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Montane Vole Control with Rozol® Paraffinized Pellets in Orchards of the Pacific Northwest

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ABSTRACT: Rozol® Paraffinized Pellets (0.005% chlorophacinone) were effective in reducing populations of montane voles greater than the EPA standard of 70%. This test substance was applied using a Vicon spreader at the approximate rate of 10 lbs/acre to the vegetation of the orchard floor. An intensive mark-recapture census was used as a direct census to determine population change as a result of the test substance application. An apple slice index was used as an indirect method to confirm population change. Few carcasses were found during the carcass search practiced in this study. Determination of active ingredient residues was done on the vole carcasses found during the carcass search events.

KEY WORDS: apple orchard, chlorophacinone, deer mouse, efficacy, mark-recapture census, *Microtus montanus*, montane vole, *Peromyscus maniculatus*, rodent control, rodenticide, whole-body residue

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INTRODUCTION

The montane vole (Microtus montanus) is the predominant vole species that causes damage to orchards of the Pacific Northwest. In one study, it was reported that montane vole damage resulted in losses in production of 21% from trees of a red delicious apple orchard and a 51% loss of production from trees in a golden delicious apple orchard (Askham 1988). Little published information is available about the biology of montane voles that reside in orchards of the Pacific Northwest. Vole populations tend to be cyclical in nature, and the amount of damage realized within an orchard can be dependent upon vole population numbers, amount and duration of snow, as well as age and planting density of trees. LiphaTech Inc. is applying for a §3 Federal EPA registration of Rozol® Paraffinized Pellets (0.005% chlorophacinone) for use as a broadcast application at the rate of 10 lbs/acre to control voles in orchards. Rozol® Paraffinized Pellets have been used for years in orchards to control voles as allowed by 24(c) labels acquired at the state level.

During October 2002, a field efficacy study was performed to show that Rozol® Paraffinized Pellets could reduce vole populations in orchards by 70% or greater as required by EPA. This study was performed following Good Laboratory Practices (GLP) requirements.

METHODS AND MATERIALS

Study Site

The test and control plots were located in an orchard 7.5 miles south of the town of Ephrata in Grant County, Washington. Elevation at this site is approximately 1,280 feet above sea level. The tree variety of the orchard is red delicious, with crab apple trees and banana apple trees planted throughout the orchard to serve as pollinators. Trees were approximately 22 years old, planted with 10-foot spacing between trees within a row, and at a density of 242 trees per acre. The orchard is watered using sprinkler irrigation. Tall fescue is the predominant plant species growing on the orchard floor, with clover,

dandelion, mallow, and orchard grass making up a small percentage of the vegetation.

Four treatment plots, in a block configuration, were used in this study. The block of 4 plots covered an area of 4.36 acres with each treatment repetition being 1.09 acres. Each of the 4 plot repetitions was broken up into 66 subplots of 720 square feet each. This grid pattern was established to help better document vole movement and home ranges by identifying where voles are trapped during the direct census event and found during the carcass search. Twenty-one of the subplots of each plot repetition served as a buffer zone between other plot repetitions within the block of 4 plot repetitions. The remaining 45 were used to calculate the efficacy of the trial through the direct census method used in this study. A buffer zone of approximately 23 feet around the perimeter of the block of 4 plot repetitions was treated with the test substance. The untreated control plots were set up in the same configuration except that a treated buffer was not established around the perimeter of the plots.

Population Census

Two methods were used to monitor for change in vole population within each treatment and untreated control plot repetition. A mark-recapture census (MRC) was used as a direct measure of the vole population and to obtain information on vole movement within the orchard. An apple slice index (ASI) was used as an indirect census method.

The MRC was performed 2 times, with each being a 2-consecutive-day event, in both the plots repetitions of the treatment and of the untreated control. A Sherman live-catch trap was placed in each of the subplots of a plot repetition for acclimation 3 days before the actual census event began. An acclimation period was performed on the 3 consecutive days preceding the pre-treatment MRC as well as during the 3 consecutive days preceding the post-treatment MRC. During the acclimation period, each trap door was clipped in the open position and oats

were placed on the trap trigger pan and in front of the trap. A dime-sized portion of peanut butter was added to the inside trap surface on the last day of acclimation. At this time, the air temperature was low enough to preclude slugs and ants from being attracted to the peanut butter in

the trap.

immediately Trapping began following acclimation period. Trapping was only performed during the day since it was determined prior to the study, by snap-trapping areas of the orchard that were not included in the study, that the voles were mainly active during the day. Not trapping overnight avoided catching deer mice (Peromyscus maniculatus) and helped to avoid trap mortality that might have been realized during an overnight trapping period. Each trap was set in the morning and baited with oats, a dime-sized portion of peanut butter, and a small slice of potato to supply trapped animals with a source of moisture. Traps were checked and captured voles ear-tagged (or ear-tag numbers were recorded in the case of repeat catches), the afternoon of the same day they were set. All trap doors were left in the closed position after they were checked.

The ASI was performed 2 times. The first (pre-treatment) ASI was done the day following the last day of the first (pre-treatment) MRC event. A second ASI was performed 7 days after the test substance was broadcast in the orchard. ASI events helped in the determination of when the second (post-treatment) MRC could be done and served as a general indicator of population trend. To perform the ASI, 1 apple slice was placed in each of 10 randomly chosen subplots of a plot repetition. The slices were placed in the subplots during the morning and graded for activity the afternoon of that same day.

Bait Application

A Vicon Granular Applicator Model PS203, mounted on a John Deere 5400N orchard tractor, was used to apply the test substance. A low-flow kit, provided by the manufacturer of the Vicon Spreader, was used to plug 2 of the 3 discharge holes in the applicator hopper. The test substance was applied at approximately 10 lbs/acre (actual application was 10.16 lbs/acre) to the vegetation of the orchard floor. An applicator uniformity test at this rate showed that the test substance was broadcast at an average density of 5.5 pellets per square yard.

Efficacy Determination

Change between pretreatment MRC and posttreatment MRC events was determined for the treatment plot repetitions using the equation in Table 1. The change for the untreated control plot repetitions was calculated using the equation found in Table 2. Another equation was used to determine the recapture percentage. The recapture percentage is the percentage of animals captured, ear-tagged, and released during the first 2-day MRC event that were recaptured during the second 2-day MRC event. The equation used to calculate this percentage is found in Table 3.

The ASI gives more of a general measurement of population trend by measuring feeding activity. The grading of apple slices was done following a scale found in Godfrey and Askham (1986). The scale used in

grading the apple slices is shown in Table 4. Each score is then multiplied by the number of occurrences of that particular grading score, and the product of all grading categories are summed. This sum is divided by the number of apple slices placed in the plot repetition being indexed to give the ASI score for that repetition. The two scores obtained from the first ASI and the second ASI are then used in the equation presented in Table 5 to obtain the percentage change realized between the two ASI events in that particular plot repetition.

Carcass Search / Wildlife Observations

Both the treatment and untreated control plot repetitions were searched for carcasses by walking each area on a daily basis starting on the second day after application of test substance to the treatment plots. During the daily carcass search, the person performing the search walked all subplots at a pace that allowed visual scanning of the area of each subplot. Dead animals were collected, assigned unique identification numbers, and their locations noted by plot and subplot number.

Carcasses of *M. montanus* were frozen and shipped to the National Wildlife Research Center for whole-body analysis to determine chlorophacinone residues. The only other carcass found were those of *P. maniculatus*. These were frozen for subsequent necropsy to determine if signs of anticoagulant poisoning were present. During the study, observations regarding wildlife in the orchard were

RESULTS AND DISCUSSION

Efficacy

required.

The calculations of the MRC data used in determining the efficacy of the test substance in controlling *M. montanus* are found in Table 6. This intensive direct census technique showed that the test substance exceeded the required EPA standard of 70% population reduction

The results from the percentage recapture equation (Table 7) also show that within the treatment plots, very few individuals ear-tagged during the pre-treatment MRC were present during the post-treatment MRC. Table 8 shows the calculations used to determine the change in population between the pre-treatment MRC and the post-treatment MRC for the untreated control plot repetitions

and the average change of all 4 repetitions.

The results from the percentage recapture equation in Table 9 show that within the untreated control, over one-third of the individuals ear-tagged during the pre-treatment MRC are present during the post-treatment MRC. This is a large percentage of recaptures, considering the population growth that the untreated control experienced between the pre-treatment MRC and the post-treatment MRC. When the average percentage recapture for the treatment plots is compared to the untreated control plots, there were 94.2% fewer recaptures in the treatment.

The ASI is more sensitive to environmental differences between plot repetitions. Two plot repetitions of the untreated control and two plot repetitions of the treatment were not used, due to such influences. Plot repetitions 1 and 2 of the untreated control were not

Table 1. Equation used to calculate the efficacy of test substance in controlling voles in the plot repetitions of the treatment.

Treated Block population change % = (T29surv + T30surv) - (T12 + T13nt) x 100

Where:

T12 = number of voles trapped on Oct 12.

T13nt = number of voles trapped on Oct 13 that did not have ear-tags from Oct 12.

T29surv = total number of voles captured on Oct 29, both with and without eartags.

T30sury = total number of voles captured on Oct 30 that were not captured on October 29.

Table 2. Equation used to calculate the change in population numbers between mark-recapture census (MRC) events performed in the repetitions of the untreated control.

Control Block population change % = (C29surv + C30surv) - (C12 + C13nt) x 100 C12 + C13nt

Where:

C12 = number of voles trapped on Oct 12.

C13nt = number of voles trapped on Oct 13 that did not have ear-tags from Oct 12. C29surv = total number of voles trapped on Oct 29, both with and without eartags.

C30sury = total number of voles captured on Oct 30 that were not captured on Oct 29.

Table 3. Equation used to calculate the recapture percentage or number of voles tagged in each plot repetition during the first mark-recapture census (MRC) that were recaptured in that plot repetition during the second MRC.

Number of voles ear-tagged during first

MRC that were recaptured during second MRC X 100

Number of voles captured, tagged and released during first MRC event

Table 4. Scale used in grading apple slices of apple slice index event for feeding activity.

0 = no biting on apples

1 = one small area chewed on apple slice

2 = two or more areas chewed on apple slice (less than half the apple slice eaten)

3 = more than half the apple slice eaten

4 = the apple slice completely eaten

Table 5. Equation for calculating the percentage difference between the first and second apple slice index event for each plot repetition.

Percentage change between Apple Slice Index (ASI) for given plot repetition=

Score of first ASI x 100
Score of first ASI

Table 6. Efficacy calculation for the plot repetitions of the treatment and the average control for all plot repetitions of the treatment plot.

Piotilo	Number of voles captured on Oct 12 T12	captured on Oct 13	Number of voles captured on Oct 29 T29surv	captured on Oct 30
TRT Plot 1	10	13 untagged (of 17 total)	2	3
TRT Plot 2	13	9 untagged (of 13 total)	3	3
TRT Plot 3	17	10 untagged (of 18 total)	2	2
TRT Plot 4	14	12 untagged (of 18 total)	3	0

The corresponding efficacy calculations are:

TRT Plot 1: $\frac{(2+3) - (10+13)}{(10+13)} \times 100 = \frac{-18}{23} \times 100 = -78.3\%$ TRT Plot 2: $\frac{(3+3) - (13+9)}{(13+9)} \times 100 = \frac{-16}{22} \times 100 = -72.7\%$ TRT Plot 3: $\frac{(2+2) - (17+10)}{(17+10)} \times 100 = \frac{-23}{27} \times 100 = -85.2\%$ TRT Plot 4: $\frac{(3+0) - (14+12)}{(14+12)} \times 100 = \frac{-23}{26} \times 100 = -88.5\%$

Average population change of the treatment plot repetitions = -81.2%

Table 7. Percentage of voles ear-tagged and released in the plot repetitions of the treatment during the pre-treatment markrecapture census (MRC) that were recaptured during the post-treatment MRC.

Plot ID	Number of voles captured, tagged & released on Oct 12/13	Number of voles recaptured Oct 29/30 with tags from Oct 12/13	Recapture percentage
TRT Plot 1	23	**************************************	1/22 = 4.3%
TRT Plot 2	22	0	0/22 = 0.0%
TRT Plot 3	27	0	0/26 = 0.0%
TRT Plot 4	26		1/26 = 3.8%

Average recapture percentage for the treated plot repetitions = 2.0%

Table 8. Calculations used to determine the change of population between the pre-treatment mark-recapture census (MRC) and the post-treatment MRC for the untreated control plot repetitions and the average change of all 4 repetitions.

Plot ID	Number of voles captured on Oct 12 C12	Number of voles captured on Oct 13 C13nt	Number of voles captured on Oct 29 C29surv	O Number of voles captured on Oct 30 C30surv
UTC Plot 1	12	17 untagged (of 20 total captured)	21	14
UTC Plot 2	20	9 untagged (of 15 total captured)	29	20
UTC Plot 3	10	5 untagged (of 9 total captured)	18	8
UTC Plot 4	15	18 untagged (of 20 total captured)	31	18

The corresponding population change calculations are

UTC Plot 1:

(21+14) - (12+17) x 100= 6 x 100= +20.7%

(12+17)

29

UTC Plot 2:

 $(29+20) - (20+9) \times 100 = 20 \times 100 = +68.9\%$

(20+9)

UTC Plot 3:

 $(18+8) - (10+5) \times 100 = 11 \times 100 = +73.3\%$ (10+5)

15

UTC Plot 4:

(31+18) - (15+18) x 100= 15 x 100= +48.5%

(15+18)

Average population change in the control block = +52.9%

Table 9. Percentage of voles ear-tagged and released in the plot repetitions of the untreated control during the pretreatment mark-recapture census (MRC) that were recaptured during the post-treatment MRC.

Plot ID	Number of voles captured, tagged & released on Oct 12/13	Number of voles recaptured Oct 29/30 with tags from Oct 12/13	Recapture percentage
UTC Plot 1	29	5	5/29 = 17.2%
UTC Plot 2	29	8	8/29 = 27.6%
UTC Plot 3	15	9	9/15 = 60.0%
UTC Plot 4	33	11	11/33 = 33.3%

Average recapture percentage of the control plot repetitions = 34.5%

Table 10. Average active ingredient (chlorophacinone) observed in each of four Microtus montanus carcasses found in the treatment plots.

Specimen#	Weight (g) of skinned animal	Average A.I. (ppm)*
100	12.6	0.864
102	8.1	0.851
104	6.6	2.210
108	16.5	1.700

^{*}observed concentrations not adjusted for QC recoveries

Table 11. Average A.I. (chlorophacinone) observed in each of two *Microtus montanus* carcass found in the untreated control plots.

Specimen #	Weight (g) of skinned animal	Average A.I. (ppm)*
2	13.0	less than method level of detection (MLOD**)
3	12.2	less than method level of detection (MLOD**)

^{*} observed concentrations not adjusted for QC recoveries

Table 12. Wildlife observations during the course of the study.

Common Name	Scientific name	Notes
Red-tailed hawk (dark phase)	Buteo jamalcensis	Sitting on wind machine in treated block
American kestrel	Falco sparvarius	Hunting open fields next to orchard
Ring-necked pheasant	Phasianus colchicus	Feeding on apples on ground
Morning dove	Zenaldura macroura	Seen flying through orchard and on dirt roads between orchard blocks
Common flicker (Northern flicker)	Colaptes auratus	In poplars at edge of orchard
Black billed magple	Pica pica	In orchard prior to harvest
Common Raven	Corvus corax	Flying high over orchard
American Robin	Turdus migratorius	Migrating through during early part of study - seen feeding on crab apples
Audubon's warbler (Yellow-rumped warbler)	Dendroica coronata	Migrating through
Slate-colored junco	Junco hyemalis	Migrating through

harvested, and apples were still available in these plots during the second ASI event. Plot repetitions 1 and 2 of the treatment had banana apple trees for pollinators. These large, moist apples were present during the first ASI and seemed to be attractive to the voles. During the second ASI, these apples had all deteriorated, thus not competing with the ASI. By averaging the ASI results from the two remaining plots of the untreated control, a 31.5% increase in feeding is obtained. An average of the percentage change between ASI events for plot repetitions 3 and 4 of the treatment shows that there was a 78.8% decrease in feeding. These results support a conclusion that the test substance gave acceptable control of *M. montanus*.

Carcass Search, Analysis and Necropsy, Wildlife Observations

The carcasses of four *M. montanus* and of three *P. maniculatus* were found in the treatment plots during the carcass search. Two *M. montanus* carcasses and one *P. maniculatus* carcass were found in the untreated control plots. One of the two vole carcasses from the untreated control was found the day after the final carcass search, but it was also submitted for carcass analysis.

Table 10 shows the amount of chlorophacinone that was determined to be in each of the vole carcasses found in the treatment plot. The small number of carcasses found did not allow for statistical analysis of the results. All three of the *P. maniculatus* carcass necropsies showed signs of anticoagulant poisoning.

Analysis of the two M. montanus carcasses from the untreated control is presented in Table 11. As would be expected, these animals were not found to have

chlorophacinone residues. Necropsy of the *P. maniculatus* found in the untreated control showed no signs of anticoagulant poisoning.

In frequent visits to the orchard during the study, no wildlife were observed feeding on the test substance. Avian predators that were observed resting in the orchard were only seen hunting in the open fields surrounding the orchard. Wildlife observations are summarized in Table 12. Most of the birds were migrants and were not seen by the study midpoint. No mammalian predators or scavengers were seen in the orchard.

LITERATURE CITED

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^{**} MLOD is reported to be 0.14 ppm