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Novel and current rodenticides for pocket gopher *Thomomys* spp. management in vineyards: what works?

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Abstract

BACKGROUND: Rodenticides are often included as part of an integrated pest management approach for managing pocket gophers (*Thomomys* spp.) given that they are relatively quick and inexpensive to apply. Strychnine has historically been the most effective toxicant for pocket gophers, but its use is currently limited in the United States; alternative registered toxicants have not proven effective. Recent research with baits containing cholecalciferol plus anticoagulant toxicants proved effective against pocket gophers in a lab setting. Therefore, we established a field study to compare cholecalciferol plus anticoagulant combinations [0.03% cholecalciferol plus 0.005% diphacinone (C + D), 0.015% cholecalciferol plus 0.0025% brodifacoum (C + B2)] with strychnine (0.5%) for pocket gopher management.

RESULTS: Strychnine treatments resulted in 100% efficacy after two treatment periods. Both C + D and C + B2 resulted in efficacy significantly greater than 70% after two treatment periods (83 and 75% respectively). Efficacy from C + B1 (85%) was not significantly greater than 70%, but did yield high overall efficacy as well.

CONCLUSION: Although strychnine remains the most effective rodenticide for pocket gopher control, the cholecalciferol plus anticoagulant baits tested would be a good alternative when strychnine is unavailable. C + D may be the best option given that it uses a first-generation anticoagulant as the synergist. © 2016 Society of Chemical Industry

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Keywords: brodifacoum; cholecalciferol; diphacinone; pocket gopher; rodenticide; strychnine

1 INTRODUCTION

Pocket gophers (Thomomys spp.) are one of the most damaging wildlife species to agriculture and natural resource areas throughout the western United States,¹⁻³ with losses in California agricultural lands estimated between 5.3-8.8% when pocket gophers are present.⁴ Pocket gophers are fossorial rodents that feed primarily on root systems of plants, on the cambium layer of trees and vines and on aboveground herbaceous plant parts.⁵ Many techniques are used to manage pocket gophers, including habitat modification, cultural practices, exclusion, trapping, burrow fumigation and rodenticide baiting. Management of pocket gophers is generally most effective when utilizing an integrated pest management (IPM) approach that incorporates multiple techniques.⁶ Of these potential management tools, rodenticide application is generally the preferred option by pest control professionals and land managers given that it is the quickest and least expensive management tool available for pocket gophers.^{2-4,6}

Although quick and relatively inexpensive to use, the effectiveness of rodenticide applications has often been variable.³ Four primary toxicants are currently available for pocket gopher management in most of the western United States: the first-generation anticoagulants chlorophacinone and diphacinone, and the acute toxicants zinc phosphide and strychnine. Historically, strychnine has usually been the most effective of these toxicants for pocket gopher management, although resistance to strychnine has been a problem in some agricultural areas.³ Additionally, strychnine supplies are quite low in the United States, leading to loss of most strychnine pocket gopher products. A number of strychnine-alternative rodenticide products are available for use, but few of these have proven effective (\bar{x} efficacy = 50, 0–30 and 40–50% for chlorophacinone, diphacinone and zinc phosphide products respectively; Witmer G and Baldwin R, unpublished, 2014).^{7–10} This has left pest control professionals and land managers searching for an alternative option when rodenticides are needed to manage extensive problem situations (i.e. large areas where management using other options is cost prohibitive).

Recent investigations have begun to explore the possibility of using a combination of cholecalciferol plus an anticoagulant to

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manage damaging vertebrate species.^{11–13} When an anticoagulant is combined with cholecalciferol, the anticoagulant acts as a synergist to increase the potency of a lower concentration of cholecalciferol by blocking vitamin K₂-dependent proteins that help regulate calcium in host organisms.^{11,14} This lower concentration can reduce the potential for poor bait acceptance, which is often present at higher concentrations of cholecalciferol, while increasing the effectiveness of the rodenticide when compared with cholecalciferol by itself.¹⁴ The combination rodenticides also result in shorter times-to-death than when used individually (e.g. \bar{x} time-to-death for pocket gophers = 7.5 and 10.4 days for combination rodenticides and first-generation anticoagulants respectively; Witmer G and Baldwin R, unpublished, 2014), and often use less of each toxicant; collectively, these reduce secondary toxicity risks.^{11,13}

Currently, registration is being sought for cholecalciferol plus diphacinone (C + D) in New Zealand for brushtail possum (*Trichosurus vulpecula*), roof rat (*Rattus rattus*) and Norway rat (*Rattus norvegicus*) management, as it has proven quite effective. In the United States, recent research has indicated that C + D is also effective for California vole (*Microtus californicus*) control in artichoke fields.^{12,13} As such, we felt that C + D might be a viable alternative to strychnine for pocket gopher management in the United States.

Brodifacoum is a second-generation anticoagulant that, along with strychnine, is often considered to be the most effective rodenticide still in use in the United States.¹¹ However, brodifacoum is not registered as a field rodenticide in the United States given substantial concerns over secondary toxicity;¹⁵ it would not initially appear to be a good candidate as a pocket gopher rodenticide. That being said, efficacy of rodenticides has often been low for pocket gophers,7-9 potentially owing to their avoidance of baits that deliver the active ingredient to the pocket gopher (i.e. pocket gophers typically eat green vegetation and root systems, while most rodenticide baits are necessarily seeds or pelletized products³). Strychnine has historically worked well for pocket gopher management, as it is highly toxic yet exhibits good bait acceptance.³ Pocket gophers are the only species in California and most western states where strychnine can still be used, in part because the rodenticide is applied within the burrow system, thereby substantially reducing direct non-target exposure to the rodenticide. Secondary toxicity concerns are also lower for pocket gopher bait application than for most other rodent species given that pocket gophers spend relatively minimal time above ground.¹⁶ Therefore, a highly toxic product may be more effective at managing a species that often consumes sublethal amounts of less toxic rodenticides,³ while still posing a low secondary toxicity risk owing to minimal exposure to predators and scavengers; brodifacoum could fit this situation. We can further reduce this risk by combining cholecalciferol with brodifacoum, which allows a decrease in the concentration of both active ingredients that is needed to maintain efficacy, while combining cholecalciferol with an anticoagulant could shorten time-to-death, further reducing any secondary non-target exposure.

Given these potential benefits, we recently completed a laboratory study to test the efficacy of cholecalciferol plus anticoagulant rodenticides. Three experimental pelletized options proved effective: 0.03% cholecalciferol plus 0.005% diphacinone (C + D) (Connovation Ltd, Manukau, New Zealand) and 0.015% cholecal-ciferol plus 0.0025% brodifacoum (C + B1) and 0.03% cholecalciferol plus 0.0025% brodifacoum (C + B2) (Bell Laboratories, Inc., Madison, WI). A 0.015% cholecalciferol plus 0.00125% brodifacoum version was ineffective (60% efficacy; Witmer G and Baldwin

R, unpublished, 2014). Although these results were promising, we needed verification in the field to determine their potential utility as a pocket gopher management tool. Therefore, the objectives of the present study were: (1) to determine whether cholecalciferol plus anticoagulant rodenticides were effective at reducing pocket gopher activity in treated fields, and (2) to determine how the efficacy of cholecalciferol plus anticoagulant products compared with the efficacy of a currently registered strychnine product. If one or more of the cholecalciferol plus anticoagulant rodenticides proved effective, they could serve as a viable addition or replacement to strychnine given the current paucity of strychnine in the United States combined with the need to alternate between active ingredients for long-term rodent management.¹³

2 MATERIALS AND METHODS

Study sites were located in three vineyards around the city of Lodi in San Joaquin County, California. Cover crops of mostly monocots were planted every other row; the alternate rows were disced to reduce pocket gopher habitat. Drip irrigation was centered underneath the vines to supply water. This forced most pocket gopher activity into these irrigation zones and allowed us to focus most of our treatment efforts in these areas, although rodenticide applications were made outside these irrigation zones when active burrow systems were present in those areas.

At each study site, we established five treatment blocks. Each block was approximately 1.0 ha in size and square in shape. In the interior of each block, we established a 0.4 ha treatment plot. Within each treatment plot, we established a three by three grid structure of monitoring plots, with each monitoring plot 9.1×9.1 m in size. These monitoring plots were separated by 18.2 m, allowing us to fit all nine into a 0.4 ha area. The outer 0.6 ha served as a buffer zone to help limit movement of pocket gophers into the monitoring area.

Four treatments and a control (i.e. no rodenticide application) were randomly assigned to each treatment block at each study site, with all treatment and control blocks contiguous at each study site. The rodenticides we tested included three experimental pelletized baits (C + D, C + B1, C + B2) and a grain bait that consisted of wheat, milo and oats coated with strychnine (active ingredient concentration 0.5%; Avalon Gopher Grain Bait, RCO International, Inc., Harrisburg, OR). We applied rodenticide baits using the funnel-and-spoon method. For this approach, we used a long screwdriver to probe into tunnel systems. Once a tunnel system was found, we removed the probe and inserted a funnel into the open hole. We then applied the appropriate amount of bait using a 14.8 cm³ (approximately 10–11 g; C + D, C + B1, C + B2) or 4.9 cm³ (approximately 5 g; strychnine) measuring spoon. Once the rodenticide was applied, we used a small piece of toilet paper to plug the hole, and then covered the plug with soil to eliminate light from the tunnel system. Occasionally, the soil around the probe hole collapsed, creating a large opening in the tunnel. When this happened, we applied bait to both sides of the tunnel system and covered the openings with soil. Rodenticides were applied an estimated 1-3 times per burrow system, depending on the size of the burrow system. This increased the likelihood that a pocket gopher would encounter the rodenticide at some point within its burrow system. Each application was marked with a wire flag so that we could count the number of applications post-treatment. We also weighed the amount of rodenticide applied per treatment block to determine the total weight of rodenticide applied per treatment area. This allowed us to determine the mean weight of rodenticide

applied per application to ensure there was limited variation in the amount of rodenticide deposited per application. All rodenticide applications occurred between 19 June and 30 July 2015.

We applied rodenticide baits throughout the treatment plot and for approximately 9.1 m on all sides of the treatment plot. Treatments throughout the entire buffer zone would have been preferred, but we did not have sufficient labor to achieve this application design given the high density of pocket gophers within the study sites. We anticipated that the area of the buffer zone we treated would be sufficient to eliminate immediate reinvasion of the treatment plot, but acknowledge that some reinvasion could have occurred. However, if such reinvasion did occur, it would bias our efficacy estimates low, so in effect our efficacy estimates could be somewhat conservative.

To assess efficacy, we compared activity in monitoring plots before and after treatment applications using the open-hole method.^{17,18} Following this approach, we opened holes into two tunnels within each monitoring plot when possible. We made note of plots where one or fewer tunnels were available. We then checked monitoring plots 2 days after holes were opened, and recorded whether holes were plugged or remained unplugged; because pocket gophers maintain closed burrow systems, they will plug any holes that open into tunnels if they are present. We initiated bait application following completion of the monitoring process. We repeated the monitoring process post-treatment, with holes in C+D, C+B1 and C+B2 plots opened 14-17 days after rodenticide application was completed, while strychnine plots were opened 9–19 days post-treatment. These timeframes were deemed sufficient to access mortality given recent lab trials that determined time-to-death for these rodenticides \overline{x} time-to-death = 5.3, 6.4, 10.8 and 1.0 days for C + D, C + B1, C + B2 and strychnine respectively; Witmer G and Baldwin R, unpublished, 2014). Following this second monitoring period, we again applied rodenticide to all treatment plots unless previous monitoring indicated 100% efficacy of that rodenticide. If we observed no remaining pocket gopher activity in a plot post-treatment (i.e. 100% efficacy), we did not apply the rodenticide a second time. We conducted a final monitoring period again using the open-hole method post-treatment. We calculated efficacy by dividing the number of plots that had no pocket gopher activity post-treatment by the number of plots that had pocket gopher activity pretreatment; this proportion was multiplied by 100 to provide a percentage efficacy value. We considered a rodenticide to be effective for pocket gopher control if it achieved a minimum threshold of 70% reduction in activity after two treatments.¹⁹ We used one-sample t-tests to observe whether our post-treatment efficacy values differed from 70%.²⁰ Because sample sizes were limited (n = 3) owing to restrictions on the size of treatment areas when testing unregistered pesticides, we used $\alpha = 0.1$ to determine significant differences, but we also calculated effect sizes (Cohen's d) to determine the magnitude of the difference irrespective of sample size. A Cohen's d > 0.8 is generally considered to be a large effect.²¹ All aspects of this study were approved by the University of California, Davis, Institutional Animal Care and Use Committee (protocol no. 18637).

3 RESULTS

The number of applications per treatment block varied extensively (\bar{x} range = 82–396) across fields and between first and second applications owing to differences in density of pocket gophers in each field and the variable efficacy of each rodenticide

following the initial application (i.e. high efficacy meant fewer second-treatment applications). The mean weight of cholecalciferol plus anticoagulant rodenticide used per application varied slightly across treatment sites and periods, in part owing to inconsistent filling of the measuring spoon given the large size of the pellets. Nonetheless, the mean weight of rodenticide deposited per application was strongly consistent across the cholecalciferol plus anticoagulant rodenticides (\overline{x} range = 11.5–11.8 g). The weight of strychnine deposited per application was strongly consistent across sites and applications (\overline{x} range 4.9–5.1 g), presumably owing to the small size of grain fitting within the measuring spoon more precisely than with larger pellet sizes. Additional detail on the number of applications and weight of each rodenticide applied can be found in the supporting information.

We did not observe any rodenticides exhibiting efficacy significantly >70% after a single application (\overline{x} range = 46-79%, $t_2 \leq 0.9$, $P \geq 0.464$) (Table 1). Following a second application, strychnine ($\overline{x} = 100\%$, $t_2 = \infty$, P < 0.001, Cohen's $d = \infty$), C + D $(\bar{x} = 83\%, t_2 = 3.1, P = 0.089$, Cohen's d = 1.814) and C + B2 ($\bar{x} =$ 75%, $t_2 = \infty$, P < 0.001, Cohen's $d = \infty$) were significantly above the EPA-mandated efficacy level of 70% (Table 1). Although mean efficacy of C + B1 was not statistically >70% ($t_2 = 1.6$, P = 0.255) (Table 1), we still observed relatively high efficacy ($\overline{x} = 85\%$). This lack of significance was driven by moderate variability in efficacy across fields (SD = 17). Nonetheless, effect size was large (Cohen's d = 0.896),²¹ indicating that additional testing is justified for this lower-concentration rodenticide. Control blocks generally showed a slight increase in activity, indicating that the observed reduction in pocket gopher activity was due to the applied treatments (Table 1).

4 **DISCUSSION**

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Both strychnine and cholecalciferol plus anticoagulant rodenticides proved effective at reducing pocket gopher populations in vineyards. Although our results with strychnine were similar to those observed in some past studies,^{8,22-24} other investigations have not shown strychnine bait application to be an effective management option.^{7,9} Reasons for this disparity are unclear, but it could be due to differences in the concentration of the active ingredient applied, the bait used to carry the active ingredient (e.g. wheat, milo, oats and pellets), variability in bait acceptance across different species of pocket gophers, variability in crops where the bait was applied and the ability of the applicator to place the rodenticide into an active tunnel system.²⁵ Alternatively, continual reliance on strychnine for pocket gopher management can lead to behavioral or physiological resistance to the toxicant where individuals within the population learn to either avoid or consume sublethal amounts of the rodenticide.^{3,26,27} We observed this same response in a related captive study of potential rodenticides (Witmer G and Baldwin R, unpublished, 2014); this highlights the need to utilize an integrated approach when managing pocket gophers rather than relying on strychnine as the sole method for pest control.

It is worth noting that, although efficacy from the initial strychnine application did not differ significantly from 70%, the actual mean efficacy was above the 70% threshold ($\bar{x} = 79\%$) (Table 1). Pocket gophers are fossorial rodents that actively tunnel throughout the year. However, they do not create mounds on a daily basis. In some situations, it can be several days or more between mounding activities for some pocket gophers within a population.^{28,29} No removal activities (e.g. rodenticides, burrow fumigants, traps) are **Table 1.** Percentage efficacy of four different rodenticides [0.03% cholecalciferol plus 0.005% diphacinone [C + D], 0.015% cholecalciferol plus 0.0025% brodifacoum [C + B2] and 0.5% strychnine (Strych)] plus a control (Cont) for pocket gopher management across two different treatment periods. Rodenticide applications occurred in summer 2015 across three fields in San Joaquin County, California. Mean and standard deviation (SD) values are provided for comparative purposes

	First treatment					Second treatment				
	C + D (%)	C + B1 (%)	C + B2 (%)	Strych (%)	Cont (%)	C + D (%)	C + B1 (%)	C + B2 (%)	Strych (%)	Cont (%
Field 1	75	33	50	71	0	75	67	75	100	25
Field 2	56	44	25	67	0	89	89	75	100	0
Field 3	14	100	63	100	-20	86	100	75	100	-40
Mean	48	59	46	79	-7	83 ^a	85	75 ^a	100 ^a	-5
SD	31	36	19	18	12	7	17	0	0	33

effective if they are not applied within an active tunnel system. As such, repeat applications are often needed to maximize exposure of all individuals in a population. The fact that we observed such high efficacy for pocket gophers after a single application of strychnine suggests that it can continue to be one of the more effective tools for pocket gopher management as long as it is just one part of an IPM program.

One way to mitigate the potential for behavioral or physiological resistance to strychnine is to rotate rodenticide applications with another toxicant. Additionally, effective alternative toxicants would be greatly beneficial given the current limited availability of strychnine products in the United States. Although both C+D and C + B2 were effective alternatives, the C + D product has the advantage of utilizing a first-generation anticoagulant as the synergist to cholecalciferol. Diphacinone is much less toxic and has a substantially shorter half-life in tissues when compared with brodifacoum.¹¹ Therefore, it should pose less risk of secondary toxicity than C + B2 to non-target scavengers and predators. That being said, pocket gophers are strongly fossorial rodents that spend relatively little time above ground.¹⁶ As such, secondary exposure should be fairly minimal, regardless of the toxicant used. Additionally, C+B2 contains half the toxicant normally included in brodifacoum baits, so this should substantially lessen risks as well. We did test a lower-concentration cholecalciferol (0.015%) plus brodifacoum (0.00125%) rodenticide in a laboratory setting, but we did not find it to be as effective (efficacy 60%; Witmer G and Baldwin R, unpublished, 2014). It is possible that a rodenticide bait that contained 0.03% cholecalciferol plus 0.00125% brodifacoum might be more effective, but this has yet to be tested.

We should note that, although the C+B1 combination rodenticide did not differ from 70%, it did yield a greater mean efficacy value ($\overline{x} = 85\%$) than any of the other combination products tested (Table 1). Further testing may prove this combination to be an effective option as well, which would be beneficial given the lower concentration of cholecalciferol (0.015%) when compared with the other tested combination rodenticides (0.03%). It should be pointed out that this study was conducted in a vineyard setting. Given the similarity in available food sources between tree and vine crops, we believe that our observed results will likely be comparable across these crops. However, results may be different in grasslands and forage crops given greater abundance of alternative and highly preferred food sources. Further testing is certainly warranted in these settings. Nonetheless, a growing body of literature exists highlighting the efficacy and potential benefits of cholecalciferol plus anticoagulant toxicants (Witmer G and Baldwin R, unpublished, 2014).^{11–13} These toxicants seem to exhibit great promise as an effective tool in the proverbial IPM toolbox.

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SUPPORTING INFORMATION

Supporting information may be found in the online version of this article.

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