Zinc Phosphide-Treated Bracts as an Alternative Rodenticide in Artichoke Fields for Meadow Vole (*Microtus californicus*) Control

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ABSTRACT: Artichoke growers in Monterey County, California currently use a fresh artichoke bract chlorophacinone bait to control their primary vertebrate pest, the California meadow vole. Upon suspected chlorophacinone resistance by meadow voles in artichoke fields, an alternative has been sought. We studied the effect of zinc phosphide-treated artichoke bracts on California meadow voles. We found that zinc phosphide-treated artichoke bracts were effective in reducing meadow vole populations on treated plots by 95-98%. Our results suggest that zinc phosphide-treated artichoke bracts are effective in reducing California meadow vole populations in artichoke fields and may provide a useful alternative for areas in which anticoagulant resistance by voles is suspected.

KEY WORDS: anticoagulants, artichokes, California meadow vole, *Microtus californicus*, resistance, rodenticide, vertebrate pest control, zinc phosphide

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INTRODUCTION

The California meadow vole (*Microtus californicus*) is the most common species of vole in California, and a serious pest of artichoke crops. Artichoke growers in Castroville, California currently use a chlorophacinone rodenticide (0.01% chlorophacinone oil artichoke bract bait) to control voles and decrease crop damage. However, concerns have been raised by growers about an apparent decrease in efficacy of chlorophacinone for meadow vole control. In response to this, Salmon and Gibson (2003) studied the efficacy of chlorophacinone on vole control. They found a poor dose-response correlation of chlorophacinone on meadow voles, as well as a decrease in overall efficacy of chlorophacinone, both indicators of anticoagulant resistance. Salmon and Gibson (2003) also examined zinc phosphide-treated artichoke bracts as an alternative toxicant and found it to be effective in controlling voles, reaching up to 100% mortality in outdoor pen trials. The objective of our research was to examine the effectiveness of using 0.5% zinc phosphide-treated fresh artichoke bracts in controlling vole populations in a field application. Because bait acceptance of artichoke bracts by meadow voles was found to be higher than available alternative bait carriers (Marsh *et al.* 1984), it was the only carrier tested for field use.

METHODS AND MATERIALS

Study Area

The study was conducted near the town of Castroville, Monterey County, California from January through February, 2004. The climate in Monterey County is typical Mediterranean with low rainfall and hot, dry summers. Field #5 at Strobel Ranch, Sea Mist Farms was selected as the study site based on its history of heavy vole infestation. Rodent control using anticoagulant baits had been temporarily suspended prior to this study at the request of the researchers. Terrain at the study site was flat or moderately sloping, and oxalis weeds (*Oxalis* sp.) were prevalent, although weed control had been performed in the furrows 1 month prior to the study. We established seven 1-ha plots, comprised of 4 treatments and 3 controls. To make the most efficient use of the 4-ha treatment area restriction imposed by our University of California research authorization, we selected a 4-ha artichoke field and divided it into 4 adjacent 1-ha plots. Census areas within each plot were located at least 15 m from the edge of the plot and were at least 30 m from the census area of any adjacent plots. Control plots were selected in the same manner and were located at least 500 m from treated plots.

Census Methods

Two indexing methods (indirect and direct) were conducted for 2 days each, pre- and post-treatment for a total of 8 indexing days (Table 1). The direct and indirect indexing methods were conducted on separate days.

Indirect Method

Various indices have been used for estimating vole populations including measuring consumption of apple slices (Hayes and Cullinan 1984, Tobin *et al.* 1992). We used a chew card method (Caughley *et al.* 1998), but we

Table 1. Schedule of events for zinc phosphide baiting trial in Castroville, CA, 2005.

Date	Actions Performed	Stage
1/25/04	Chew Index Day 1, Acclimate Traps	Pre-Treatment
1/26/04	Chew Index Day 2, Acclimate Traps	
1/27/04	Trap Index Day 1	
1/28/04	Trap Index Day 2	
1/30/04	Zinc Phosphide Baiting Day	Treatment
2/2/04	Chew Index Day 1, Acclimate Traps	Post-Treatment
2/3/04	Chew Index Day 2, Acclimate Traps	
2/4/04	Trap Index Day 1	
2/5/04	Trap Index Day 2	

used artichoke bracts as the chewing device as they were already well accepted by voles in the artichoke fields. Because of the communal living habits of voles, we used a binary (chewed vs. not chewed) rather than a quantitative (amount chewed) index.

One hundred wire flags were placed in a grid near the center of each plot. The grids were established by placing a flag at the base of every fourth plant in 10 consecutive artichoke rows. Each grid measured approximately 30 × 70 m. On each morning of the index period, a single fresh artichoke bract was attached through the wire support at the base of each flag, flush with the ground. Each evening a researcher removed the bract and recorded whether the bract was chewed or not chewed. Bracts that were completely missing were assumed to be taken by voles and recorded as "chewed." This process was conducted for 2 days each, pre- and post-treatment. Bracts were completely removed in the afternoon, to avoid chewing from nocturnal mice known to inhabit the fields. The chew index was limited to 2 days to prevent voles from becoming accustomed to finding the bracts.

Direct Method

Twenty trapping stations were established on each plot with 3 live-catch traps at each station, for a total of 60 traps per plot. The trapping stations were 10 m apart, and the traps were set near runways and burrows located within 3 m of the center of the station. The traps were positioned on rows 1, 5 or 6, and 10 of the chew index grid with 6-7 trap stations per row. The trapping grid measured approximately 40 × 40 m. Traps were locked open and baited in the morning to serve as an acclimation period for 24-48 hours before the onset of trapping.

Following the acclimation period, the traps were set and baited with artichoke hearts each morning between 0600 and 0900 for 2 consecutive days. In the morning of Day 2, oats were added to the traps to help sustain the vole throughout the day. Brown cotton was added to the traps as a bedding material on days of rain. The traps were checked between 1530 and 1730 and then closed for the night. All voles caught were marked with ear tags and released at the point of capture. Captured voles were examined for signs of an ear tag that may have fallen off and all recaptures were recorded. All voles were examined and determined to be in good health before being released. Voles that died in the traps were recorded and left in the field. The same method was repeated post-treatment.

Hantavirus Safety Guidelines

Hantavirus is a human disease known to be carried by deer mice (*Peromyscus* spp.) and transmitted through their urine, feces, and saliva (Johnson 2001). Pre-trial trapping indicated that deer mice were present in the artichoke fields and that they were most active at night. For this reason, we trapped only during the daylight hours. To decrease potential exposure to hantavirus, traps were opened downwind and away from the trapper's the virus (D. Bryson, Liphatec, Inc., pers. commun.). Traps were re-set the next morning. If a trap was excessively soiled by deer mouse urine or feces, it was replaced with a clean trap.

Baiting

Several batches of bait were mixed according to the USDA zinc phosphide label specification for fresh vegetable bait (EPA Reg. No. 56228-6). The batches were mixed according to the following procedure: to 10 lbs of bracts (4.55 kg), we added 1 ounce (28.35 g) of canola oil followed by 40 g of zinc phosphide (ZnP) powder (63.2% active ingredient). The canola oil was added first to help the ZnP powder adhere to the bracts and to reduce airborne ZnP particulates. Bracts were mixed in 80-lb batches for a minimum of 10 minutes per batch in a standard capacity bait mixer at the Kleen Globe bait mixing facility, Castroville, CA. Batches of placebo bracts (containing canola oil only) were mixed for the 3 control plots. Placebo bracts were mixed before zinc phosphide-treated bracts to avoid contamination. Handlers and applicators wore appropriate personal protection equipment as required for zinc phosphide.

All treated and control bracts were moved into plastic-lined bins and transported by tractor to the test site. Following the pre-treatment indexing period, Plots 1-3 were treated with placebo artichoke bracts and Plots 4-7 were treated with 0.5% zinc phosphide artichoke bracts at a rate of 4-5 bracts per plant, the rate used for chlorophacinone treated artichoke bracts (CDFA Label 10965-50067-AA SLN CA-930022). Bracts were hand-placed on the ground at the base of each plant.

DATA COLLECTION AND ANALYSIS

For the direct (trapping) index method, we calculated the number of voles caught per available trap each trapping day, pre- and post-treatment. Voles found dead in traps were not included in analysis, nor were non-target captures, and the traps in which they were found were not counted as "available" in analysis. We calculated the percent reduction in the population index using the following formula:

$$\{(pre\text{-}post) / pre\} \times 100\% \quad [1]$$

where "pre" and "post" refer to the average pre treatment post treatment voles per trap available, respectively. For the indirect (chew) index, we calculated the number of bracts chewed of 100 offered for each day. The average for pre and post treatments were calculated using Equation 1, but substituting the number of bracts chewed for the number of voles trapped.

The percent reduction between pre and post treatment for both indices was compared using a 2-way Analysis of Variance (ANOVA) using the GLM Procedure with a Tukey adjustment comparing least squares means (SAS Version 9.1). Differences were considered significant at $\alpha = 0.05$.

RESULTS

Trapping Index

Percent reduction on treated plots ranged from 89.9 to

Table 2. Trapping ... population index.

	Plot	Pre-Treatment			Post-Treatment			% Change Pre to Post Treatment
		1/27/04	1/28/04	Avg. 1	2/4/04	2/5/04	Avg. 2	
Control	C-1	0.133	0.050	0.092	0.133	0.190	0.162	76.1%
Control	C-2	0.276	0.085	0.181	0.305	0.246	0.276	52.5%
Control	C-3	0.283	0.153	0.218	0.373	0.373	0.373	71.1%
Treatment	T-4	0.464	0.250	0.357	0.035	0.036	0.036	-89.9%
Treatment	T-5	0.439	0.207	0.323	0.000	0.017	0.009	-97.2%
Treatment	T-6	0.339	0.119	0.229	0.000	0.017	0.009	-96.1%
Treatment	T-7	0.321	0.109	0.215	0.017	0.017	0.017	-92.1%

Table 3. Chew index results from zinc phosphide trial showing the number of bracts chewed out of 100 available bracts in each plot. Negative numbers indicate a decrease in the population index.

	Plot	Pre-Treatment				Post-Treatment				% Change Pre to Post Treatment
		1/25/04	1/26/04	Total 1	Avg. 1	2/2/04	2/3/04	Total 2	Avg. 2	
Control	C-1	37	28	65	32.5	37	40	77	38.5	18.5%
Control	C-2	65	41	106	53.0	41	46	87	43.5	-17.9%
Control	C-3	58	45	103	51.5	52	55	107	53.5	3.9%
Treatment	T-4	77	90	167	83.5	5	5	10	5.0	-94.0%
Treatment	T-5	75	77	152	76.0	5	2	7	3.5	-95.4%
Treatment	T-6	69	82	151	75.5	6	1	7	3.5	-95.4%
Treatment	T-7	79	78	157	78.5	2	0	2	1.0	-98.7%

showed a difference in the change in population indices between sessions depending on the treatment ($F_{1,5}$ = 73.82, P = 0.0004). The population index was significantly lower after baiting in the treated area (P = 0.0009), with no difference in control plots (P = 0.0762).

Chew Index

The average number of bracts consumed on treatment days ranged from 75.5 to 83.5%, with an average population reduction of 94 to 98.7% (Table 3). On control plots, the number of bracts chewed ranged from 32.5 to 53% with a population change ranging from a decrease of 17.9% to an increase of 18.5% (Table 3). Two-way ANOVA showed a difference in the change in chew index between sessions depending on the treatment ($F_{1,5}$ = 88.84, P < 0.0001). We found no difference in chew index between treated and control plots (P = 0.2209). Differences in chew index overall varied pre and post treatment (P < 0.0001). We found a method × day interaction (P < 0.0001), suggesting more detailed analysis is required.

DISCUSSION

Both indices show a decrease in activity in the treatment plots after treatment, while the control plots showed in all but one case an increase in activity. This suggests that the decrease is a result of zinc phosphide treatment and not the result of other factors, such as disease. We included Figures 1 and 2 to provide a graphical representation of the results, illustrating the sharp decline in activity after treatment. Even with natural variations in activity, it is evident that treatment with 0.5% zinc phosphide had a dramatic effect on both activity indices.

Although there was variation in daily activity levels on each plot, if the treatment was ineffective, we would expect similar changes in activity from pre- to post-treatment in all 7 plots. In both figures, it is apparent that the activity in the treated plots changed considerably more than in control plots. This change in activity supports our hypothesis that a 0.5% zinc phosphide artichoke bract treatment is an effective treatment for controlling meadow voles in artichoke fields.

Daily vole activity can differ depending on weather conditions. For example, on the second day of the pre-treatment trapping session, there was a sharp decline in trapping success. This coincided with warmer temperature in the afternoon, when compared to the other 3 trapping days. To determine the effect of this weather change on the results, we recalculated the percent change in activity for the trapping index to exclude the day in question. By removing this day, the average change of activity in control plots changed from an average increase of 66.6% to an average increase of only 17.9%. However, the index changes in the treatment plots were relatively unaffected (Table 4).

We included the recaptured voles in our daily measure of trapping activity, but we did not perform a capture/recapture analysis. One could argue that this does not take into account the trap affinity that may have developed and introduces bias into the pre-treatment population estimate. We reduced the potential for trap

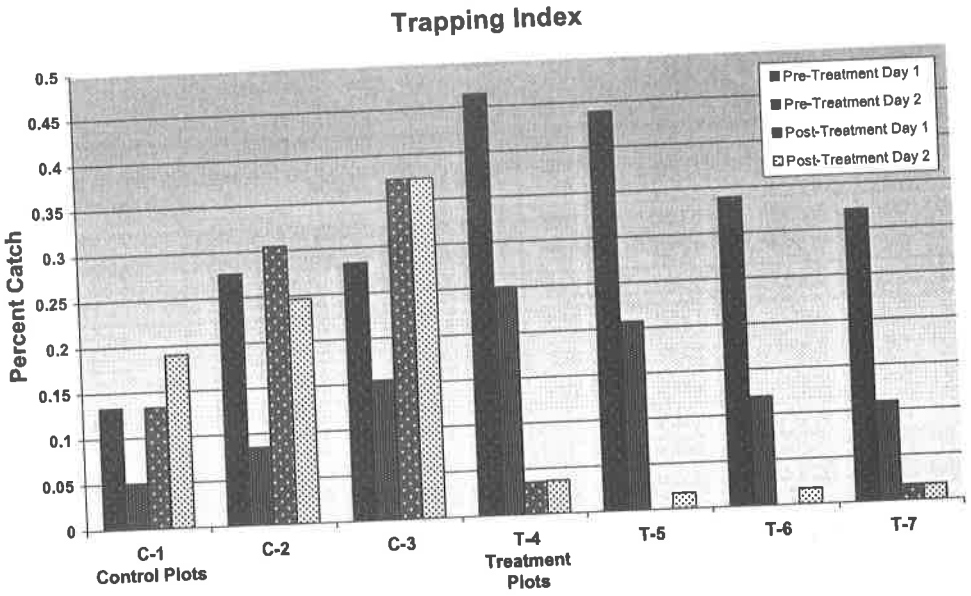

Figure 1. Daily percent catch of voles during zinc phosphide trial.

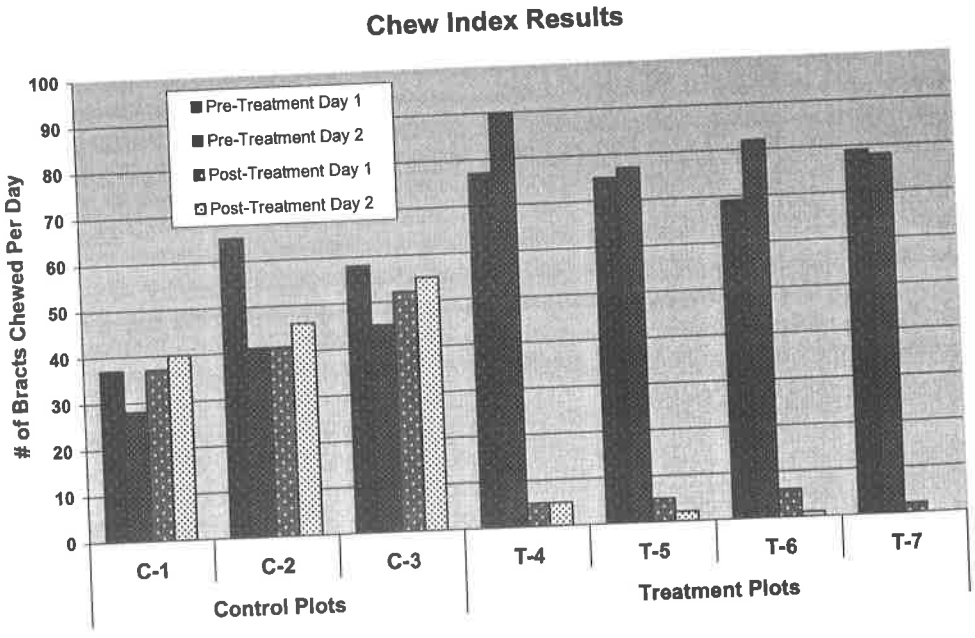

Figure 2. Daily chew index results from zinc phosphide trial.

Table 4. Percent change in activity indices pre to post-treatment, adjusted to exclude Jan. 28th results. Negative numbers indicate a decrease in the population index.

Plot		Trapping Index Including 1/28	Average	Trapping Index Excluding 1/28	Average	Chew Index	Average
Control	C-1	76.1%	66.6%	21.8%	17.9%	18.5%	1.5%
	C-2	52.5%		0.0%		-17.9%	
	C-3	71.1%		31.8%		3.9%	
Treatment	T-4	-89.9%	-93.8%	-92.2%	-95.5%	-94.0%	-95.9%
	T-5	-97.2%		-97.9%		-95.4%	
	T-6	-96.1%		-97.3%		-95.4%	

affinity by limiting trapping to 2 days in each session. Fifteen percent of the voles were recaptured in the same trapping session (37 of 244 live voles), and we considered this margin of error acceptable.

Dead voles found in traps were not included in our results, nor was the trap they were found in counted as an available trap. Our reason for not including them was because they could not be used in our index, as they would not be available for activity at a later time and because they did not die of natural causes. The number of traps tripped without an animal in it were not recorded and assumed to be constant over all plots. Carcass searches produced 14 carcasses in treated plots within 24 hours of baiting, a timeframe consistent with mortality from zinc phosphide poisoning. No carcasses were found in control plots. However, it is important to note that carcass searches are limited by the dense foliage of artichoke fields and the fact that many of the voles likely died underground. And although the carcass searching data cannot be used to establish population levels or changes to those levels, it does further support the observation that the zinc phosphide treatment was the cause of the change in activity levels pre to post-treatment.

MANAGEMENT IMPLICATIONS

The fast-acting nature and overall efficacy of zinc phosphide-treated artichoke bracts suggest they are viable alternative to chlorophacinone. Video surveillance conducted during a similar project showed that voles were the primary consumer of artichoke bract bait, with minimal feeding by deer mice. During the post-treatment carcass search, no non-target carcasses were found, suggesting minimal risks to non-target species. Additionally, the low persistence of zinc phosphide in the environment and the fact that it does not accumulate residue in the carcass makes zinc phosphide an attractive alternate for use on anticoagulant resistant voles (Staples *et al.* 2003).

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