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Development of a New Deer Repellent for the Protection of Forest Resources

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ABSTRACT: We have identified hydrolyzed casein as a promising repellent for minimizing damage to forest resources inflicted by browsing ungulates. Eight and twelve percent hydrolyzed casein formulations prepared in water with a latex-based agricultural sticker significantly reduced browse damage by captive black-tailed deer (*Odocoileus hemionus columbianus*) to western redcedar (*Thuja plicata*) saplings. These repellent formulations can be prepared by the user at significant cost savings versus commercial products. *West. J. Appl. For.* 21(2): 108–111.

Key Words: Black-tailed deer, browsing, *Odocoileus hemionus columbianus*, reforestation, wildlife damage, wildlife management.

Largely because of deer management strategies and increased forage availability, deer populations have rapidly increased throughout North America throughout the past decades (Cote et al. 2004). Overabundance has contributed to significant economic losses in transportation, agriculture, and forestry, as well as to transmission of disease. Deer damage to agriculture has been recognized as a substantial economic problem for some time (Wywiałowski 1998). In the Pacific northwestern United States, damage to forest resources by black-tailed deer is considered a significant impediment to reforestation efforts (Nolte and Dykzeul 2002). Increased agricultural losses because of deer browse have also been reported in Europe (Santilli et al. 2004).

Browse damage can be lethal to plants, while also reducing the future value of crops via decreased yields and plant deformities (Nolte 1998). However, browse damage is not limited to commercial agriculture and reforestation efforts. Overabundant deer may have a significant impact on plant community structure and ecosystem properties (Cote et al. 2004). High rates of deer browsing suggest the possible extinction of valuable forest understory herbs (Mcgraw and Furedi 2005).

A number of commercially available products are marketed to deter browsing of trees and shrubs by deer. These products contain a broad range of presumed active ingredients—some more effective than others (Nolte and Wagner 2000). The majority of these products are contact

repellents that must be applied directly onto the plants to be effective. Among contact repellents, four different modes of action have been proposed: flavor aversion learning, taste modification, chemical irritation, and fear (Nolte and Wagner 2000).

We recently demonstrated that a number of methionine-containing proteins minimize browsing by making treated plants less palatable to deer (Kimball and Nolte 2005). Among these, casein has the potential for commercial use as a deer repellent. Here, we describe three experiments conducted to evaluate several protein sources as repellents for protecting conifers from deer browse damage. We sought to develop a new repellent formulation that effectively minimized browse damage, was easy to prepare in water, and was relatively inexpensive versus commercial repellent products.

Materials and Methods

Treatments

Baker's soy flour was provided by Archer Daniels Midland Co. (Decatur, IL). Egg albumen was provided by Belovo, Inc. (Pinehurst, NC). Complete milk protein (CMP), edible acid casein (EAC), and hydrolyzed casein (HC) were purchased from American Casein Co. (Burlington, NJ). Methionine was purchased from Aldrich Chemical Co. (Milwaukee, WI) and hydrated lime (Kemilime) from Ash Grove Cement Co. (Overland Park, KS). Methionine was added to lime to yield a test treatment that was 5% methionine for experiment 1. Deer-Away Big Game Repellent powder (BGR-P; IntAgra, Inc., Minneapolis, MN) was used as a positive control in experiment 2. Ready-to-use premixed Plantskydd Deer Repellent (Tree World, Inc., Des Moines, IA) was used as the positive control in experiment

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3. The agricultural sticker Tactic (Loveland Products, Greeley, CO) used to adhere treatments to the conifer seedlings in all experiments was provided by the manufacturer.

Deer and Facilities

Forty-eight captive 1- to 3-year old, black-tailed deer (*Odocoileus hemionus columbianus*) were used for the bioassays conducted in 0.2 ha outdoor pens. The same 24 deer were used in experiments 1 and 2; whereas 24 different deer were used in experiment 3. Shelter and ad libitum pelleted basal ration (USDA Deer Pellet; X-Cel Feeds, Tacoma, WA), water, and mineral block were provided throughout each experiment. Naturally occurring forage in the pens was limited to an assortment of cool season grasses. The experiments were conducted February 2004 to February 2005 and approved by the Institutional Animal Care and Use Committee of the USDA National Wildlife Research Center.

Experiment 1

In each of eight 0.2-ha pens, western redcedar (*Thuja plicata*) saplings (~60 cm) were planted in five unique plots consisting of 12 trees per plot arranged in a 3 × 4 arrangement with 1 meter spacing. To assure independence of treatments, plots were separated by at least 3 meters and treatments were randomly assigned (each treatment represented by one plot in each pen). Treatments were applied by spraying individual saplings uniformly with 0.054% (v/v) Tactic solution in tap water with a tank-type garden sprayer. The wetted saplings were immediately dusted by hand with the appropriate powdered treatment. The four methionine-containing treatments were: egg albumen, CMP, 5% methionine in lime, and Baker's soy flour. Lime was included in the experiment as a control.

Three deer were confined to each pen for the duration of the experiment and provided ad libitum access to test trees for 22 days. The number of bites on each tree were recorded on days 1, 2, 3, 4, 5, 7, 8, 12, 16, 19, and 22, or until the individual tree was completely consumed (defined as 25 cumulative bites) according to previously established procedures (Nolte 1998). Severe browse damage was defined as 10 cumulative bites to an individual tree. Experiment 1 was conducted from Feb. 2 to 27, 2004.

Experiment 2

Experiment 2 was similarly conducted with five plots replicated in eight pens except that each plot consisted of nine western redcedar saplings in a 3 × 3 array. The five treatments consisted of three casein-related sources: CMP, EAC, and HC, a positive control (BGR-P), and a control [0.054% (v/v) Tactic spray only]. Three deer were confined to each of eight pens for the 16 days of the experiment. The number of bites on each tree were recorded on days 1, 2, 3, 6, 9, 13, and 16, or until the individual tree was completely consumed. Experiment 2 was conducted from May 26 to June 11, 2004.

Experiment 3

The final experiment was similarly conducted with six plots (12 western redcedar saplings per plot in 3 × 4 array) in each of eight 0.2-ha pens. The six treatments used in experiment 3 consisted of four HC solutions, Plantskydd,

and a control (Tactic only). For this experiment, the sticker was mixed with tap water at a concentration of 0.26% (v/v). The four HC solutions were prepared by adding the powder to the sticker solution to yield HC concentrations of 2, 4, 8, and 12% (w/v).

Treatments were applied to the saplings by handheld spray bottle. During application, occasional shaking of the container was required to keep HC suspended in the 8 and 12% solutions. Three deer were confined to each pen for the duration of the experiment and provided ad libitum access to test trees for 21 days. The number of bites on each tree were recorded on days 1, 2, 3, 4, 7, 9, 14, and 21, or until the individual tree was completely consumed. Experiment 3 was conducted from Jan. 27 to Feb. 16, 2005.

Statistical Analyses

For each experiment, a Kaplan-Meier survival analysis was performed to compare survivability distribution functions among treatments, using the Wilcoxon test of equality (PROC LIFETEST; SAS/STAT 2002, SAS/STAT version 9.1; SAS Institute Inc., Cary, NC). Failure day was defined as the first experimental day when severe browse (10 cumulative bites) was measured on an individual tree. Trees that survived to the end of the experiment (did not meet definition of failure) were assigned a failure day of 25 and censored according to the standard application of survival analysis.

Failure data were also analyzed by ranking failure day among treatments in each pen (1 = shortest failure day) and subjecting the rank data to Kruskal-Wallis analysis (Iman 1982). Rank was the response for the nonparametric analysis with treatment the fixed effect. Multiple comparisons of mean ranks were made using Fisher's least significant difference (LSD option; SAS/STAT 2002). A separate analysis was conducted for each of the three experiments.

Results

Survivability functions differed significantly among treatments in all experiments ($P < 0.0001$; Figures 1, 2, and

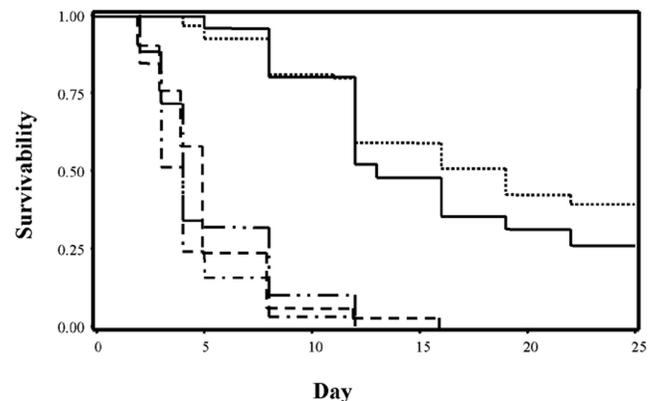


Figure 1. Survivability functions for western redcedar (*Thuja plicata*) saplings treated with three different protein sources, 5% free methionine in lime, and a control (lime only) in experiment 1. Deer had ad libitum access to the treated saplings for 22 days. , complete milk protein; _____, albumen; ----, soy; - . - . , free methionine; - - - , lime (control).

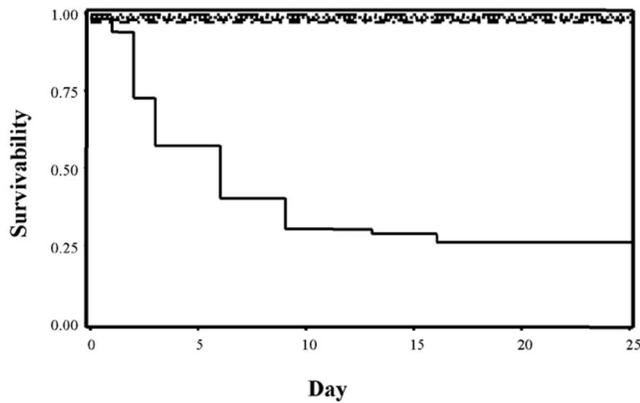


Figure 2. Survivability functions for western redcedar (*Thuja plicata*) saplings treated with three different sources of casein, Deer Away Big Game Repellent powder (BGR-P), and a control (latex sticker only) in experiment 2. Deer had ad libitum access to the treated saplings for 16 days., complete milk protein (CMP); - - -, edible acid casein (EAC);, hydrolyzed casein (HC); - - -, BGR-P (positive control); ____ control.

3). In experiment 1, all trees treated with Baker's soy flour, 5% methionine, or lime were severely browsed (defined as 10 bites) by day 16 of the experiment (Figure 1). Forty percent of trees treated with CMP and 26% of trees treated with albumen were not severely browsed by the end of the experiment. Kruskal-Wallis analysis of the ranked data in experiment 1 demonstrated a significant treatment effect ($P < 0.0001$) with multiple comparisons of the means indicating that failure day rank followed the order: CMP = albumen > soy = 5% methionine > lime.

Kruskal-Wallis analysis of the ranked data from experiment 2 demonstrated that CMP, EAC, HC, and BGR-P were each effective in minimizing browse with respect to the control ($P < 0.0001$). One hundred percent of trees treated with the casein sources or BGR-P were protected from browse damage (Figure 2). Conversely, 71% of control trees were severely browsed by the end of the 16-day experiment.

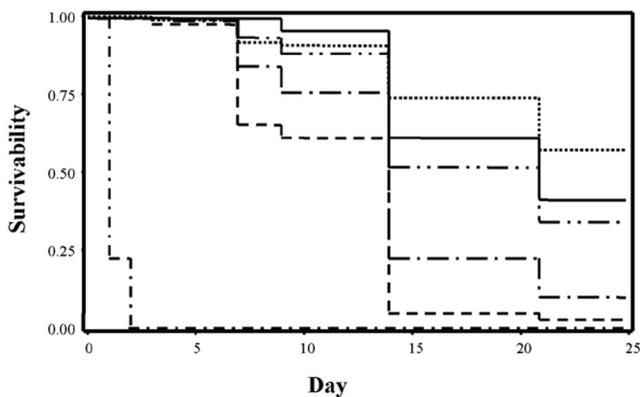


Figure 3. Survivability functions for western redcedar (*Thuja plicata*) saplings treated with four concentrations of HC, Plantskydd, and a control (latex sticker only) in experiment 3. Deer had ad libitum access to the treated saplings for 21 days., Plantskydd; ____ 12% HC;, 8% HC; - - -, 4% HC; - - -, 2% HC; - - -, control.

The results of experiment 3 established that deer avoidance of HC-treated trees was concentration-dependent (Figure 3). Kruskal-Wallis analysis of the ranked data demonstrated a significant treatment effect ($P < 0.0001$) with multiple comparisons of the means indicating failure day rank followed the order: Plantskydd = 12% HC = 8% HC > 4% HC > 2% HC > control.

Discussion

In each experiment, treatments were visibly apparent as white- or cream-colored powder adhering to unbrowsed foliage throughout the tests. There was no indication that plot assignment, or proximity of a treatment plot to another, impacted relative preference for the treatments. This is consistent with the observation that deer repellents such as BGR-P and Plantskydd have "aversive distances" of less than 1 meter (Nolte and Wagner 2000). This is the distance from a repellent-treated food source that deer will avoid an untreated test food. In practice, the aversive distance of contact repellents is typically 0 meters, which is why reapplication is frequently necessary to protect new growth. Therefore, 3 meters was considered sufficient distance between plots to avoid confounding effects of treatment interaction in these experiments.

The treatments used in experiment 1 were chosen because they each contain methionine. Many proteins do not contain methionine or are methionine-limited (Friedman 1996). For example, porcine collagen (gelatin) contains little methionine and was not avoided by deer (Kimball and Nolte 2005). Among experiment 1 treatments, CMP and albumen contain approximately six times more protein-bound methionine than Baker's soy. Previous chemical analyses demonstrated that methionine was protein-bound (i.e., not present as the free amino acid) in these sources (Kimball and Nolte 2005). The proteins used in these experiments all contained less than 5% methionine.

Experiment 1 confirmed that CMP and albumen were more effective repellents than soy flour, which has low methionine content. More importantly, results of experiment 1 suggest that proteins with protein-bound methionine were more effective than the free amino acid. Although Kruskal-Wallis analysis established that the 5% methionine treatment was more effective than the control (lime only), it was no more effective in reducing browse damage than the soy treatment that has low levels of protein-bound methionine. Additionally, consumption of the lime control in experiment 1 indicated that avoidance of protein treatments did not result from simple tactile or visual cues.

Experiment 2 was designed to compare the repellency of three casein sources versus a positive control. CMP contains all protein fractions present in skim milk (whey and casein). EAC is the casein fraction of skim milk. HC is the enzymatic digest of casein. Enzymatic hydrolysis yields small peptides and free amino acids from the intact proteins. BGR-P was chosen as the positive control for this experiment because it is a powder that could be delivered in the same manner as the casein treatments. Furthermore, it is a contact repellent with proven efficacy in bioassays with

captive deer (Nolte and Wagner 2000). Activity of BGR-P is attributed to short-chained aliphatic aldehydes produced by auto-oxidation of egg lipids (Oita et al. 1977). The results of experiment 2 demonstrated that CMP, EAC, and HC were each as effective as the positive control.

The results of experiments 1 and 2 indicated that methionine-containing proteins as well as hydrolysates of methionine-containing proteins are potentially effective deer repellents that warrant further investigation. They further suggest that of the numerous products of casein hydrolysis, the active ingredient(s) are probably small peptides containing methionine—not methionine present as the free amino acid. The decision to focus on HC for the development of a new repellent was based on the assumption that its water solubility would be greater than intact proteins as a consequence of the hydrolysis process. The choice of HC was further justified when it was demonstrated that white-tailed deer (*Odocoileus virginianus*) avoided HC-treated food, but not EAC-treated food in one-choice feeding trials after food deprivation (Kimball et al. 2005).

Experiment 3 was conducted to determine the effective HC concentration required to minimize deer browsing. Ready-to-use, premixed Plantskydd was used as the positive control for this experiment because the liquid could be applied to the test trees in identical manner as the HC formulations. Plantskydd (active ingredient dried blood) has also exhibited proven efficacy as a contact repellent in bioassays with captive deer (Nolte and Wagner 2000). The efficacy of the HC treatments was directly proportional to HC concentration. Plantskydd, 8% HC, and 12% HC all effectively reduced deer browsing of a preferred conifer species throughout the 3-week period of the experiment.

Browse damage to saplings treated with Plantskydd, BGR-P, and various casein sources varied among the three experiments. For example, not a single tree treated with CMP was severely browsed in experiment 2. Conversely, 60% of trees treated with CMP were severely browsed in experiment 1. The motivation of herbivores to use a particular resource is subject to many variables, including experience with the food, nutritional state, and food alternatives (Provenza 1995). This is true for deer browsing of agricultural resources in natural systems as well as the experiments described here. It is unlikely that the deer's motivation to browse test trees was consistent among experiments. Accordingly, the results from each experiment must be considered independently. Any comparisons among the three experiments can only be made with respect to the control treatments.

These experiments indicate that HC is an effective repellent for reducing browse damage to forest resources. Specifically, a liquid formulation consisting of 8% or 12% HC with 0.26% latex-based sticker shows great promise for operational use. Although this formulation offers no advantages versus commercially available products with respect

to labor investments (it must be delivered to seedlings in the same manner as the commercial products), potential savings in material costs are significant.

At the time of publication, a 12% HC formulation would cost approximately \$6 USD per 4.0 L in total material costs. Four liters would be capable of treating ~500 30-cm seedlings. This price is based on the current retail price for the latex sticker and the cost of HC when purchased in bulk (e.g., 900 Kg pallet). The price per equivalent volume when HC is purchased in smaller quantities (e.g., 22 Kg bags) would be approximately \$8 USD. The cost of an HC repellent formulation could be significantly reduced by using an 8% HC formulation, which was found to be as effective as the 12% formulation in these bioassays. However, because the price of HC is subject to worldwide milk stores and economics, the price of a HC repellent formulation could fluctuate proportionally. By comparison, commercial deer repellent products purchased in bulk or concentrate typically cost between \$15 and \$25 USD per equivalent coverage.

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