + 0.004% sarolaner bait was well taken and provided a significant reduction in the number of yellowjackets before the yellowjacket populations declined in late October. The 0.025% fluralaner was also well taken, and the reduction in yellowjackets trapped exceeded 80%.

Irvine Regional Park 2021

Methods and Materials

Monitoring

The foraging activity of yellowjackets was measured using a total of 56 UCR traps (Fig. 20). Monitoring began on 6/14/2021 and ended on 11/17/2021. The collection jars were changed every 14 days. The heptyl butyrate vials were replaced as needed.

Efficacy Trial #1

The 0.05% fluralaner bait was prepared with 500 ml of diluted chicken juice (1:1), 33.3 g of PAA crystals, and 250 mg of fluralaner. The selamectin + sarolaner bait was prepared with 500 ml of diluted chicken juice (1:1), 33.3 g of PAA crystals, and 12 tubes of Revolution Plus (720 mg of selamectin + 120 mg sarolaner). The mixtures were allowed to condition for 24 hours in the refrigerator.

The salsa cups and lids were weighed, and filled with ≈ 25 g of bait, and weighed again. Four bait stations were assembled for each type of bait, with four cups of bait in each station. The fluralaner bait stations were hung at sites 25, 26, 27, and 50. The selamectin + sarolaner baits were hung at sites 38, 39, 40, and 42. Four cups of each bait were placed in two evaporation control stations (Sites 41). The baits were hung on 8/24/2021 and retrieved on 8/26/2021. The traps at sites 1-10 served as controls.

After 24 hours, the cups were covered, placed on ice, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The trap counts were analyzed with a Wilcoxon signed-rank test.

Efficacy Trial #2

The same sites from 8/24/2021 were treated again on 9/21/2021 to 9/23/2021 with the same baits as described above.

Results

Monitoring

The only species trapped was *V. pensylvanica*. Two queens were trapped between 6/14/2021 and 6/28/2021. A total of 17,795 yellowjackets were trapped during 2021 (Fig. 29).

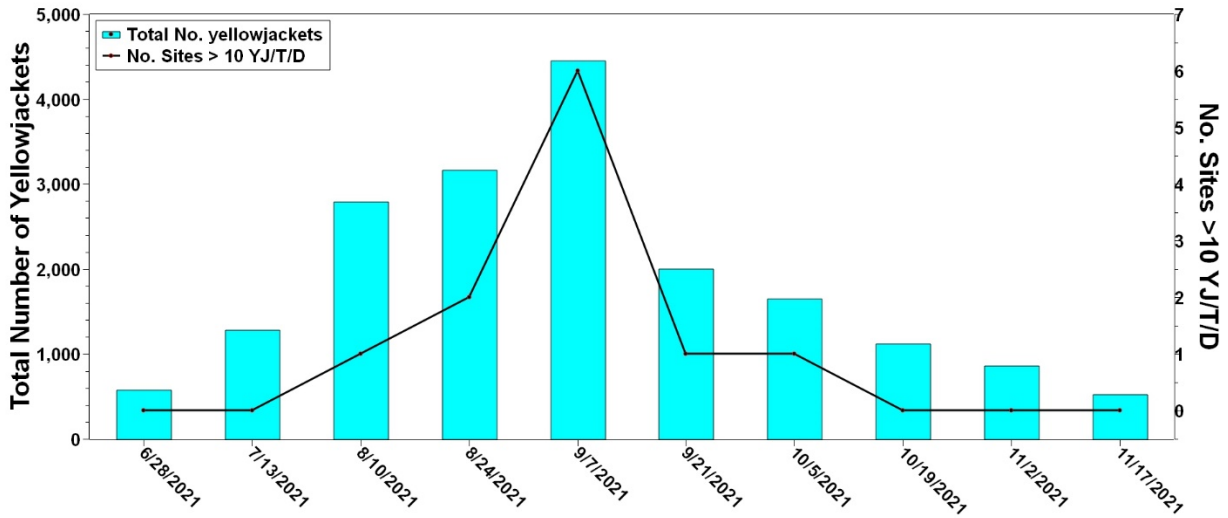

Fig. 29. The total number of yellowjackets trapped and the number of sites with >10 yellowjackets/trap/day (YJ/T/D) at Irvine Regional Park in 2021.

Efficacy Trial # 1

The weather had been warm, and the baits in the evaporative control lost about 70% of their weight over the 48-hour baiting period. After compensating for the water loss, the yellowjackets removed 40.0 g of 0.05% fluralaner (about 9% of the total) and 219.2 g of selamectin + sarolaner (about 54% of the total). The 0.05% fluralaner baits provided significant reductions in the number of yellowjackets trapped after 14 days (W = 34, n = 8, **P** = 0.01) and 28 days (W = 36, n = 8, **P** = 0.007, Table 20). Similarly, the 0.144% selamectin + 0.024% sarolaner provided significant reductions after 14 days (W = 55, n = 10, **P** = 0.002) and 28 days (W = 55, n = 10, **P** = 0.002). The untreated controls declined after 14 days (W = 46, n = 10, **P** = 0.01), but were unchanged after 28 days (W = 35, n = 10, **P** = 0.08). The selamectin + sarolaner bait was more readily removed by the yellowjackets than the fluralaner bait.

Table 20. The efficacy of 0.05% fluralaner and 0.144% selamectin + 0.024% selamectin PAA gel baits.

Toxicant	Bait Taken g (% of total)	Monitoring Sites	Average YJ/T/D (% reduction)		
			Pre-baiting	14 days	28 days
0.05% fluralaner	40.0 (9.4%)	8	5.78	1.93 (55.2%)	2.23 (50.9%)
0.144% selamectin + 0.024% sarolaner	219.3 (54.5%)	10	6.96	2.45 (64.8%)	1.52 (78.2%)
Untreated		10	4.00	3.03 (24.2%)	3.16 (21.0%)

EfficacyTrial # 2

The weather was warm, and the baits in the evaporative controls lost about 70% of their weight. After compensating for the water loss, the amount of 0.5% fluralaner bait removed was very low compared with the 0.144% selamectin + 0.024% sarolaner bait (Table 21). The fluralaner resulted in significant reductions in the numbers of yellowjackets after 14 days (W = 28, n = 10, P = 0.01), 28 days (W = 36, n = 10, **P** < 0.01), and 42 days (W = 36, n = 10, **P** < 0.01). The selamectin + sarolaner also provided significant reductions after 14 days (W = 34, n = 9, **P** = 0.04), 28 days (W = 55, n = 9, **P** = 0.002), and 42 days (W = 55, n = 9, **P** = 0.002). In the untreated controls, there was a significant reduction after 14 days (W = 35, n = 10, **P** = 0.3) and 42 days (W = 45, n = 10, **P** = 0.003).

Table 21. The efficacy of 0.05% fluralaner and 0.144% selamectin + 0.024% selamectin PAA baits against western yellowjackets.

Toxicant	Bait Taken g (% of total)	Monitoring Sites	Average YJ/T/D (% reduction)			
			Pre-baiting	14 days	28 days	42 days
0.05% fluralaner	2.5 (0.6%)	8	1.29	0.38 (70.6%)	0.39 (69.4%)	0.21 (83.3%)
0.144% selamectin + 0.024% sarolaner	228.4 (53.1%)	10	1.52	1.10 (27.7%)	0.76 (50.2%)	0.49 (68.1%)
Untreated		10	3.16	2.67 (15.4%)	1.86 (41.2%)	1.57 (50.2%)

Discussion

The number of YJ/T/D never reached the treatment threshold at most sites in Irvine Regional Park. However, it was decided to try to reduce the numbers at those sites with > 10 YJ/T/D. Those sites were baited on 9/21/2021. The selamectin + sarolaner bait was well accepted by foraging yellowjackets and provided significant reductions in the number of yellowjackets trapped after the first baiting.

The numbers of yellowjackets trapped at the 56 different sites in the park significantly declined after 9/21/2021, and the second baiting data was not conclusive.

Silent Valley RV Park 2019

Silent Valley Recreational Vehicle Camp is located about 8 km south of Banning, CA in the San Jacinto Mountains (33°50′57.51″ N, 116°51′08.45″ W; elev. 1,093 m) on California Route 243. The year around park consists of about 186 ha with 850 campsites. The Park supports multiple activities, including a small lake, swimming pools, a restaurant, and a general store. The campground is covered with numerous oaks, and the park is surrounded by native chaparral. Over the last 10 years, there have been sporadic problems with yellowjackets.

Methods and Materials

Monitoring

A total of 43 UCR-style monitoring traps were hung along the park's perimeter, beginning on 6/3/2019 and ending on 10/7/2019. The traps were checked every 14 days, and the heptyl butyrate vials were changed as needed.

Efficacy Trial

The dinotefuran bait was prepared with 100 ml of chicken juice, 600 ml of water, and 40 g of PAA crystals. The mixture was placed in the refrigerator, and the hydrogels were allowed to condition for 48 hours. A 0.1% aqueous solution of dinotefuran was prepared. Appropriate quantities of 0.1% dinotefuran were added to 100 g of the conditioned hydrogel to make baits containing 0.0025, 0.001, and 0.00075% dinotefuran. The baits were stored overnight in the refrigerator.

Plastic salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. The entire cup (cup + bait + lid) was weighed again. The bait cups were refrigerated and transported to the field on ice packs.

Three bait cups of each bait concentration were placed in UCR-style bait stations. The three bait stations were hung within about 15 m of each other. Evaporation controls with five cups were hung alongside bait stations to estimate water loss from hydrogel baits. After 24 hours, the bait cups and evaporation controls were removed, covered, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The amount of bait taken by the yellowjackets was analyzed with a one-way ANOVA and means compared with a Tukey’s HSD.

Results

Monitoring

Queens of *V. pensylvanica* were collected on 6/17/2019 (n=4), 7/1/2019 (n=3), 7/15/2019 (n=2), and 7/29/2019 (n=1) along with a few workers. Low numbers of *V. atropilosa* and *V. sulphurea* workers were collected beginning 7/29/2019 (Table 22). However, the most predominant species trapped (n = 29,451) throughout the summer was *V. pensylvanica* (Fig. 30).

Table 22. The number of *V. atropilosa* and *V. sulphurea* workers trapped at Silent Valley RV Park during 2019.

Species	7/29/2019	8/12/2019	8/26/2019	9/9/2019	9/23/2019	10/7/2019
atropilosa	0	27	44	5	10	0
sulphurea	3	5	11	10	1	2

Efficacy Trial

In the evaporation controls, the 0.00075%, 0.001%, and 0.0025% dinotefuran baits lost 15.0%, 14.9%, and 13.7% of their weight in 24 hours. After adjusting for water loss, the yellowjackets removed the baits in the following order: 0.001% > 0.00075 > 0.0025% ($F = 54.94$, df = 2, 24; $\mathbf{P} < 0.001$).

The 0.001% and 0.0025% dinotefuran baits resulted in 62.5% and 58.7% reductions in the number of yellowjackets trapped at day 28, respectively (Table 23). Of the 270 g of bait deployed at each site, about 24.9%, 18.7%, and 2.8% of the 0.00075%, 0.001%, and 0.0025% dinotefuran baits, respectively, were removed by yellowjackets within 24 hours.

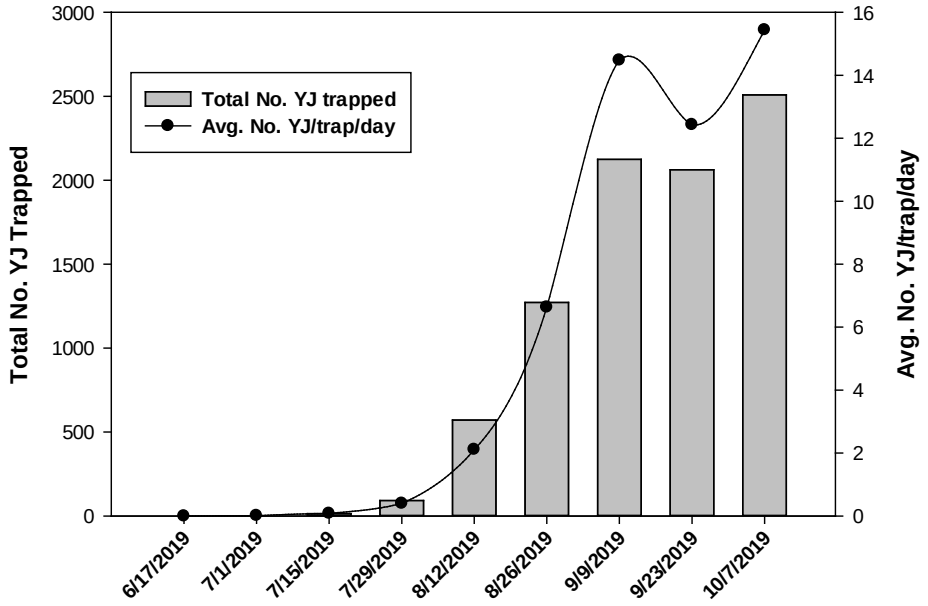

Fig. 30. The total number of *V. pensylvanica* trapped and the average number of yellowjackets/trap/day (YJ/T/D) for Silent Valley in 2019.

Table 23. Dinotefuran bait removal by yellowjackets and the average number of yellowjackets trapped (YJ/T/D) before and after baiting.

Treatment	YJ/T/D (no. traps)[a]	Total bait[b] removed (g)	Average YJ/T/D (% Reduction)	
			14 days	28 days
0.0025%	16.4 (1)	7.6 a	8.5 (48.3%)	6.8 (58.7%)
0.001%	11.8 (3)	67.5 c	7.7 (35.1%)	4.4 (62.5%)

0.00075%	11.0 (4)	50.6 b	10.2 (7.6%)	11.2 (0.0%)
Untreated	16.8 (14)		13.0 (23.5%)	16.0 (5.1%)

[a] The number of monitoring traps near the bait stations in parenthesis.
[b] Means followed by the same letter are not significantly different at **P** < 0.05.

Discussion

There were not enough monitoring stations at all the bait stations to permit the statistical analysis of the baiting trial. The amount of 0.0025% dinotefuran baits removed by yellowjackets was less than 10 g. The 0.001% dinotefuran provided a 62.5% reduction at 28 days, and the two lower concentrations of dinotefuran were better accepted. The lowest concentration, 0.00075%, failed to reduce the number of yellowjackets trapped. This data is consistent with other studies that suggested that higher concentrations of dinotefuran kill the foragers too quickly and prevent them from recruiting other foragers (Rust et al. 2010).

Silent Valley Park 2020

Methods and Materials

Monitoring

The foraging activity of yellowjackets was monitored using UCR-style traps. Initially, 43 traps were hung under trees and bushes about 0.5-1.5 m off the ground on 6/29/2020. An additional 10 traps were added on 7/13/2020. On 7/27/2020, 3 more traps were added, making up a total of 56 traps along the perimeter of the RV park. The length of each monitoring session was 14 days. The monitoring continued until 11/2/2020.

Choice Tests with ALG and PAA Hydrogel Baits

Hydrogels made from biodegradable alginate (ALG) or polyacrylamide gels (PAA) were conditioned with diluted chicken juice. A choice test was conducted to determine which hydrogel was more attractive and taken by yellowjackets. Details regarding the production of the ALG and PAA gels see Irvine Regional Park 2019 (Methods and Materials section page 39). The choice test was conducted on 8/24/2020.

Plastic salsa cups and lids were weighed, and ≈ 20 g of each hydrogel was added to each cup. Then the entire bait cup (cup+ gels + lid) was weighed again. A total of 20 cups were prepared for each hydrogel type. The cups were placed on ice packs and transported to the field.

The UCR-style bait stations served as choice test arenas. The salsa cups with PAA and ALG gels were placed into the stations. The choice tests were placed at 4 active monitoring sites where the number of yellowjackets trapped per day ranged from 10.9 to 30.8. The choice tests were conducted for 4 hours. A site with low yellowjacket activity was selected for the evaporative control. The evaporative controls were placed in a bait station covered with window screen. After 4 hours, the cups were retrieved, covered, placed in a cooler, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The data was analyzed with a paired *t*-test.

Choice Preference Tests

Choice tests were conducted to determine the acceptability of the isoxazolines, fluralaner and sarolaner, and the neonicotinoid clothianidin mixed in minced chicken.

The baits were prepared with 80 ml of chicken juice, 420 g of minced chicken, and 1 tube of Bravecto (250 mg fluralaner) resulting in a 0.05% fluralaner bait. The 0.05% bait was diluted with untreated chicken to provide 0.025, 0.0125, and 0.00625% fluralaner. The selamectin + sarolaner baits were prepared by mixing 4 tubes of Revolution Plus (240 mg selamectin + 40 mg sarolaner) into 80 ml of chicken juice and 420 g of the minced chicken. The initial concentration (0.048% selamectin + 0.008% sarolaner) was diluted by mixing with untreated chicken to make the lower concentration listed in Table 24. The clothianidin bait was prepared by mixing 50 g of clothianidin cockroach bait (Maxforce Impact, 1% clothianidin) into 80 ml chicken juice and 420 g of minced chicken to make a 0.1% clothianidin yellowjacket bait. Appropriate quantities of 0.1% clothianidin bait were mixed with untreated minced chicken to make 0.025, 0.0125, and 0.00625% baits.

Table 24. Concentrations of the active ingredients in the selamectin + sarolaner baits prepared in minced chicken.

tubes /500 g chicken	Active ingredient (mg)		% Concn		
	selamectin	sarolaner	selamectin	sarolaner	combination
4	240	40	0.048	0.008	0.056
2	120	20	0.024	0.004	0.028
1	60	10	0.012	0.002	0.014
0.5	30	5	0.006	0.001	0.007

Plastic salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. Then, the entire bait cup (cup+ minced chicken baits + lid) was weighed again. Bait cups were refrigerated until they were deployed.

Three UCR-style bait stations were deployed for each choice preference test. One cup of each concentration and an untreated chicken cup were placed in a bait station for each toxicant. The stations were placed at sites with high trap counts. The choice tests were conducted on 9/8/2020 and lasted 4 hours. Bait cups were also placed in a bait station covered with screen to serve as evaporative controls. After 4 hours, the cups were covered with a lid, returned to the laboratory, and weighed.

After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The data was analyzed with Chi-square analysis.

Efficacy Trials

Three contiguous monitoring sites in which the average trap counts exceeded 100 yellowjackets per trap for the previous 14 days on 9/8/2020 were baited on 9/21/2020. The baits were prepared with minced chicken and PAA hydrogels (Table 25). The baits with minced chicken were prepared with 80 ml of the chicken juice and 420 g of minced chicken and thoroughly mixed. One and a half tubes of Bravecto were added to the mixture to make a 0.05% and 0.025% fluralaner final concentration, respectively. Four tubes of Revolution Plus were

added to the minced chicken to make a final concentration of 0.048% selamectin + 0.008% sarolaner. The baits were stored in the refrigerator and transported to the field on ice packs.

About 15 g of PAA crystals were soaked in 300 ml of chicken juice (1:1 dilution) to prepare the PAA hydrogels. Four tubes of Revolution Plus were added to the mixture to make a final concentration of 0.048% selamectin + 0.008% sarolaner. The sodium selenate and sodium selenite baits were made by adding 10 or 50 mg of each to the chicken juice and crystals. All gels were conditioned in the refrigerator overnight.

Plastic salsa cups and lids were weighed, and ≈ 30 g of bait was added to each cup. Then the entire bait cup (cup + bait + lid) was weighed again. Bait cups were refrigerated until they were deployed.

UCR-style bait stations were provisioned with three bait cups containing 30 g of each bait and hung within 15 m of the monitoring traps. Bait cups in a bait station covered with window screen served as evaporative controls. After 24 hours, the bait cups were collected, covered, returned to the laboratory, and weighed. The data were analyzed with Chi-square analysis.

The number of yellowjackets trapped was compared with Wilcoxon signed-rank test and, when possible, a paired *t*-test with small sample sizes.

Table 25. Yellowjacket baits tested at Silent Valley on 9/21/2020.

Toxicant	Bait base[a]	Bait Sites	Pre-Count YJ/T/D (9-8-2020)
Fluralaner, 0.05%	chicken	F91, F87, F80	16.1
Fluralaner, 0.025%	chicken	F73, F69, F59	14.9
Selamectin, 0.048% + sarolaner, 0.008%	chicken	F50, F48, F44	8.2
Selamectin, 0.048% + sarolaner, 0.008%	PAA	F39, F27, F17	12.6
0.01% Na Selenate	PAA	A551, A560, A568	12.6
0.005% Na Selenate	PAA	A699, A708, A721	10.4
0.01% Na Selenite	PAA	B776, B804, BIP	22.3
0.005% Na Selenite	PAA	C438, C421, C407	14.1
Control site		BRH, B848, B843	12.0

[a] Minced chicken, PAA – polyacrylamide gels

Results

Monitoring

The yellowjackets trapped during the 2020 season included 8 *V. pensylvanica* queens and 36,547 *V. pensylvanica* workers, 400 *V. atropilosa* workers, and 1,701 *V. sulphurea* workers (Fig. 31).

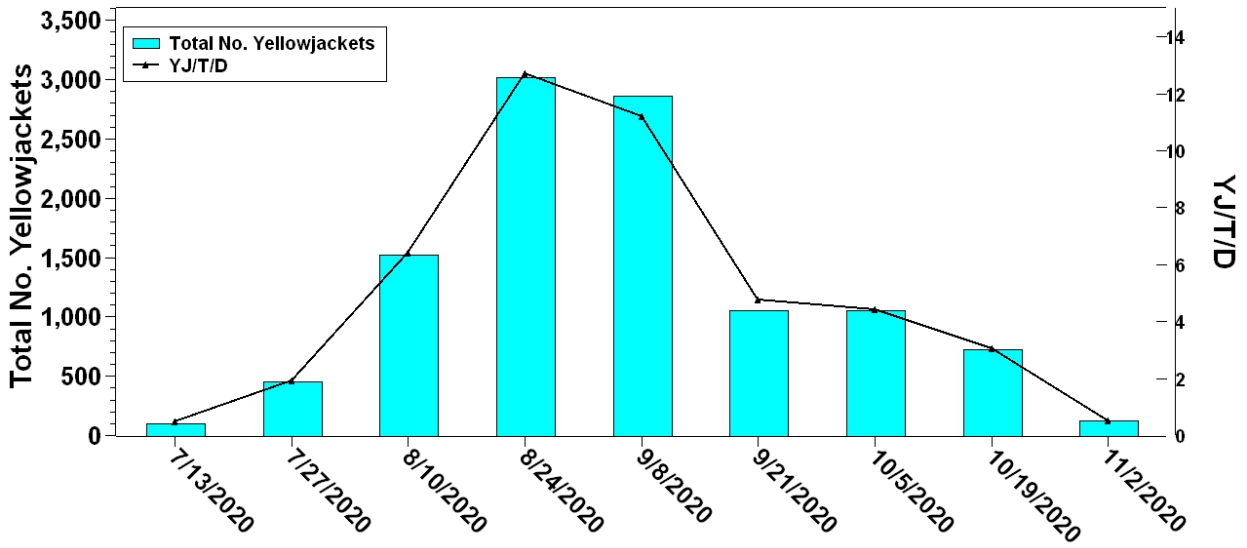

Fig. 31. The total number of yellowjackets trapped and the average number of yellowjackets/trap/day (YJ/T/D) for the untreated sites at Silent Valley in 2020.

Choice Tests with ALG and PAA Hydrogel Baits

When the choice tests were observed at 90 minutes, the majority of the yellowjackets were on the cups with PAA hydrogels. The workers cut the PAA hydrogels into manageable pieces with their mandibles. The amount of PAA gel bait removed by yellowjackets was significantly greater than the ALG bait at 24 hours (Table 26; $t = 2.41$, df = 6, **P** = 0.05).

Table 26. The amount of ALG and PAA gel baits removed by yellowjackets[a]

Gel bait	n	Amount taken (Avg ± SD)	% removed
ALG	7	14.2 ± 4.85	70.9
PAA	7	19.4 ± 0.18	96.5

[a] Tests completed at 4 hours.

Choice Tests with Experimental Toxicants

The yellowjackets took significantly less of the 0.05% fluralaner than the lower concentrations and the untreated chicken (Table 27, $\chi^2 = 33.5$, df = 4, **P** < 0.005). Yellowjacket foragers retrieved a total of 135.4 g of treated bait within 4 hours. There was a 63.6% reduction in the number of yellowjackets trapped 14 days later at those three bait sites.

There was no preference for any of the concentrations of selamectin + sarolaner or the untreated chicken ($\chi^2 = 3.98$, df = 4, **P** > 0.05). Yellowjacket workers removed 249.6 g of treated bait. There was an 83.8% reduction in the number of yellowjackets trapped 14 days later at the three baiting sites compared with a 19% reduction at 11 traps in an untreated area of the park.

The two highest concentrations of clothianidin taken were significantly less than the two lowest concentrations and the untreated bait ($\chi^2 = 24.18$, df = 4, **P** < 0.005). Foragers took only 20.8 g of treated bait in 4 hours. There was a 55.3% reduction in the number of yellowjackets trapped 14 days later.

There was a 19.0% reduction in the number of yellowjackets trapped 14 days after the choice tests in the control areas.

Table 27. Bait acceptance studies with fluralaner, selamectin + sarolaner, and clothianidin mixed in finely minced chicken.[1]

Bait	Concn. (%)	Avg. Amount Taken (± SD)	% Total Taken
Fluralaner	0.05	8.9 (±1.95)	15.7
	0.025	12.0 (± 0.84)	21.6
	0.0125	11.1 (±1.64)	19.9
	0.00625	9.4 (± 4.62)	16.8
	0.0	10.5 (± 3.29)	18.9
Selamectin + sarolaner	0.056	24.3 (± 0.66)	23.8
	0.028	18.7 (± 2.78)	18.3
	0.014	18.2 (± 0.52)	17.8
	0.007	22.0 (±7.61)	21.5
	0.0	19.1 (± 5.73)	18.7
Clothianidin	0.1	1.0 (± 0.17)	11.1
	0.05	1.3 (± 0.75)	14.3
	0.025	2.1 (± 1.02)	23.8
	0.0125	2.6 (±1.72)	29.5
	0.0	15.6 (± 0.21)	21.2

1 Choice tests conducted for 4 hours on 9/8/2020.

Efficacy Trials

Only a small amount of 0.05% fluralaner bait in minced chicken was removed compared with the 0.025% fluralaner bait after 24 hours (Table 28). The 0.048% selamectin + 0.008% sarolaner PAA and chicken baits were taken by yellowjackets. Yellowjackets readily accepted the sodium selenate and sodium selenite PAA baits.

Table 28. The amount and percentage of bait taken by yellowjackets in 24 hours.

Bait[a]	Concn. (%)	Bait Base	Amount Taken (g)	% Total Bait Taken
Fluralaner	0.05	Chicken	23.73	7.16
Fluralaner	0.025	Chicken	106.79	30.71
Selamectin + sarolaner	0.048 + 0.008	PAA	80.55	20.22
Selamectin + sarolaner	0.048 + 0.008	Chicken	78.67	21.82
Na selenate	0.01	PAA	169.84	58.86
Na selenate	0.005	PAA	217.72	55.90
Na Selenite	0.01	PAA	161.83	41.64
Na Selenite	0.005	PAA	84.08	21.77

[a] Baits were placed out 9/21/2020 and picked up 24 hours later.

It was not possible to statistically test the trap counts for all the baits applied on 9/21/2021. Some monitoring sites were too far away from bait stations, and thus, the number of monitoring traps nearby was limited. The selamectin + sarolaner formulated in the PAA gels provided significant reductions in the number of yellowjackets trapped at days 14, 28, and 42 (W= 21, n = 6, **P** = 0.03, Table 29). The selamectin + sarolaner in the minced chicken also provided significant reductions in the number of yellowjackets trapped at days 14, 28, and 42 (W= 21, n = 6, **P** = 0.03).

There were no significant reductions in the average number of yellowjackets trapped after baiting with 0.01% sodium selenite for 42 days. The 0.005% sodium selenite failed to significantly decrease the number of yellowjackets trapped over the entire 42 days.

The average number of yellowjackets trapped in the untreated controls remained unchanged from 9/21/2020 until 10/19/2020 and significantly declined at day 42 (W = 64, Z = 2.82, **P** = 0.002).

Table 29. The average number of yellowjackets per trap per day (YJ/T/D) before and after baiting with experimental baits.

Toxicant	Bait base	Average YJ/T/D (% reduction)			
		Pre-baiting	14 days	28 days	42 days
Fluralaner, 0.05%	Minced chicken	2.9	0.5 (82.2%)	0.3 (89.0%)	0.1 (96.3%)
Fluralaner, 0.025%	Minced chicken	2.59	0.60 (76.5%)	0.27 (89.4%)	0.14 (94.4%)
Selamectin 0.048% + sarolaner 0.008%	Minced chicken	2.33	0.42 (82.2%)	0.22 (89.0%)	0.09 (96.3%)
Selamectin 0.048% + sarolaner 0.008%	PAA	1.64	0.47 (71.1%)	0.32 (80.4%)	0.10 (94.2%)
Sodium selenate, 0.01%	PAA	6.60	8.92 (0.0%)	0.55 (91.7%)	0.45 (93.1%)
Sodium selenate, 0.005%	PAA	6.02	11.69 (0.0%)	5.10 (15.4%)	0.48 (92.1%)
Sodium selenite, 0.01%	PAA	3.29	3.08 (6.4%)	1.52 (53.6%)	0.12 (96.4%)
Sodium selenite, 0.005%	PAA	0.86	0.62 (28.2%)	0.52 (38.9%)	0.29 (66.7%)
Untreated		8.18	8.17 (0.2%)	4.42 (46.0%)	0.60 (92.7%)

Discussion

The polyacrylamide gels (PAA) conditioned in chicken juices and water were more readily removed than the alginate hydrogels. Within 4 hours, nearly 100% of the PAA gels were removed. At 24 hours, about 70% of the ALG gels were removed. The yellowjackets were able to manipulate the PAA crystals easier than the ALG beads. If the ALG beads were smaller and possible cut into pieces, they might be easier to handle.

The yellowjacket foragers readily took the baits containing 0.025% fluralaner and all concentrations of selamectin + sarolaner. Significantly less 0.05% fluralaner in minced chicken was taken, indicating that this concentration was somewhat repellent. All the concentrations of clothianidin had minimal amounts of bait removed, suggesting that clothianidin was too fast-acting and repellent. All concentrations of sodium selenate and sodium selenite were taken by foraging yellowjackets but failed to reduce the number of yellowjackets trapped.

The number of yellowjackets trapped peaked in late August and early September. The numbers declined in September when the first efficacy trial was being applied. The numbers of yellowjackets remained constant in the untreated sites throughout September. The selamectin + sarolaner formulated in minced chicken and PAA gels baits provided significant reductions for 28 days after baiting.

Silent Valley RV Park 2021

Commercial supplies of heptyl butyrate had not been available for the previous 12 months. We had enough heptyl butyrate to conduct the monitoring in 2021. However, we explored the possibility of using another attractant. Heptyl crotonate was effective as an attractant (Wagner and Reierson 1964). The relative efficacy of heptyl crotonate and heptyl butyrate has not been determined. Dr. Jocelyn Millar synthesized the heptyl crotonate and preference tests were conducted to compare both compounds.

Methods and Materials

Monitoring

A total of 56 UCR traps were placed along the perimeter of the RV park. The monitoring began on 6/14/2021 and continued until 9/27/2021. The length of each monitoring session was 14 days, except for the last period, which was 21 days.

Synthesis of Heptyl Crotonate

A mixture of crotonic acid (137.6 g, 1.6 mol), 1-heptanol (216 g, 2.05 mol), and 8 ml concentrated sulfuric acid was refluxed in 500 mL benzene overnight, with a Dean-Stark trap to remove water. The solution was then cooled to room temp, diluted with 500 mL hexane, and washed sequentially with water, saturated aq. $NaHCO_3$, brine, then dried over anhydrous Na_2SO_4 and concentrated under reduced pressure. The crude product was fractionally distilled, removing the excess heptanol by heating to 60°C at a 9 mm Hg vacuum. The remainder was then distilled with a Kugelrohr distillation apparatus in two batches (bp~65-70 °C at 0.25 mm Hg), giving a quantitative yield, >98.5% pure by gas chromatography on a nonpolar DB-5 column.

Choice Tests with Heptyl Butyrate and Heptyl Crotonate

The attractiveness of heptyl butyrate (HB) and heptyl crotonate (HC) foraging activity of yellowjackets was compared using UCR-style traps provisioned with 8-ml vials containing about 7.2 ml of heptyl butyrate or heptyl crotonate and a 5-cm piece of dental wick. One trap with HB and one with HC were hung under trees and bushes about 0.5-1.5 m off the ground and about 5 m apart. A total of 8 sites were used. The test began on 7/27/2021 and ended on 8/9/2021. The traps were returned to the lab, and the number of yellowjackets and species was recorded.

Traps with either the 8-ml vial containing about 7.2 ml of HB or HC and a 5-cm piece of dental wick were hung outdoors. The vials were weighed daily. The evaporation rates of HB and HC were determined by determining the change in weight of the vials and dental wick.

The trap catches were compared with a paired *t*-test.

Results

Monitoring

The numbers of *V. pensylvanica* never reached the critical threshold of 10 YJ/T/D throughout the 2021 season (Fig. 32). The total number of yellowjackets trapped included, 1 queen and 6,010 workers of *V. pensylvanica*, 1 queen and 2 workers of *V. atropilosa*, and 7 queens and 234 workers of *V. sulphurea*. The trap counts peaked on 8/23/2021 and gradually declined through September.

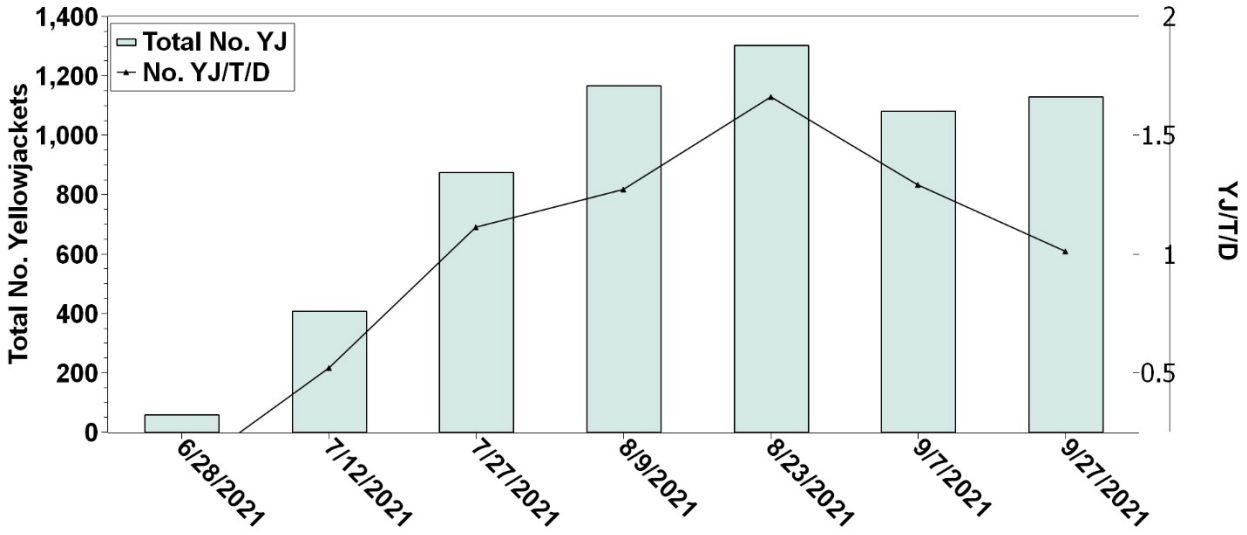

Fig. 32. The total number of yellowjackets trapped and the average YJ/T/D for 2021 at Silent Valley RV Park.

Choice Tests with Heptyl Butyrate and Heptyl Crotonate

The traps with HB and HC caught two yellowjacket species, *V. pensylvanica* and *V. sulphurea*. There were no significant differences in the numbers of *V. pensylvanica* in the HC (n = 196) or HB traps (n = 149) (T = 1.13, df = 7, **P** = 0.296). Similarly, there were no differences

in the number of *V. sulphurea* caught by either attractant (HC = 21 and HB = 16, T = -0.47, df = 7, **P** = 0.65).

Both HB and HC had similar evaporation rates (0.3 g/day; ≈ 0.35 ml/day) from the vials. Thus, even the smallest vial (7 ml) provided at least 20 days of attractant in the field.

Discussion

The total number of yellowjackets trapped in 2021 (6,010) declined dramatically from that in 2020 (36,547). Climatic conditions may have contributed to this decline and are discussed in Appendix I.

The heptyl crotonate and heptyl butyrate caught similar numbers of yellowjackets and species. Both attractants evaporate slowly, and the vials needed to be replaced every 3-4 weeks.

San Diego Zoo Safari Park 2019

The Park is a 728.4 ha animal sanctuary located within the San Pasqual Valley near Escondido, CA (33°05'50.80" N, 116°59'44.60" W, elev. 137 m). It is largely surrounded by coastal sage scrub and chaparral which are ideal habitats for the western yellowjacket, *V. pensylvanica*. Several sites within the park have had serious problems with foraging yellowjackets and park personnel were trapping them with disposable traps in 2019. A cooperative project with park personnel was established in September 2019 in which park personnel set up traps and bait stations in the park and monitored them. The experimental baits were prepared at UC Riverside and transported to the park. The trapped specimens were collected and sent to UCR to be counted and identified.

All of the sites were baited because of the large number of yellowjackets, and the Park personnel and animals were being stung. Each site was baited three times to dramatically reduce the number of yellowjackets.

Methods and Materials

Monitoring

Six sites at the park were trapped with Rescue Disposable Yellowjacket Traps (Fig. 3). The sites included the Condor Enclosure (CON), Bird Breeding Complex (BBC), Wings of the World (WOTW), Cheetah Breeding Compound (CBC), Burrowing Owl Site (BUR), and Forage Warehouse (FOR). The traps were hung under trees and bushes about 0.5-1.5 m off the ground. Instead of adding pure tap water to the trap's collection bag, a solution of propylene glycol coolant was made with water (1:2 dilution) and added to the collection bag. The solution is effective in killing and preserving the insects. The contents of the bag were removed, and the excess fluid drained. The contents were placed into 1-gal plastic zip lock bags and shipped to UC Riverside where the number and species of yellowjackets were counted

Efficacy Trials

Baits were prepared at UC Riverside with fipronil (Termidor SC, BASF Corp., Research Triangle Park, NC), 400 ml chicken juice, 400 ml water, and 66.6 g PAA crystals so that the final concentration of fipronil was 0.025%. The dinotefuran baits were prepared with 200 ml

chicken juice, 200 ml water, and 33.3 g PAA crystals so that final concentrations of dinotefuran were 0.05, 0.025, and 0.0125% AI.

Salsa cups and lids were weighed and ≈ 30 g of bait was placed in each cup. The entire bait cups (cups + lids + bait) were weighed again. The baits were held on ice packs and transported to the animal park.

The cups of bait were placed inside Havahart® animal traps on the ground at each of the sites. The salsa cups with bait were collected after 24-72 hours and the cups were covered with lids. The baits were refrigerated until they were returned to UC Riverside. The cups were weighed, and the amount of bait removed was determined. Another set of baits was held in UCR-style bait stations covered with fine screen to prevent yellowjackets from foraging on the baits. These served as the evaporation control.

These bait cups were retrieved, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The 0.025% fipronil PAA baits were placed out from 9/11/2019 to 9/13/2019, 10/1/2019 to 10/3/2019, and 11/15/2019 to 11/18/2019 at the CON site. The PAA baits containing 0.05, 0.025, and 0.0125% dinotefuran were placed out on 9/24/2019 to 9/26/2019 at the remaining five sites. The five sites were baited with 0.025% dinotefuran on 10/15/2019 and 11/14/2019.

Some of the sites had sufficient monitoring traps to permit the data to be analyzed with a Wilcoxon signed-rank test. Monitoring traps from sites treated with the same concentration of bait were pooled.

Results

Monitoring

The trapping period was reduced from 14 days to 4-7 days to ensure that the sites could be baited before the end of September. The total number of *V. pensylvanica* trapped in 2019 were 21,427. A few *V. atropilosa* and *suplhurea* were also collected.

Efficacy Trial 1

Condor Breeding Site **(CON)-**The baits lost 47.7% of the weight due to evaporation of water during the 48-hour exposure beginning on 9/11/2019. The yellowjackets removed a total of 316.6 g of bait (37.5% of the total) from 30 cups placed out at the site. All 30 cups had detectable feeding during the first baiting period. The 0.025% fipronil PAA baits provided a 53.2% (W = 36, Z = 2.49, **P** = 0.01), 53.9% (W = 24, Z = 1.65, **P** = 0.09), and 44.4% (W = 36, Z = 2.49, **P** = 0.01) reduction in the average number of yellowjackets/trap/day (YJ/T/D) at day 3, 10, and 17 at the condor rearing facility.

During the second baiting period (10/1/2019 to 10/3/2019), the baits lost 35.1% of their weight due to evaporation. A total of 178.6 g of bait were retrieved. There was no detectable removal of bait in 10 of the 30 cups. The second baiting on 10/1/2019 to 10/3/2019 with 0.025% fipronil PAA resulted in a 57.6% (W = 30, Z = 2.07, **P** = 0.04), 57.8 % (W = -10, Z = -0.74, **P** = 0.46), 80.9% (W = 36, Z = 2.49, **P** = 0.013), and 93.1% (W = 36, Z = 2.49, **P** = 0.01) reduction in trap counts on day 10, 18, 25, and 40, respectively.

The third baiting with 0.025% fipronil began on 11/14/2019 and ended on 11/18/2019. All the sites across the park experienced > 90% reduction in the average number of YJ/T/D on

12/5/2019. Monitoring was suspended and the data were not analyzed because of the lack of yellowjacket activity at the Park.

***Wings of the World* (WOTW) –** At the WOTW site, the 0.05% dinotefuran bait lost 19.0% of its weight due to evaporation during the 48-hour exposure period. After adjusting for water loss, a total of 23.2 g of bait were removed from the 6 bait cups (about 12.3% of the total available bait). The 0.5% dinotefuran bait provided a 55.3% reduction (4.25 YJ/T/D) at day 4, but the trap counts returned to 17.7 and 11.78 YJ/T/D by day 11 and 18, respectively.

The WOTW site was baited again with 0.025% dinotefuran from 10/15/2019 to 10/17/2019. The baits in the evaporative checks lost about 46.3% of their weight. When adjusted for weight loss, the yellowjackets removed 23.06 g of bait (≈ 18.9% of the total). At day 5 there was a 16.7% reduction in the average number of YJ/T/D, but the numbers were 2.3-fold higher than before baiting at day 27.

A third baiting with 0.025% dinotefuran was performed on 11/15/2019 to 11/18/2019. All the sites across the park experienced > 90% reduction in the average number of YJ/T/D on 12/5/2019. The monitoring was suspended, and the data were not analyzed because of the lack of yellowjacket activity at the Park.

***Bird Breeding Complex* (BBC), *Forage Warehouse* (FOR) –** Both sites were baited with 0.025% dinotefuran PAA bait from 9/24/2019 to 9/26/2019. In the evaporation control, the bait lost 22.1% of its weight in 48 hours. After adjusting for water loss, the yellowjackets retrieved 78.4 and 73.4 g of bait from FOR and BBC, respectively.

The 0.025% dinotefuran bait provided 65.3, 18.4, and 44.1% reduction in the average number of YJ/T/D at days 4, 7, and 14, respectively. The reductions were significant at day 4 (W = 95, Z = 2.97, **P** = 0.001) and day 14 (W = 92, Z = 2.60, **P** = 0.009).

The second baiting with 0.025% dinotefuran occurred from 10/15/2019 to 10/18/2019. The control baits lost 46.3% of their weight during the 72-hour exposure. After adjusting for water loss, the yellowjackets retrieved 70.5 and 42.7 g of bait from BBC and FOR, respectively. The baiting provided 0.0%, 37.7%, and 0.0% reductions in the average number of YJ/T/D at day 4, 11, and 26, respectively.

The third baiting with 0.025% dinotefuran began on 11/14/2019 and ended on 11/18/2019. All the sites across the park experienced > 90% reduction in the average number of YJ/T/D on 12/5/2019. The monitoring was suspended, and the data were not analyzed because of the lack of yellowjacket activity in the Park.

***Cheetah Breeding Compound* (CBC), *Burrowing Owl Site* (BUR) –** Both sites were baited with –0.0125% dinotefuran PAA baits on 9/24/2019. The evaporative controls lost 19.3% of their weight during the 48-hour exposure. After adjusting for water loss, the yellowjackets took 30.3 and 69.7 g of bait from BUR and CBC sites, respectively.

There was a significant 35.7% reduction in the average YJ/T/D at day 4 (W=21, n = 6, **P** =0.03), but the reductions were not significantly lower at days 11 and 18. There was an average of 5.19 YJ/T/D at day 18.

The second baiting with 0.0125% occurred on 10/15/2021. The evaporation controls lost 46.3% of their weight during the 72-hour exposure. When adjusted for water loss, the yellowjackets took 4.4 and 64.3 g of bait from BUR and CBC, respectively.

There were no reductions in the average YJ/T/D after the second baiting with 5.19 YJ/T/D before baiting and 8.50, 6.58, and 5.57 YJ/T/D 4, 11, and 26 days after baiting, respectively.

The third baiting with 0.0125% dinotefuran occurred on 11/14/2019. All the sites across the park experienced > 90% reduction in the average YJ/T/D on 12/5/2019. The monitoring was suspended, data was not analyzed because of the lack of yellowjacket activity in the Park.

Discussion

The monitoring traps at all the sites revealed very large numbers of yellowjacket foragers. The pre-bait monitors trapped a combined total of 4,112 yellowjackets (Avg. 128.5/trap, n = 32) with almost all being *V. pensylvanica*. The greatest numbers of yellowjackets trapped was at the CON site with > 47 yellowjackets/trap/day.

Even though the 0.025% fipronil baits provided a steady decline of yellowjackets trapped, the numbers being collected in the monitoring traps were still well above 10 YJ/T/D threshold. Rust et al. (2017) reported that 0.025% fipronil provided > 75% reductions in the number of yellowjackets trapped. About 25-50% of the bait (1:3 chicken juice:water dilution) applied was removed and the baits were simply not attractive enough to compete with the food sources at the CON site.

The dinotefuran baits provided inconsistent results. The amount of bait removed varied between concentrations and the sites baited. Only 13% of 0.05% bait was taken compared with 17.3% at 0.0125% and 28.2% at 0.025%. In the second baiting, only 13.1% of the 0.025% dinotefuran bait was taken. The baits were simply not attractive enough to lure the yellowjackets away from food sources being placed out at each site.

San Diego Zoo Safari Park 2020

Methods and Materials

Monitoring

The monitoring period was expanded to 3 weeks to reduce the amount of labor and traps being installed. Additional traps were installed so that each site had a minimum of 6 disposable traps. Monitoring traps were initially installed on 5/7/2020 and the last traps were collected 10/7/2020.

Bait Stations

In the first and second bait trials, the bait cups were placed inside Havahart® animal traps on the ground at each of the sites. In the subsequent bait trials, UCR circular baits stations were used to prevent ants from feeding on the baits. The bait stations were hung from a bush or tree with a wire and a Perky-Pet® ANT GUARD® (Woodstream Corp., Lititz, PA) was used to prevent ants from feeding on the baits (Fig. 5). The bait stations were placed next to an existing monitoring station.

Efficacy Trials

The first baiting trial began on 7/29/2020 to 7/30/2020 with three sites being baited. The baits included 0.05% fluralaner (98% technical grade, BOC Sciences Shirley, NJ), 0.05%

selamectin + sarolaner (Revolution Plus), and 0.05% fluralaner (Bravecto® 250 mg). The toxicants were mixed into 80 ml of chicken juice and 420 g of finely minced chicken. The mixture was stirred and thoroughly mixed providing 0.05% baits.

A second baiting trial was initiated on 8/18/2020 at WOTW. The 0.05% fluralaner bait was prepared with 430 g of minced chicken, 70 ml of chicken juice and 1 vial of Bravecto. The mixture was stirred and refrigerated overnight.

The third baiting trial was initiated on 9/3/2020 at CBC, CON, FOR, and BUR. The 0.048% selamectin + 0.008% sarolaner was prepared with 8 tubes of Revolution Plus, 430 g of minced chicken, and 70 ml of chicken juice. The 0.05% fluralaner bait was prepared with 430 g of minced chicken, 70 ml of chicken juice and 1 vial of Bravecto or 250 mg of technical fluralaner. The 0.05% fipronil bait was prepared with 430 g of minced chicken, 70 ml of chicken juice and 2.6 ml of Termidor SC. The mixtures were stirred and refrigerated overnight.

Salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. The entire cup (cup + lid + bait) was weighed again. Cups and bait were refrigerated and transported to the park on ice packs.

These bait cups were retrieved, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

Results

Monitoring

The total number of *V. pensylvanica* queens and workers trapped was 17 and 41,595, respectively (Fig. 33). BBC was the only site not baited during 2020 and it served as an untreated seasonal control.

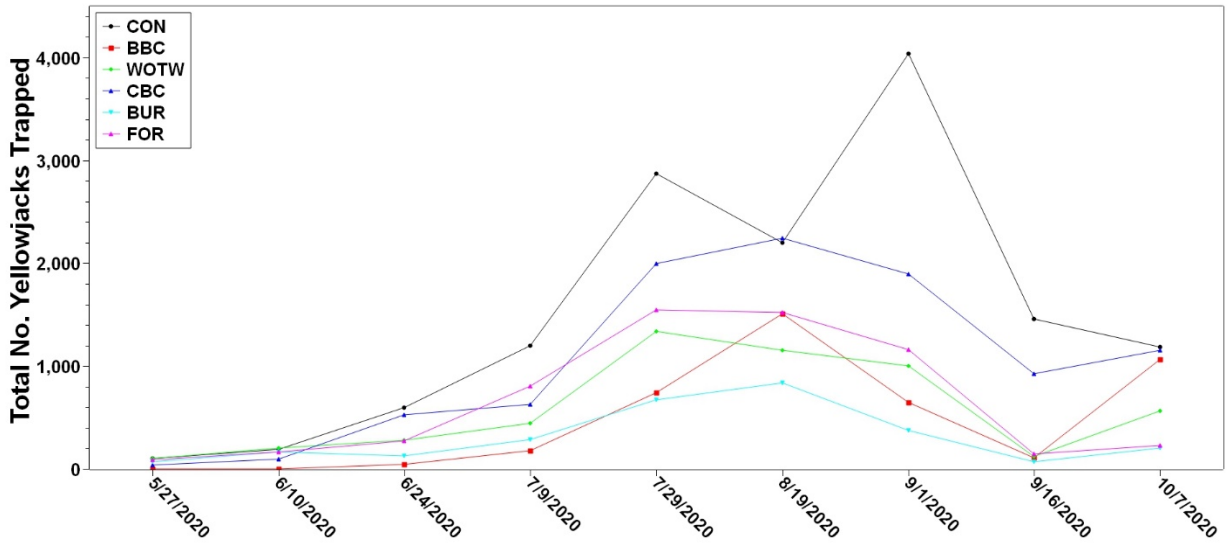

Fig. 33. The total number of yellowjackets trapped at each of the 6 sites in the Park during 2020.

Efficacy Trial #1

The evaporative controls of the 0.05% fluralaner (tech.), 0.05% fluralaner (Bravecto), and the 0.048% selamectin + 0.0048% sarolaner lost 81.0, 56.6, and 67.4% of their weight in 48 hours. After adjusting for water loss, the yellowjackets removed 135.4, 72.8, and 44.8 g of the 0.05% fluralaner (tech.), 0.05% fluralaner (Bravecto), and the 0.048% selamectin + 0.008% sarolaner, respectively. Some of the ant baits were covered with ants.

None of the baits provided significant reductions in the average YJ/T/D (Table 30). The average YJ/T/D increased at the untreated site BBC. The numbers remained above the critical threshold after 32 days at CON and FOR and these sites were baited again.

Table 30. The efficacy of 0.05% fluralaner and 0.048% selamectin + 0.008% sarolaner in minced chicken.

Site	Bait (formulation)	Bait Taken (g)	Average YJ/T/D (% reduction)		
			Pre-baiting	Day 19	Day 32
CON	0.05% fluralaner (tech.)	135.4	11.97	10.01 (16.4%)	25.88 (0.0%)
FOR	0.05% fluralaner (Bravecto)	72.8	12.88	12.65 (1.75%)	14.87 (0.0%)
CBC	0.048% selamectin + 0.008% sarolaner (Rev. Plus)	44.8	8.33	9.35 (0.0%)	7.9 (5.1%)
BBC	Untreated		3.72	7.20 (0.0%)	4.61 (0.0%)

Efficacy Trial #2

The evaporation controls lost 53.3% of their weight during the 48-hour exposure. After adjusting for the water loss, yellowjackets took 41.3 g of the 0.05% fluralaner bait. There were lots of ants in the bait cups and the Havahart traps.

There were significant reductions in the average YJ/T/D at days 28 (W= 55, Z = 2.78, **P** = 0.006) and 49 (W = -55, Z = -2.83, **P** = 0.005, Table 31). The average YJ/T/D declined at BBC (untreated control) at day 28 but increased by day 49.

Table 31. The efficacy of 0.05% fluralaner bait in minced chicken at WOTW site.

Site	Bait (formulation)	Average YJ/T/D (% reduction)			
		Pre-baiting	Day 14	Day 28	Day 49
WOTW	0.05% fluralaner	5.50	7.17 (0.0%)	0.87 (84.1%)	2.69 (51.0%)
BBC	Untreated	7.20	4.61 (0.0%)	0.79 (82.3%)	5.06 (0.0%)

Efficacy Trial #3

A third baiting trial was conducted from 9/1/2020 to 9/3/2020. The baits were applied in UCR-style circular bait stations suspended from the ground to prevent ants from feeding on the baits. All the baits were formulated in minced chicken. The minced chicken lost 43.9% of its weight in 48 hours. After adjusting for water loss, the yellowjackets removed 89.0, 86.3, and 92.4 g of selamectin + sarolaner, fluralaner (technical), and fluralaner (Bravecto) baits,

respectively. The 0.05% fipronil baits gained weight and it is likely that the sprinkler system had sprayed the baits (Table 32).

The 0.05% fluralaner bait (tech.) provided significant reductions in the average YJ/T/D at days 13 (W = 78, Z = 3.04, **P** = 0.002) and 34 W = 76, Z = 2.96, **P** = 0.003). The 0.05% fluralaner formulated with Bravecto also provided significant reductions at days 13 (W = 21, n = 6, **P** = 0.03) and 34 (W = 21, n = 6, **P** = 0.03). The selamectin + sarolaner provided significant reductions in the number of yellowjackets at days 13 (W= 43, Z = 2.17, **P** = 0.03) and 34 (W = 47, Z = 2.37, **P** = 0.02). There was a significant decrease in the average YJ/T/D in the untreated control at day 13 (W = 51, Z = 2.57, **P** = 0.006), but the numbers returned to pre-baiting levels at day 34.

Table 32. The third baiting trial was initiated on 9/1/2020 at four sites.

Site	Bait (formulation)	Bait Taken (g)	Average YJ/T/D (% reduction)		
			Pre-baiting	Day 13	Day 34
CON	0.05% fluralaner (tech.)	86.3	25.88	8.11 (68.7%)	5.64 (78.2%)
FOR	0.05% fluralaner (Bravecto)	92.4	14.87	1.88 (87.3%)	1.80 (87.9%)
CBC	0.048% selamectin + 0.008% sarolaner (Rev. Plus)	89.0	5.06	3.87 (51.1%)	5.49 (30.6%)
BUR	0.05% fipronil	NA	4.83	0.88 (81.7%)	1.60 (67.0%)
BBC	Untreated		4.61	0.79 (82.9%)	5.06 (0.05%)

Discussion

The first and second baiting trials failed to significantly reduce the numbers of yellowjackets trapped. The third baiting with 0.05% fluralaner did provide significant reductions in the number of yellowjackets at days 13 and 34. In the untreated control site, the numbers of yellowjackets increased during the first baiting period, dropped on 9/16/2020, and then returned to levels in August. The reason for this dramatic drop in the number of yellowjackets at BBC during the first 14 days of September is unknown. It is the only time in the study where we observed this dramatic of a decline followed by a resurgence in the untreated sites.

Less than 30% of the baits applied were taken by yellowjackets. The baits simply failed to attract enough yellowjackets away from other competitive food sources being provided.

San Diego Zoo Safari Park 2021

Methods and Materials

Monitoring

The monitoring was initiated on 5/25/2021 and the last traps were collected on 11/11/2021. The CON and FOR sites were baited on 8/17/2021 and 9/29/2021. CBC and BCC are more than 2,100 m away from the CON and FOR sites and served as the untreated control sites.

Efficacy Trial #1

The 0.05% fluralaner bait was prepared with 250 ml of chicken juice, 250 ml of water, 33.33 g PAA crystals, and 1 tube of Bravecto (250 mg fluralaner). The mixture was stirred and conditioned in the refrigerator overnight.

Salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. The entire cup (cup + lid + bait) was weighed again. Cups and bait were refrigerated and transported to the park on ice packs.

These bait cups were retrieved, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

Efficacy Trial # 2

The 0.05% fluralaner bait was prepared with 250 ml of chicken juice, 250 ml of water, 33.33 g PAA crystals, and 1 tube of Bravecto (250 mg fluralaner). The 0.025% fluralaner bait was prepared with the same ingredients, except only 0.5 tubes of Bravecto (125 mg fluralaner) were used. Both mixtures were stirred and conditioned in the refrigerator overnight.

Salsa cups and lids were weighed and ≈ 30 g of bait was added to each cup. The entire cup (cup + lid + bait) was weighed again. Cups and bait were refrigerated and transported to the park on ice packs.

These bait cups were retrieved, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The number of yellowjackets trapped before and after baiting was compared with a Wilcoxon signed-rank test.

Results

Monitoring

A few *V. atropilosa* and *V. sulphurea* were taken in the first monitoring event, but all the remaining yellowjackets were *V. pensylvanica*. A total of 14,914 yellowjackets were trapped during the 2021 season. The number of YJ/T/D at CBC and BBC peaked on 9/29/2021 (Fig. 34). The numbers never reached the treatment threshold of 10 YJ/T/D at any of the sites.

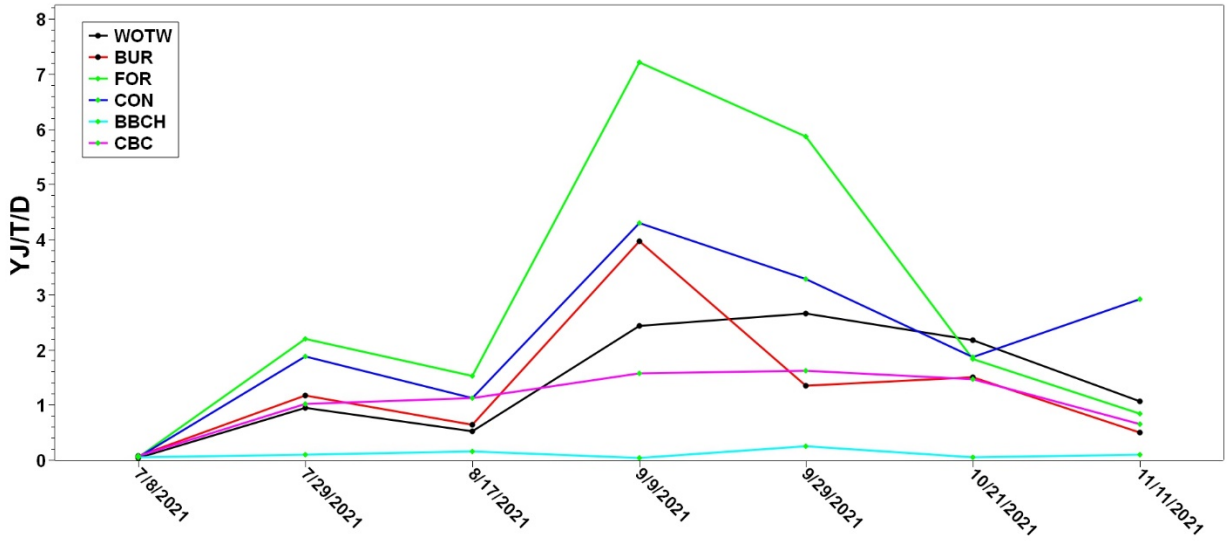

Fig. 34. The average number of yellowjackets/trap/day collected at the Park in 2021.

Efficacy Trial #1

The 0.05% fluralaner PAA bait lost 57.3% of their weight in the evaporation control during the 48-hour exposure. After compensating for the water loss, the yellowjackets removed 91.2 g of bait.

The average YJ/T/D prior to baiting was 1.12. It increased to 4.30 YJ/T/D at day 21 and 3.28 YJ/T/D at day 41.

Efficacy Trial # 2

Even though the trap counts were well below the threshold of 10 YJ/T/D two sites were baited because of large numbers of yellowjackets and complaints. The baits lost 17.5 % of their weight in the evaporative controls during the 24-hour period. When the bait taken was adjusted for water loss, the yellowjackets removed 80.5 and 30.3 g of 0.05% fluralaner and 0.025% fluralaner, respectively (Table 33).

There were no significant declines in number of yellowjackets trapped in the site baited with 0.05% fluralaner bait. At day 42, there was a significant reduction in the numbers trapped at FOR with the 0.025% bait (W = 21, n = 6, **P** = 0.03).

Table 33. The efficacy of 0.025% and 0.05% fluralaner in PAA crystals.

Site	Bait	Bait Taken (g)	Average YJ/T/D (% reduction)		
			Pre-baiting	Day 21	Day 42
CON	0.05% fluralaner	80.5	3.28	1.87 (43.1%)	2.92 (10.9%)
FOR	0.025% fluralaner	30.3	5.86	1.83 (68.7%)	0.84 (85.6%)
CBC	Untreated		1.62	1.46 (9.9%)	0.65 (59.7%)
BBC	Untreated		0.25	0.05 (80.5%)	0.1 (59.4%)

Discussion

The winter and spring rainfall was well below normal and may have impacted the development of the yellowjacket colonies (See Appendix I). The number of YJ/T/D was below the threshold of 10 YJ/T/D at each of the sites throughout the 2021 season. The numbers of yellowjackets at both sites were still a problem and baiting trials were conducted. The amounts of bait taken were less than 100 g during both trials. The numbers of yellowjackets trapped at the two baited sites were not significantly lower after baiting.

Ronald W. Caspers Wilderness Park 2020

The Ronald W. Caspers Wilderness Park consists of 3,238 ha (8,000 acres) of protected wilderness preserve in the coastal Santa Ana Mountains (33°31′57.76″ N, 177°33′04.31″ W, elev. 115 m). The Park has native Coastal Live Oaks and stands of California Sycamore. It provides camping, picnicking, hiking, and equestrian activities.

The numbers of yellowjackets created a severe situation during the summer of 2020 when we were contacted. The monitoring traps were checked weekly so that the site could be baited in September.

Methods and Materials

Monitoring

Fifty UCR-style traps were installed between 9/23/2020 and 9/24/2020 along the main roadway in the park (Fig. 35). The monitoring sites were separated by at least 100 m. Traps were serviced once a week with the last traps being collected 10/26/2020.

Efficacy Trial # 1

The baits were prepared with minced chicken and PAA gels and three different toxicants, 0.024% selamectin + 0.004% sarolaner, 0.025% sodium selenate, and 0.025% fipronil. The PAA baits consisted of 33.3 g of PAA crystals, 100 ml of chicken juice, and 300 ml water (1:3 dilution) and either 125 mg of sodium selenate or 125 mg of fipronil (Termidor SC). The hydrogels were allowed to condition in the refrigerator overnight. The meat bait was prepared by mixing 2 tubes Revolution Plus into the 80 ml of chicken juice and 420 g of finely minced chicken. The baits were stored in the refrigerator.

Salsa cups and lids were weighed, and ≈ 30 g of gel or minced chicken were added to each cup. Then the entire bait cup (cup+ bait + lid) was weighed. The baits were transported to the field on ice packets.

Three UCR bait stations were placed at each of three sites associated with a monitoring location with high yellowjacket trap counts. The selamectin + sarolaner bait stations were hung at Sites 25, 26, and 27. The sodium selenate baits were hung at Sites 34, 36, and 37. The fluaralaner baits were hung at Sites 2, 5, and 6.

After 9 days, the baits were retrieved and covered with tight lids. The cups were placed in on ice packs, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The yellowjacket trapping data before and after baiting was analyzed with a Wilcoxon signed-rank test or a paired *t*-test.

Results

Monitoring

The 50 UCR-style traps collected 28,262 *V. pensylvanica* in 7 days with 49 of sites with > 10 YJ/T/D on 10/1/2020.

Efficacy Trial # 1

The evaporation controls of minced chicken and the PAA gels lost 60.9% and 77.1% of their weight in 4 days, respectively. After adjusting for the water loss, the yellowjackets took 412.9 g of 0.024% selamectin + 0.004% sarolaner bait (80.8% of the total), 446.1 g of 0.025% sodium selenate (86.0% of the total), and 352.1 g of 0.025% fipronil (67.0% of the total).

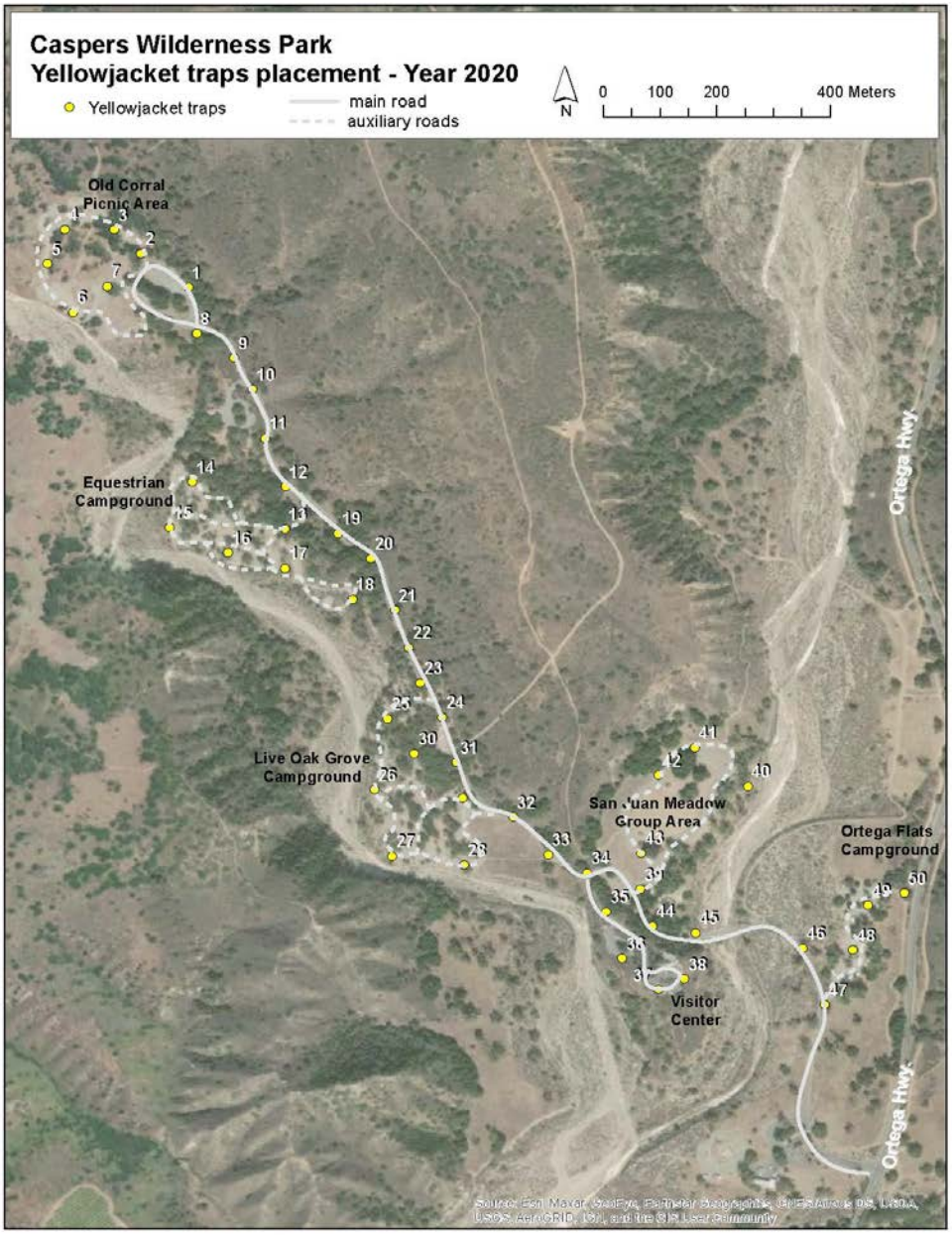

Fig. 35. Yellowjacket trap placement at Ronald W. Caspers Wilderness Park.

The selamectin + sarlonaer bait provided significant reductions at days 7 (W = 55, Z =2.78, **P** = 0.005, Table 34), 14 (W = 55, Z = 2.78, **P** = 0.005), and 21 (W = 55, Z = 2.78, **P** = 0.005). The sodium selenate provided significant reductions at days 7 (W = 36, Z =2.49, **P** = 0.013), 14 (W = 36, Z = 2.49, **P** = 0.013), and 21 (W = 36, Z = 2.49, **P** = 0.013). The 0.025% fipronil provided a significant reduction in the number of yellowjackets at 7 days (W = 36, Z =2.49, **P** = 0.013), 14 days (W = 36, Z = 2.49, **P** = 0.013), and 21 days (W = 36, Z = 2.49, **P** = 0.013). There was a significant reduction in the number of yellowjackets trapped at the 5 untreated monitors at days 14 (T = 3.45, df = 4, **P** = 0.026) and 21 (T = 3.72, df = 4, **P** = 0.020).

Table. 34. The efficacy of 0.024 % selamectin + 0.004% sarolaner, 0.025% sodium selenate, and 0.025% fipronil baits.

Toxicant	Bait Taken (g)	Monitoring Sites	Average YJ/T/D (% reduction)			
			Pre-baiting	7 days	14 days	21 days
selamectin + sarolaner	412.9	10	76.66	1.00 (98.7%)	0.54 (99.3%)	0.14 (99.8%)
sodium selenate	489.6	8	75.84	10.05 (86.7%)	2.34 (96.9%)	0.64 (99.2%)
fipronil	455.8	8	107.34	1.00 (99.1%)	0.30 (99.7%)	0.27 (99.8%)
Untreated		5	18.57	9.51 (48.8%)	1.66 (97.1%)	0.46 (97.5%)

Discussion

The baiting occurred late in the season after we were alerted by Park personnel of complaints of large populations of yellowjackets in mid-September. All three baits were readily accepted by the yellowjackets and provided significant reductions in the number of yellowjackets trapped in the park 7 days after baiting and before the untreated controls significantly declined at 14 days.

Ronald W. Caspers Wilderness Park 2021
Tree of Life Nursery 2021

The exact same sites at the Ronald W. Caspers Wilderness Park were monitored again in 2021. Additional monitoring sites were established at the Tree of Life Nursery which was about 1,000 m west of the Park (see Tree of Life Nursery Report). These sites served as a seasonal control so that the entire Park could be baited.

Methods and Materials

Monitoring

Fifty UCR-style traps were installed on 6/15/2021 along the main roadway at the same sites in the park as in 2020 (Fig. 36). The traps were checked every two weeks until 11/15/2021. Ten UCR traps were installed along a transect at the Tree of Life Nursery to serve as seasonal checks. The traps were installed on 6/15/201 and checked every two weeks with the last collection on 11/15/2021.

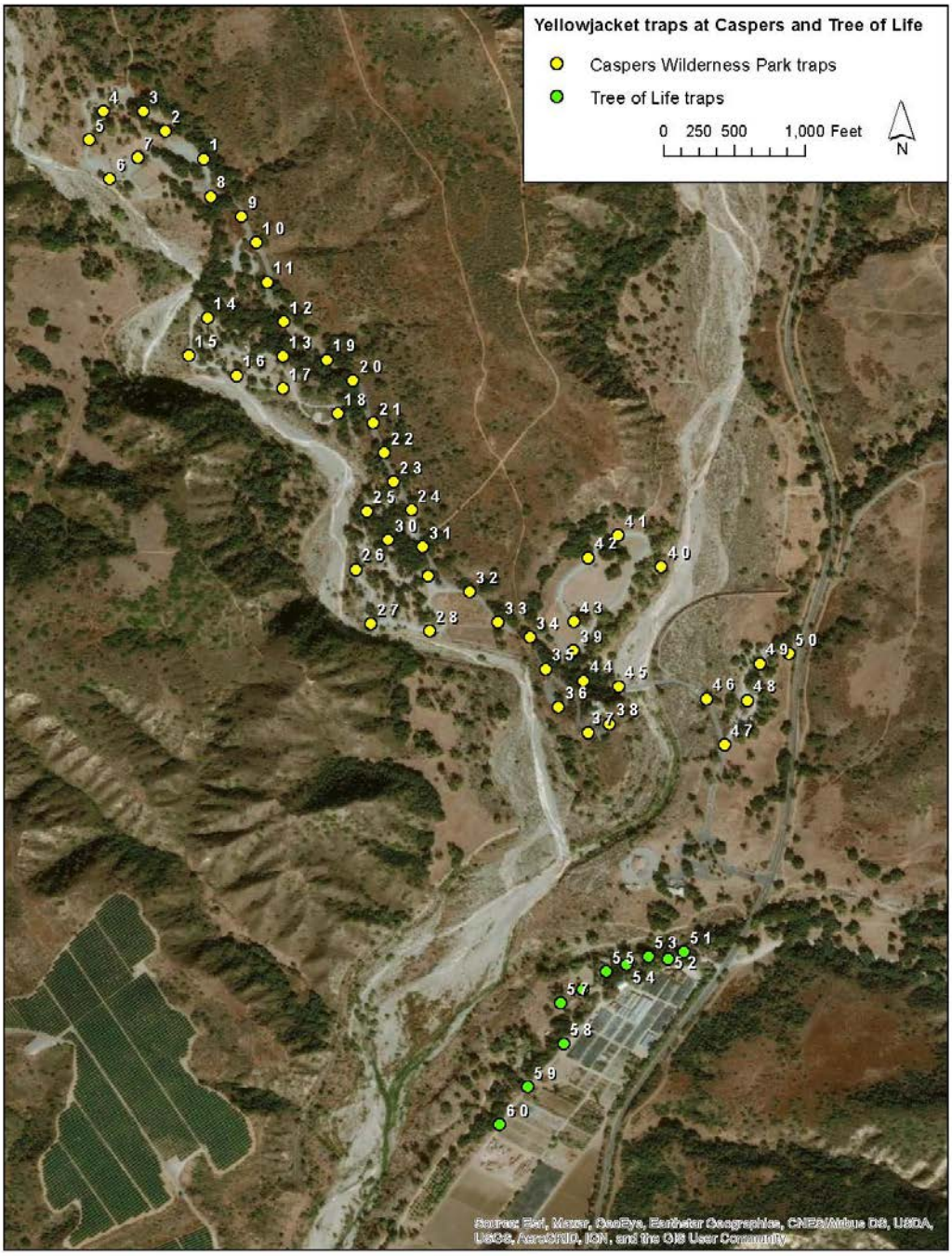

Fig. 36. Monitoring trap locations at Ronald W. Caspers Park (yellow dots) and the Tree of Life Nursery (green dots).

Efficacy Trial # 1

The 0.05% fluralaner bait was prepared with 250 ml of chicken juice, 250 ml of deionized water, 33.33 g of PAA crystals, and 1 tube of Bravecto (250 mg fluralaner). The 0.096% selamectin + 0.016% sarolaner bait was prepared with 250 ml of chicken juice, 250 ml

of deionized water, 33.33 g of PAA crystals, and 8 tubes of Revolution Plus. The baits were allowed to condition in the refrigerator overnight.

Salsa cups and lids were weighed, and ≈ 30 g of bait were added to each cup. Then the entire bait cup (cup+ bait + lid) was weighed. The baits were transported to the field on ice packets.

Four bait cups were placed in each of four UCR bait stations. The fluralaner bait stations were place at Sites 33, 36, 40, and 47. The selamectin + sarolaner baits were placed out at Sites 3, 10, 17, and 27. The baits went out on 8/11/2021 and were retrieved on 8/13/2021.

After 2 days, the baits were retrieved and covered with tight lids. The cups were placed in on ice packs, returned to the laboratory, and weighed. After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The yellowjacket trapping data before and after baiting was analyzed with a paired *t*-test.

Efficacy Trial # 2

The same baits were applied again on 9/20/2021 (see methods above).

Results

Monitoring

The only species of yellowjackets trapped were *V. pensylvanica*. A total of 15,344 yellowjackets were trapped over the summer and fall.

Efficacy Trial # 1

The evaporation controls for the fluralaner and selamectin + sarolaner baits lost 65.3 and 60.3%, respectively. After compensating for the water loss, the yellowjackets took 316.1 g of fluralaner bait (78.4% of the total) and 384.5 g of selamectin + sarolaner bait (95.1% of the total, Table 35). The selamectin + sarolaner provided significant reductions at days 9 (W=462, Z = 4.75, **P** < 0.001), 14 (W = 461, Z = 4.74, **P** < 0.001), and 28 (W = 462, Z = 4.15, **P** < 0.001). The fluralaner bait provided significant reductions at days 9 (W = 188, Z =3.77, **P** < 0.001), 14 (W = 184, Z = 3.69, **P** < 0.001). and 28 (W = 190, Z = 3.81, **P** < 0.001). The number of yellowjackets trapped in the untreated area at Tree of Life Nursery remained unchanged for 14 days after the baiting and then there was a significant reduction in the number of yellowjackets at 28 (W = 55, Z = 2.78, **P** = 0.006).

Table 35. The efficacy of 0.05% fluralaner and 0.096% selamectin + 0.016% sarolaner baits in PAA crystals at Ronald W. Caspers Wilderness Park.

Toxicant	Bait Taken (g)	Monitoring Sites	Average YJ/T/D (% reduction)			
			Pre-baiting	9-10 days	14-15 days	27-28 days
Selamectin + sarolaner	384.5	31	5.78	0.45 (92.2%)	0.71 (87.8%)	0.40 (93.1%)
Fluralaner	316.1	19	4.26	1.25 (78.4%)	1.35 (76.6%)	0.75 (87.0%)
Untreated[a]		10	8.19	11.18 (0.0%)	5.189(36.6%)	2.43 (70.3%)

[a] Trap counts from the Tree of Life Nursery.

Efficacy Trial # 2

The evaporation controls of the fluralaner and the selamectin + sarolaner baits lost 52.1% and 41.7% of their weight in 48 hours. After adjusting for water loss, the yellowjackets removed 148.5 g of fluralaner bait (35.6% of the total) and 213.7 g of selamectin + sarolaner bait (49.3% of the total, Table 36).

The selamectin + sarolaner provided significant reductions in the number of yellowjackets trapped at days 12 (W = 430, Z = 4.64, **P** < 0.001), 28 (W = 435, Z = 4.70, **P** < 0.001), 42 (W = 404, Z = 4.59, **P** < 0.001), and 56 (W= 406, Z = 4.62, **P** < 0.001). The fluralaner provided significant reductions of yellowjackets trapped at days 12 (W = 186, Z = 3.73, **P** < 0.001), 28 (W = 170, Z = 3.41, **P** < 0.001), 42 (W = 171, Z = 3.71, P < 0.001), and 56 (W = 190, Z = 3.81, **P** < 0.001). The trap counts in the untreated section of Tree of Life Nursery remained unchanged from 9/17/2021 until 10/18/2021. There were significant reductions at days 42 (W = 55, Z = 2.78, **P** = 0.002) and 56 (W = 55, Z = 2.78, **P** = 0.002).

Table 36. The efficacy of the second baiting with 0.05% fluralaner and 0.096% selamectin + 0.016% sarolaner baits in PAA crystals.

Toxicant	Bait Taken (g)	Monitoring Sites	Average YJ/T/D (% reduction)				
			Pre-baiting	12 days	28 days	42 days	56 days
Selamectin + sarolaner	213.7	31	0.46	0.11 (75.4%)	0.08 (83.1%)	0.04 (91.4%)	0.01 (97.0%)
Fluralaner	148.5	19	0.86	0.28 (67.4%)	0.30 (65.0%)	0.11 (87.0%)	0.03 (96.3%)
Untreated[a]		10	2.43	2.84 (0.0%)	1.85 (23.9%)	0.55 (77.4%)	0.19 (99.2%)

[a] Trap counts from the Tree of Life Nursery.

Discussion

Both the fluralaner and selamectin + sarolaner baits provided significant reductions in the number of yellowjackets trapped. The yellowjackets removed more of the selamectin + sarolaner than the fluralaner. The numbers of YJ/T/D were well below the treatment threshold at the time of the second baiting, but park personnel wanted an additional baiting before the end of the season. The second baiting also significantly reduced numbers for the next 28 days.

Tree of Life Nursery 2021

The Tree of Life Nursery is located along Rte. 74 about 6.4 km east of Rancho Mission Viejo, CA (33°31'50.53" N, 117°33'01.90"W, elev. 116 m). The nursery is adjacent to the Ronald W. Casper Wilderness Park. The area consists of native Coastal Live Oaks and stands of California Sycamore. The site was selected to serve as a seasonal control for the baiting studies in the nearby Ronald W. Caspers Wilderness Park.

Methods and Materials

Monitoring

Ten UCR-style traps were spread along a transect along the northeast edge of the nursery about 115 m apart (Fig. 31). The nearest monitoring traps in the Ronald W. Casper Wilderness Park was ≈ 381 m away. The traps were installed on 6/15/2021 and checked every 14 days. The last traps were collected on 11/15/2021.

Monitoring Attractants Choice Test

The attractiveness of heptyl crotonate (HC) and heptyl butyrate (HB) was tested from 8/9/2021 to 8/23/2021. Ten UCR traps were provisioned with HC vials and 10 UCR traps were provisioned with HB vials. One HC trap was placed about 2 m from a HB trap at each of the 10 locations. After 14 days, the traps and collection jars were returned to the laboratory and counted.

The number of yellowjackets trapped with each attractant was analyzed with a Wilcoxon signed-ranks test.

Results

Monitoring

The only species of yellowjackets trapped was *V. pensylvanica*. The number of yellowjackets steadily increased in July and reached a maximum in the first weeks of August. Only 3 sites in August had > 10 yellowjackets/trap/day during the study (Fig. 37).

Monitoring Attractants Choice Test

There were no significant differences in the number of yellowjackets trapped in either HC or HB traps (W=9, n = 10, **P** = 0.69). Both attractants were equally effective in luring yellowjackets to the traps.

Discussion

The site served as an excellent control for the baiting trials at Ronald W. Caspers Wilderness Park. The numbers of yellowjackets significantly declined in September and again in late October.

Heptyl butyrate and heptyl crotonate attracted similar numbers of *V. pensylvanica* during the study. Wagner and Reierson (1969) added heptyl crotonate to mirex baits and tripled the amount of bait removed by yellowjackets. The heptyl crotonate is acceptable substitute for heptyl butyrate.

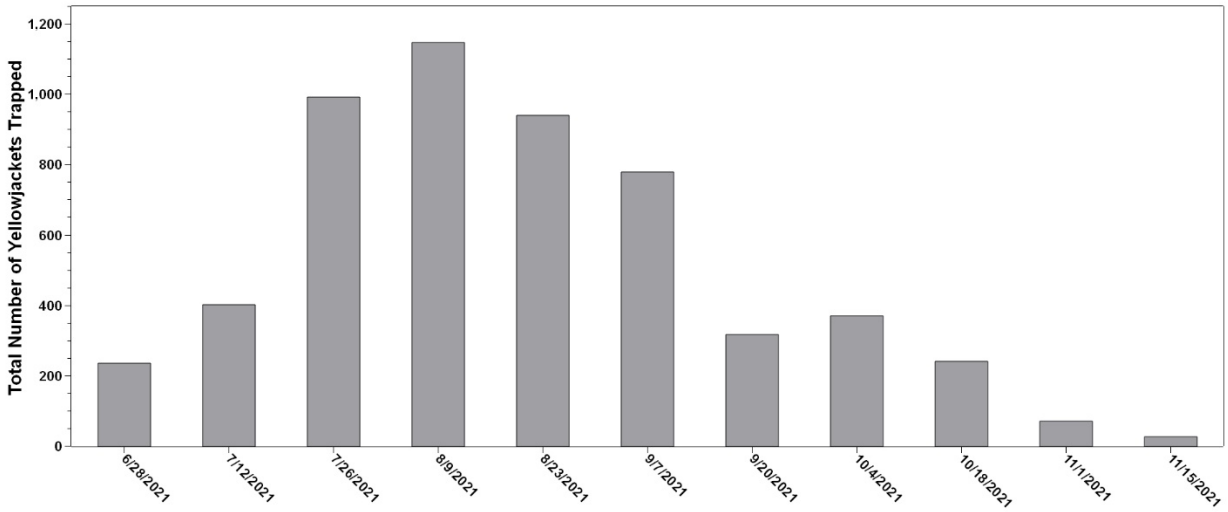

Fig. 37. The total number of yellowjackets trapped at Tree of Life Nursery in 2021.

University of California Riverside Campus Site 2019

The University of California Riverside campus is located next to the Box Springs Mountains (33°58'04", 117°19'34"W, elev. 356 m). The botanical gardens and the southern edge of the campus abuts native coastal sage scrub with mixed non-native grasses. A transect of monitoring traps was placed along a driveway and parking lot adjacent to a hillside of native scrub and grasses on the University of California Riverside campus.

Methods and Materials

Monitoring

The foraging activity of yellowjackets was measured using UCR-style traps. The monitoring traps were hung under trees and bushes about 0.5-1.5 m off the ground. A total of 10 traps were placed along a driveway and parking lot. The traps were hung about 30 m apart beginning on 5/13/2019. The traps were checked every 14 days and the number of queens and workers trapped was recorded for the entire summer and fall until 12/9/2019 (the last trap count).

Repellency Tests with Essential Oils

A solution of four essential oils (EOs) has been shown to be highly repellent to *V. pensylvanica* (Zhang et al. 2012). The EOs included natural clove oil, geranium oil, lemongrass oil, and rosemary oil. The objective was to determine if this solution would prevent yellowjackets from foraging on minced chicken. Clove oil natural (density 1.016 g/ml), geranium oil (0.91 g/ml), lemongrass oil (0.87 g/ml), and rosemary oil (0.906 g/ml) were purchased from LorAnn Oils, Inc. (Lansing, MI).

The solution was prepared with 1 ml of each of the EOs and 10 ml of acetone. Pieces of cheesecloth were cut into 19 by 19 cm squares. Each square was impregnated with the 14 ml of the solution and allowed to dry in the fume hood for about 1 hour.

To determine if the solution was repellent to yellowjacket foragers, two UCR-style bait stations were placed about 5 m apart at each of three sites on 11/25/2019. In one bait station, one cup with minced chicken was placed in the center of the cage. In the other station, a piece of cheesecloth impregnated with the EO solution was placed on the bottom of the cage and a cup with minced chicken was placed at the center of the cheesecloth. At the end of the day the piece of treated cheesecloth was removed from the cage, put into a plastic bag and stored in the refrigerator overnight. The test was repeated the next day with the same piece of treated cheesecloth.

On the second day, the amount of minced chicken removed was determined. Salsa cups and lids were weighed and ≈ 20 g of minced chicken was added to the cups. The entire cup (cup + lid + chicken) was weighed. To estimate the water loss from the minced chicken on the second day, an evaporation control station containing two cups of minced chicken was hung away from the other stations. The evaporation stations were removed at the end of the test. The cups were covered with lids and returned to the laboratory and weighed.

After adjusting for water loss of the baits, the amount of bait removed by the yellowjackets was determined (see Evaporation Control, page 6).

The data (the amount of chicken removed from EOs treated cloths and untreated cloths) were log transformed and compared with a paired *t*-test.

Results

Monitoring

The only species collected at the UCR site was the western yellowjacket, *V. pensylvanica*. A total of 11 queens were trapped during May and 5 queens were trapped in the first two weeks of June. The total number of workers trapped gradually increased during the summer and finally peaked on 10/28/2019 with 950 workers (Fig. 38). The numbers of yellowjackets trapped dramatically decreased after 11/12/2019. At the peak, the largest number of yellow jackets trapped was 6.8 YJ/T/D. The total number of workers trapped was 5,330.

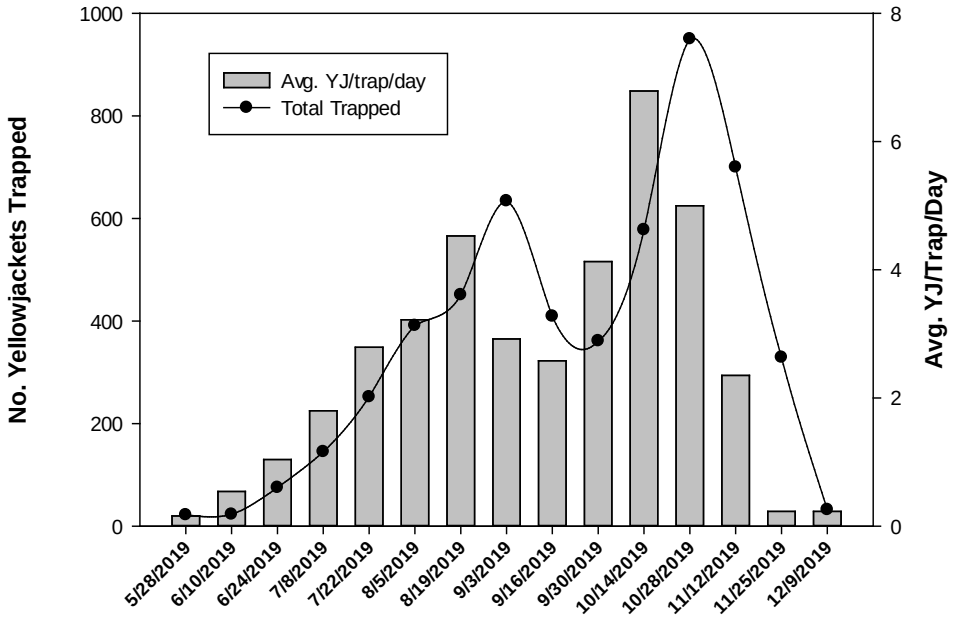

Fig. 38. The total number of yellowjackets trapped and the average number of yellowjackets/trap/day (YJ/T/D) at UCR during 2019.

Repellency Tests with Essential Oils

On the first day, the bait stations were checked hourly to determine if yellowjackets were foraging and removing the minced chicken. At 6 hours, the minced chicken on top of the treated cheesecloth was untouched whereas the yellowjackets had removed all the chicken from 2 of the 3 untreated bait stations. About half of the chicken was removed from the third untreated bait station.

On the second day, the same treated cheesecloths were tested again after being refrigerated overnight. The yellowjackets continued to forage on the minced chicken in the bait cups in the untreated control. No yellowjackets were observed in the bait stations with the treated cheesecloths during the second day but there was some removal. The minced chicken in the evaporative control lost 8.2% of its weight during the test. After adjusting for the water loss, a total of 5.1 g and 32.9 g of minced chicken was removed from the treated and untreated cloths, respectively. This difference was significantly different (t = -4.29, df = 2, **P** = 0.05, Table 37).

Table 37. The amount of minced chicken removed from the bait stations with and without the 1-day-old deposits of the EO solution.

	Treated Cheesecloth		No cheesecloth	
Sites	Taken (g)	% removed	Taken (g)	% removed
1	2.02	15.1	13.40	70.2
2	1.62	8.5	4.28	27.8
3	1.46	9.6	15.25	87.5

[a] 1-d-old deposits tested 11/26/2019 for 6.7 hours.

University of California Riverside
Campus Site 2020

Methods and Materials

Monitoring

The foraging activity of yellowjackets was monitored with 10 traps placed about 30 m apart. Monitoring began on 6/29/2020. The traps were checked every 14 days and the number of queens and workers trapped were recorded for the entire summer and fall. The last trap count was 11/2/2020.

Results

Monitoring

The only species collected at UCR was *V. pensylvanica*. No queens were trapped in 2020. The trap catches at UCR increased steadily over the summer and the total number of yellowjackets trapped peaked on 10/5/2020 (Fig. 39). The numbers dramatically declined in late October. The number of yellowjackets/trap/day never exceeded 10 YJ/T/D. A total of 6,190 yellowjackets were trapped during 2020.

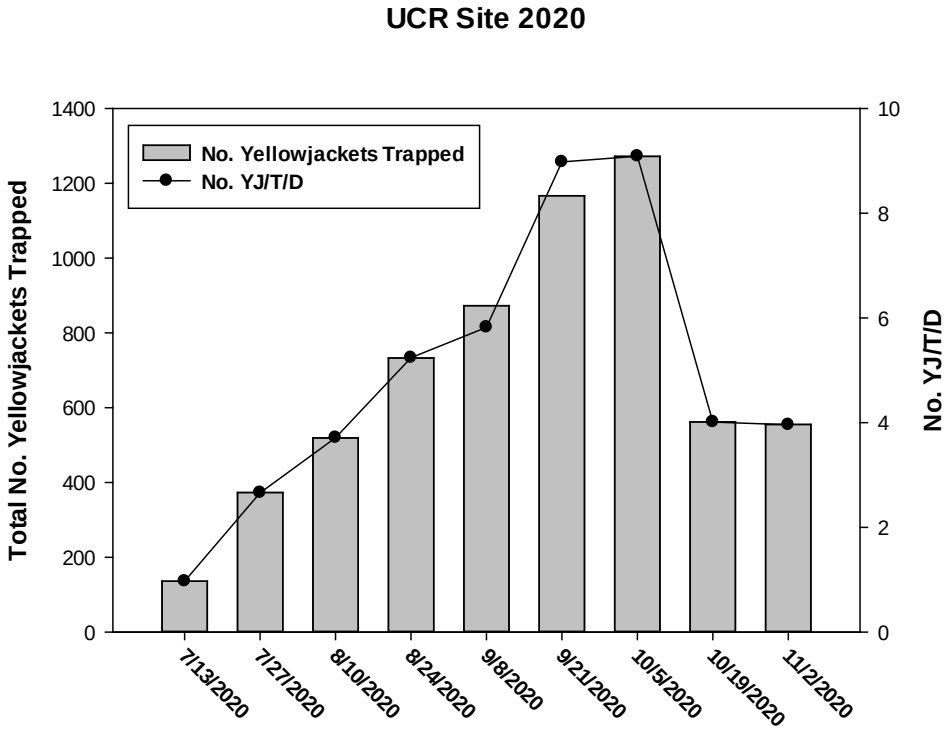

Fig. 39. The total number of yellowjackets trapped and the number of yellowjackets/trap/day (YJ/T/D) at the UCR site during 2020.

Discussion

The number of yellowjackets trapped never reached a critical threshold during 2020. The site was used a seasonal phenology.

University of California Riverside
Campus Site 2021

Methods and Materials

Monitoring

The foraging activity of yellowjackets was monitored with 10 traps placed about 30 m apart. Monitoring began on 6/14/2021. The traps were checked every14 days and the number of queens and workers trapped were recorded for the entire summer and fall. The last trap count was on 11/3/2021.

Results

Monitoring

The only species trapped was *V. pensylvanica*. No queens were trapped and a total of only 466 yellowjackets were trapped during the entire season (Fig. 40).

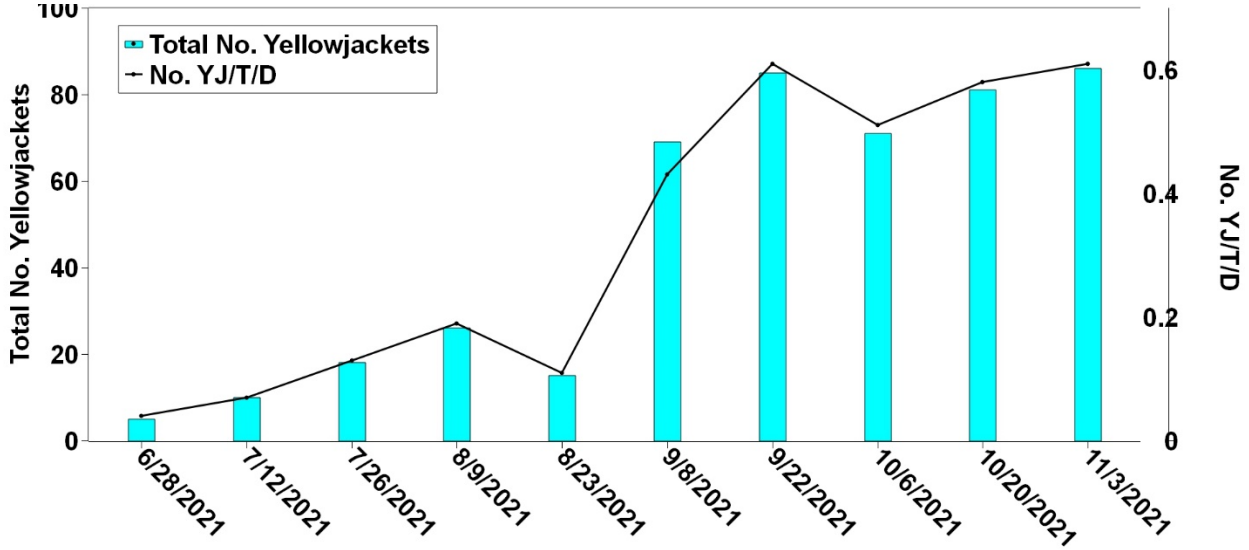

Fig. 40. The number of yellowjackets trapped and the yellowjackets/trap/day (YJ/T/D) for UCR site in 2021.

Discussion

The number of yellowjackets trapped at the UCR site never reached the action threshold of 10 YJ/T/D during 2021. The warmer temperatures and the lack of rain in 2021 negatively impacted the yellowjacket populations (Appendix I). The lack of rain probably contributed to a decline of vegetation and insects in the spring depriving queens and workers of food sources.

Acknowledgements. We would like to thank Mary Rust for assisting with the monitoring and baiting at Silent Valley RV Park. Linda Post and the Integrated Pest Management Team at the San Diego Zoo Safari Park did an outstanding job monitoring and baiting the site. The San Diego Zoo was extremely generous and assisted us in purchasing active ingredients for the study. Dr. Jocelyn Millar synthesized the heptyl crotonate.

References Cited

Daly D., M.E. Archer, P.C. Watts, M.P. Speed, M.R. Hughes, F.S. Barker, J. Jones, K. Odgaard, and S.J. Kemp. 2002. Polymorphic microsatellite loci for eusocial wasps (Hymenoptera: Vespidae). Mol. Ecol. Notes 2: 273–275

Grant C.D., C.J. Rogers, and T.H. Lauret. 1968. Control of ground nesting yellow jackets with toxic baits – a five-year testing program. J. Econ. Entomol. 61:1653–1656.

Hasegawa, E., and J. Takahashi. 2002. Microsatellite loci for genetic research in the hornet *Vespa mandarinia* and related species. Mol. Ecol. Notes 2:306–308.

Jones, O.R., and J. Wang. 2010. COLONY: a program for parentage and sibship inference from multilocus genotype data. Mol. Ecol. Resour. 10: 551–555.

Landolt, P. J., H. C. Reed, and D. J. Ellis. 2003. Trapping yellowjackets (Hymenoptera: Vespidae) with heptyl butyrate emitted from controlled-release dispensers. Fla. Entomol. 86: 323-328.

Reierson, D. A., M. K. Rust, and R. S. Vetter. 2008. Traps and protein bait to suppress populations of yellowjackets (Hymenoptera: Vespidae), pp. 267–274. In Proceedings, the Sixth International Conference on Urban Pests, 13–16 July 2008, Budapest. OOK-Press Kft., Hungary.

Reierson, D.A., and R.E. Wagner. 1975. Trapping yellowjackets with a new standard plastic wet trap. J. Econ. Entomol. 68: 395-398.

Rust M.K., D.-H. Choe, E. Wilson-Rankin, K. Campbell, J. Kabashima, and M. Dimson. 2017. Controlling yellow jackets with fipronil-based protein baits in urban recreational areas. Int. J. Pest Manage. 63: 234-241.

Rust M.K., D.A. Reierson, R.S. Vetter. 2010. Developing baits for the control of yellowjackets in California. Final Report 2010 for Structural Pest Control Board [Online]. Structural Pest Control Board, Grant No. 041–04, pp. 1–33. Available from: http://www.pestboard.ca.gov/howdoi/research/2009_yellowjacket.pdf (2010).

Simmons, E. S. 1991. Yellowjacket abatement in California parklands. Report to California Department of Parks and Recreation 17, Pest Management Series. Sacramento, USA. 39 pp.

Tay, J.-W., M.S. Hoddle, A. Mulchandani, and D.-H. Choe. 2017. Development of an alginate hydrogel to deliver aqueous bait for pest ant management. Pest Manage. Sci. 73: 2028–2038. DOI: 10.1002/ps.4616.

Thoren PA, Paxton RJ, Estoup A (1995) Unusually high frequency of (CT)n and (GT)n microsatellite loci in yellow-jacket wasp, *Vespula rufa* (L.) (Hymenoptera: Vespidae). Insect Mol. Biol. 4: 141–148

Wagner, R.E., and D.A. Reierson. 1969. Yellow jackets control by baiting. 1. Influence of toxicants and attractants on bait acceptance. J. Econ. Entomol. 62: 1192-1197.

Zhang, Q.-H, R.G. Schneidmiller, and D.R. Hoover. 2013. Essential oils and their compositions as spatial repellents for pestiferous social wasps. Pest Manage. Sci. 69: 542-552.

Summary

1). When conditioned in diluted chicken juice, the polyacrylamide hydrogels (PAA) were preferred over the alginate hydrogel beads. The PAA gels were easier to manipulate by the workers.

2). Minced chicken (Swanson's White Premium Chunk Chicken, Campbell Soup Co., Camden, NJ) and the liquid contents in the can were highly preferred by foraging yellowjackets. PAA gels conditioned in pure chicken juice or chicken juice diluted in water (1:1) were the most attractive bait base tested.

3). Heptyl crotonate and heptyl butyrate were attractive to 6 species of yellowjackets. There were no significant differences in the numbers of yellowjackets trapped. Heptyl crotonate is an acceptable substitute for heptyl butyrate.

4). Baits consisting of 0.025% fipronil in PAA or minced chicken were effective in reducing the number of yellowjackets trapped at most sites. However, dense populations of yellowjackets and considerable amounts of competitive foods at the San Diego Zoo Animal Safari reduced its effectiveness.

5). Baits containing dinotefuran and clothianidin failed to provide adequate control. Both toxicants probably killed workers too quickly and lower concentrations of each failed to reduce the number of foragers.

6). Baits containing sodium selenate and sodium selenite were readily accepted by yellowjackets but failed to provide control. The baits were not repellent and possibly even greater concentrations of bait might be tested in the future.

7). The isoxazolines, fluralaner and sarolaner, were tested. Fluralaner baits containing 0.025% and 0.05% in both PAA and minced provided varying levels of control. However, dense populations of yellowjackets and considerable amounts of competitive foods at the San Diego Zoo Animal Safari reduced its effectiveness. Higher concentrations of 0.144% and 0.024% sarolaner provided excellent reductions of yellowjackets at several locations. This combination toxicant looks extremely promising.

8). The isoxazoline and fipronil baits were also effective against *V. alascensis* in the trials where there were enough wasps to tests. The baits were not attractive and did not affect the other species of yellowjackets captured in the heptyl butyrate traps.

9). As expected, there was large decline in bait acceptance of minced chicken and PAA gels. The appearance of reproductive increases the workers focus to plants and carbohydrate foods.

10). The lack of rainfall in 2021 probably had a severe impact on yellowjacket populations at most sites.

APPPENDIX I

Several of the sites included in this study have been monitored in the past. On occasion the numbers of yellowjackets have been extremely high resulting in serious problems. Rust et al. (2017) examined the weather patterns at Irvine Regional Park from 2012-2014 and noted that a lack of rainfall in January and warmer spring temperatures may have contributed to a decline in the number of yellowjackets.

Irvine Regional Park

The total number of yellowjackets trapped in 2019, 2020, and 2021 were 8,219, 27,446, and 17,795, respectively. A wildfire in 2017 along the northern boundary of the Park may have had some impact on the low numbers of yellowjackets trapped in 2019.

From 1991-2020, the first 6 months of the year averaged 22.71 cm of rain with January and February receiving the highest level of rainfall of 7.32 and 8.03 cm of rain, respectively (fire station, Santa Ana, CA, 33°44'39.04" N, 117°52'.05.35"W, 33.5 m elev.). The only year that the Park received greater than normal rainfall was 2019 (Table 1).

The amount of rainfall from the preceding months from September to December is included in Table 2.

Table 1. Temperatures and precipitation near the Irvine Regional Park site for 2018 to 2021.

2018					2019				
	Temperature (°C)[a]					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Jan	22.2	16.3	10.4	2.79	Jan	19.2	14.4	9.67	12.78
Feb	20.2	14.8	9.4	0.81	Feb	16.2	11.8	7.6	13.94
Mar	19.7	15.6	11.3	3.10	Mar	19.8	15.7	11.6	3.61
Apr	22.1	17.5	12.9	0.13	Apr	21.7	17.7	13.7	0.08
May	21.3	17.8	14.4	0.20	May	20.2	17.2	14.4	1.27
June	23.9	20.4	16.9	0	June	23.1	19.9	16.8	0.03
total				7.04					31.70

2020					2021				
	Temperature (°C)					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Jan	20.8	15.4	10.0	0.38	Jan	20.44	14.78	9.11	4.47
Feb	22.2	16.7	11.0	0.97	Feb	20.11	15.00	9.89	0.08
Mar	20.2	16.5	12.8	8.10	Mar	19.44	14.61	9.83	2.95
Apr	22.6	18.9	15.1	6.93	Apr	22.06	17.78	13.50	0.05
May	25.6	21.6	17.6	0.03	May	22.33	19.06	15.78	0.38
June	25.2	21.5	17.7	0.03	June	24.8	21.1	17.4	0.0
total				16.43					7.93

[a] TAVG – average temperature. Santa Ana John Wayne Airport (33°40'34.28" N, 117°52'02.91" W, elev. 13.7 m)

Table 2. Temperatures and precipitation near the Irvine Regional Park site for 2018 to 2021 including September to December.

2017					2018				
	Temperature (°C)[a]					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	Rainfall[a] (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Sept	26.9	22.7	18.2	0.0	Sept	25.9	22.3	18.7	0.0
Oct	27.7	22.1	16.6	0.0	Oct	25.2	20.8	16.5	1.32
Nov	23.5	18.4	13.4	0.0	Nov	23.9	18.4	12.9	1.96
Dec	23.1	16.2	9.3	0.03	Dec	19.1	14.6	10.1	8.23
			total	0.03				total	11.51
2018					2019				
Jan	22.2	16.3	10.4	2.79	Jan	19.2	14.4	9.67	12.78
Feb	20.2	14.8	9.4	0.81	Feb	16.2	11.8	7.6	13.94
Mar	19.7	15.6	11.3	3.10	Mar	19.8	15.7	11.6	3.61
Apr	22.1	17.5	12.9	0.13	Apr	21.7	17.7	13.7	0.08
May	21.3	17.8	14.4	0.20	May	20.2	17.2	14.4	1.27
June	23.9	20.4	16.9	0.0	June	23.1	19.9	16.8	0.03
total				7.04					31.70

2019					2020				
	Temperature (°C)					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Sept	28.2	24.1	20.0	0.08	Sept	29.4	24.3	169.1	0.0
Oct	27.6	21.6	15.6	0.0	Oct	27.7	22.6	17.5	0.0

Nov	23.4	17.9	12.5	3.56	Nov	22.7	17.0	11.2	0.94
Dec	19.6	15.0	10.5	3.40	Dec	21.5	15.0	8.5	2.72
			total	7.04				total	3.66
2020					2021				
Jan	20.8	15.4	10.0	0.38	Jan	20.44	14.78	9.11	4.47
Feb	22.2	16.7	11.0	0.97	Feb	20.11	15.00	9.89	0.08
Mar	20.2	16.5	12.8	8.10	Mar	19.44	14.61	9.83	2.95
Apr	22.6	18.9	15.1	6.93	Apr	22.06	17.78	13.50	0.05
May	25.6	21.6	17.6	0.03	May	22.33	19.06	15.78	0.38
June	25.2	21.5	17.7	0.03	June	24.8	21.1	17.4	0.0
total				16.43					7.93

[a] Rainfall averages from 1991-2020. Sept-Dec = 11.7 cm; Jan-March = 19.69.

UCR Campus

Over the years the UCR site has served as a control site because the average number of yellowjacket trapped/trap/day rarely exceeds 10. The total number of yellowjackets trapped for 2019, 2020, and 2021 was 4,624, 6,190, and 466, respectively.

From historical rainfall from 1970-2021, the average rainfall for the first 6 months was 15.35 cm with February (5.3 cm) and March (4.2 cm) receiving the greatest rainfall. The rainfall exceeded the normal average in both 2019 and 2020, but there was a severe drought in the spring of 2021 which reflects the very low number of yellowjackets trapped.

Table 3. Temperature and precipitation for the UCR site for 2019 to 2021. The annual average precipitation for Riverside is 24.4 cm of rain per year (1970-2021).

2019					2020				
	Temperature (°C)					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min	rainfall (cm)
Jan	17.3	10.4	4.0	5.13	Jan	18.8	9.5	1.7	0.25
Feb	13.7	7.8	1.9	9.40	Feb	20.6	11.4	2.1	1.04
Mar	18.5	11.8	5.2	5.03	Mar	17.7	11.5	5.3	10.87
Apr	24.1	15.4	7.7	0.58	Apr	21.9	14.8	7.9	9.58
May	21.9	14.7	8.7	2.74	May	28.8	19.4	10.6	0.00
June	29.1	20.0	13.7	0.00	June	29.9	20.5	12.9	0.13
total				22.89					21.87

2021				
	Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	Rainfall (cm)
Jan	19.7	10.9	2.7	3.43
Feb	19.0	10.8	2.9	0.03

Mar	19.1	11.5	3.9	3.66
Apr	24.7	15.9	7.4	0.00
May	26.3	17.3	10.9	0.00
June	33.1	23.5	14.3	0.15
total				7.26

[a] TAVG – average temperature. March AFB (33°53'28.08" N, 117°15'43.97" W, elev. 462.4 m).

San Diego Zoo Safari Park

The total number of yellowjackets trapped during 2020, and 2021 were 41,592 and 7,231, respectively.

From 2000 to 2021, the first 6 months of the year averaged 20.7 cm of rain with January and February receiving the highest level of rainfall of 5.77 and 6.76 cm of rain, respectively. The average annual rainfall was 32.68 cm. From January to June, the Park received greater than normal rainfall during 2019 and 2020, but 2021 was extremely dry.

Table 4. Temperature and precipitation for the San Diego Zoo Safari Park for 2019 to 2021.

2019					2020				
	Temperature (°C)[a]					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min	rainfall (cm)
Jan	19.2	13.4	7.6	9.07	Jan	21.2	14.1	6.9	1.19
Feb	16.3	11.3	6.4	16.44	Feb	22.4	15.3	8.2	1.59
Mar	20.6	15.2	9.9	1.14	Mar	19.8	14.9	10.0	14.25
Apr	24.3	18.4	12.6	0.41	Apr	23.1	17.9	12.8	13.39
May	22.0	17.6	13.2	3.56	May	27.6	21.4	15.3	0.05
June	27.0	21.7	16.3	0.18	June	28.6	22.4	16.3	0.36
total				31.3					30.83

2021				
	Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	Rainfall (cm)
Jan	21.8	14.4	7.2	7.75
Feb	21.8	14.7	7.6	0.58
Mar	21.1	14.6	8.0	4.80
Apr	24.6	18.1	11.4	0.25
May	24.9	19.1	13.4	0.08
June	29.3	22.5	15.7	0.0
total				13.46

[a] TAVG – average temperature. (33°07'24.20" N, 117°05'42.06" W, elev. 196.6 m)

UC Berkeley Richmond Field Station

The total number of yellowjackets for 2019, 2020, and 2021 was 15,077, 7,070, and 10,480.

The historical rainfall from 2000 to 2021 for the first six months of the year averaged 24.24 cm of rain with January, February and March receiving an average of 8.08, 9.55, and 7.84 cm of rain. The annual average rainfall is 53.14 cm. In 2018 and 2019, the rainfall exceeded the average for the first six months.

Table 5. Temperature and precipitation for the Oakland International Airport for 2019 to 2021.

2018					2019				
	Temperature (°C)[a]					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Jan	14.7	10.7	6.6	11.66	Jan	15.7	11.8	7.9	9.52
Feb	16.7	10.9	5.1	0.86	Feb	13.7	10.1	6.6	15.42
Mar	16.1	11.7	7.1	9.27	Mar	17.2	12.8	8.3	9.27
Apr	18.2	13.7	9.1	5.44	Apr	19.9	15.8	11.6	1.20
May	18.7	14.96	11.3	0	May	19.0	15.6	12.1	2.97
June	21.6	16.7	11.9	0	June	24.4	18.8	13.8	0
total				27.23					38.38

2020					2021				
	Temperature (°C)					Temperature (°C)			
Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)	Month	Mean Max.	TAVG	Mean Min.	rainfall (cm)
Jan	15.1	10.8	6.6	4.42	Jan	16.7	11.9	7.2	6.15
Feb	19.2	13.4	7.6	0	Feb	18.6	13.2	7.7	4.44
Mar	17.8	13.3	8.8	3.84	Mar	17.9	12.7	7.5	4.4
Apr	19.7	15.4	11.1	2.31	Apr	17.6	13.5	9.4	0
May	23.7	18.4	13.1	1.12	May	19.8	15.2	10.5	0.03
June	24.2	19.2	14.2	0	June	21.5	17.7	13.4	0.0
total				11.69					14.72

[a] TAVG – average temperature. Oakland International Airport (37°43'09.83" N, 122°13'06.06" W, elev. 1.8 m)

Discussion

Yellowjacket queens emerge in May and June to begin nest construction. As the colonies are established, there will be increasing demands for insects to provision the developing larvae. Exogenous factors affecting populations of *V. germanica* and *alascensis* have been examined, but nothing definitive has been shown to effect populations. In the arid west, wildflowers and vegetation are extremely dependent on the spring rains. Bowers (2005) found that rains prior to good wildflower years were at least 30% greater than long-term averages in the Mohave Desert and at least 50% greater in the Sonoran Desert. In comparing the months prior (September through March) for Irvine Regional Park, the rainfall was far short the average rainfall over the past 30 years. Only in 2019, did the numbers approach the historical average rainfall for those months.

References Cited:

Bowers, J.E. 2005. El Niño and displays of spring-flowering annuals in the Mojave and Sonoran deserts. J. Torrey Botanical Soc. 132: 38-49.