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ORAL RABIES VACCINE (ORV) BAIT UPTAKE BY CAPTIVE STRIPED SKUNKS

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ABSTRACT: Aerial delivery of oral rabies vaccine (ORV) baits has proven effective in large-scale efforts to immunize wildlife against rabies, and in North America this strategy currently is being used to immunize foxes (*Urocyon cinereoargenteus* and *Vulpes vulpes*), raccoons (*Procyon lotor*), and coyotes (*Canis latrans*). Skunks are also a major reservoir and vector of rabies, but at present oral vaccines for use in skunks are not licensed. Furthermore, given differences in morphology (smaller jaws) and behavior (food handling and consumption), it is unknown if baits currently used in ORV campaigns would be effective for skunks. Because oral vaccine delivery is contingent upon puncture of the vaccine container (VC), baits need to be sufficiently attractive to elicit selection and consumption. Manipulation of the bait to facilitate vaccine ingestion by the target species is a critical element for an effective ORV bait. The objectives of this study were to assess manipulation and consumption of current ORV baits by striped skunks (*Mephitis mephitis*). We conducted four independent trials with penned animals and various baits to assess bait selection frequency, VC puncture frequency, and consumption. Video recorded trials were used to assess attractiveness of baits and consumption behavior of skunks. Bait characteristics, such as texture, size, and flavor influenced selection and consumption. Fish and chicken flavors were preferred and vaccine containers within selected baits were likely to be punctured. Vaccine ingestion seemed more likely if VCs were directly coated with the bait matrix. To make baits attractive to skunks and to ensure puncture of the VC, modifications to current baits should consider a smaller size, a meat-flavored matrix, a slightly pressurized VC, and a direct coating of matrix on the VC.

Key words: Bait, consumption, manipulation, *Mephitis mephitis*, oral rabies vaccine (ORV), rabies, skunk, wildlife damage management.

INTRODUCTION

In the USA, more than 92% of reported rabies cases occur in wildlife, primarily in carnivores and bats (Krebs et al., 2005). In 2004, most rabies cases reported to the Centers for Disease Control and Prevention (CDC) occurred in raccoons (*Procyon lotor*; 38%), skunks (primarily *Mephitis mephitis*; 27%), and bats (Order Chiroptera; 20%; Krebs et al., 2005). Other wildlife hosts for rabies virus include gray fox (*Urocyon cinereoargenteus*), red fox (*Vulpes vulpes*), and coyote (*Canis latrans*). Skunk rabies has the broadest geographical distribution of all terrestrial rabies virus strains in the USA (Krebs et al., 2005). Skunks are also susceptible to raccoon, fox, and bat strains of rabies virus (Krebs et al., 2002), which makes skunks a versatile host of the virus. The number of skunks infected with raccoon rabies virus is rising and it is possible that skunks could independently maintain this rabies virus variant; this poses a potentially serious rabies control challenge (Guerra et al., 2003). In southern Ontario, skunks also may maintain the arctic fox rabies virus variant that currently is affecting red foxes (Nadin-Davis et al., 1999).

The primary method to control wildlife rabies on a large scale is aerial distribution of rabies vaccine–laden baits. Oral rabies vaccine (ORV) sealed in containers inside food-based baits are used for immunizing foxes (Wandeler, 1991), raccoons (Hanlon et al., 1989; Olson et al., 2000) and coyotes (Fearneyhough et al., 1998). The only ORV approved for use in the USA, Raboral V-RG® (Merial Limited, Athens, Georgia, USA), has not been shown to immunize skunks effectively. However, protection was reported in skunks vaccinated with high-titer vaccinia recombinant virus (five of six skunks survived the 90-day challenge; Tolson et al., 1987) and new vaccines for skunks are in development (Hanlon et al., 2002; Vos et al., 2002).

Trap–vaccinate–release (TVR) programs have been reported to slow the spread of skunk rabies. In TVR programs, skunks are live-trapped, injected intramuscularly with inactivated rabies vaccine, and released (Rosatte et al., 1987, 1990a). Though effective, TVR efforts are extremely labor-intensive and expensive (Rosatte et al., 1992). Without an effective and economical vaccine delivery method for skunks, the containment and elimination of skunk rabies appears unlikely.

A need exists for an ORV bait for skunks similar to those used for foxes, coyotes, and raccoons. Literature on skunks as nontarget consumers of ORV baits is limited. Although they will chew on baits intended for other species (Bachmann et al., 1990; Rosatte et al., 1990b), indicating the potential for effective ORV delivery, low bait uptake by skunks suggests that current baits may be ineffective. Information gained in studies on the consumption and bait manipulation by coyotes (Linhart et al., 1994; Farry et al., 1998; Steelman et al., 1998), foxes (Steelman et al., 1998; Winkler and Baer, 1976), and raccoons (Hable et al., 1992; Linhart et al., 2002) has been used to optimize bait design and distribution strategies for these target species, and similar information is needed for skunks. A bait that is difficult to handle, or allows the skunk to separate the vaccine container (VC) from the bait matrix, would not be effective without puncture of the VC and subsequent contact of an adequate dose of vaccine with the oropharyngeal mucosa. The objectives of this study were to assess manipulation and consumption of current ORV baits by striped skunks (*Mephitis mephitis*): 1) by determining selection frequencies for different bait types by skunks; 2) by determining puncture frequency of VCs; 3) by determining the proportion of bait consumption; and 4) by describing bait consumption behavior.

MATERIALS AND METHODS

Animal collection

Twenty-four striped skunks of mixed sex and age were trapped (Model TLT204, Tomahawk Live Trap Company, Tomahawk, Wisconsin, USA) in urban and rural Fort Collins, Colorado (40°35′N, 105°05′W) from 24 June to 6 August 2003. Skunks were immobilized by hand injection with a 5:1 ratio of Ketamine (100 mg/ml)/Xylazine (100 mg/ml) (MWI Veterinary Supply, Meridian, Idaho, USA) at 15 mg/kg. Under anesthesia, skunks were weighed, sexed, ear-tagged (Model 1005-1, National Band and Tag Company, Newport, Kentucky, USA), and scent gland papillae were ligated (Eastland, 1987). Skunks were transported to the National Wildlife Research Center, Fort Collins, Colorado, USA, placed in individual outdoor pens, and monitored daily. Skunks were held in quarantine for >1 mo, during which time they were offered 300 g/day of Mazuri® Omnivore Zoo Feed "A" (PMI Nutrition International, LLC, Brentwood, Missouri, USA). Water was available *ad libitum*.

Design

Four independent cafeteria-style bait trials (5 days/trial) were conducted between 22 September and 17 October 2003. In these, 24 skunks were offered the same suite of baits. Baits contained VCs filled with distilled water, and were offered in a specific order on a round ceramic plate near the food and water bowls in each pen. The order of baits on the plates was consistent throughout each trial, but the order was rotated daily to account for potential bias associated with the approach path by skunks. For 3 days prior to each trial, sardines were offered on the plate to increase the skunk interest and reduce neophobia. The amount of maintenance feed also was reduced (15–20 g) during trials to increase hunger. Following each test, skunks were given approximately 200–250 g of maintenance feed until the next day's test. Skunks were not allowed access to their den boxes during the test period, which lasted 1.5 hr/day (8:30–10:00 AM). Bait selection, VC puncture, and consumption were video-recorded and data recorded as described below.

Baits

Baits were supplied by commercial sources and consisted of baits used in current ORV campaigns as well as baits with slight modifications in size or flavors. All baits were

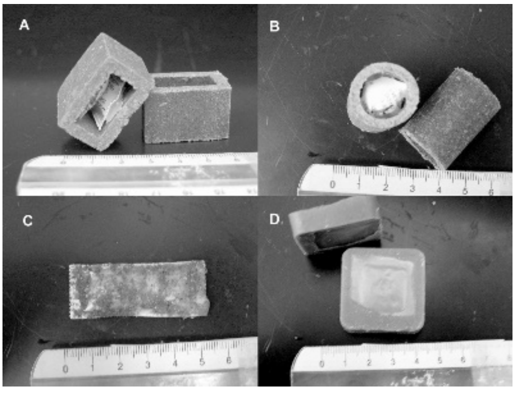

FIGURE 1. Baits used in ORV trials: (A) polymer meal standard rectangle (left) and short rectangle (right); (B) polymer meal cylinder; (C) coated sachet, and (D) Ontario slim.

formulated with 1% tetracycline hydrochloride (approximately 7–10 mg/g of bait), a standard biomarker in ORV baits. Polymer meal bait (Bait-Tek, Inc., Orange, Texas, USA) flavors included fish, chicken, cat food, and alligator bait (formulated to attract alligators). Baits were made as previously described (Linhart et al., 2002) and were extruded into three shapes (standard rectangle, shorter rectangle, and cylinder; Fig. 1A, B). The fishmeal standard rectangle bait is currently used in raccoon and fox ORV programs in the USA. The VC was a sachet similar to a condiment packet (Merial Limited, Athens, Georgia, USA); this was fitted into the hollow core of the bait and held in place with paraffin wax.

Coated sachet baits were prepared and supplied by Merial Limited. Sachets used for these baits were identical to those used with polymer meal baits, but were coated with fish essence paraffin wax and rolled in fish crumbles (Fig. 1C; Linhart et al., 2002). The process created a direct coating of the bait matrix on the VC. Fish was the only flavor used on coated sachets.

Ontario "slim" baits (Artemis Technologies, Inc., Guelph, Ontario, Canada) were a slimmer version of their predecessor, the Ontario bait (Linhart et al., 1997). Slim baits were sugar- and shortening-based with a waxy consistency, and flavors included apple, chicken, seafood, sugar, and cherry. Sugar slims are the standard bait used in current ORV campaigns, primarily in Canada. The VC was called a blister pack, which consists of a plastic reservoir covered and sealed with foil (Fig. 1D).

Bait manipulation

Video cameras (Sony Handicams CCD-TRV 318 Hi 8 and DCR-TRV 350 digital cameras) were used to: 1) observe which baits were the first to be examined and the first to be selected (the attractiveness of baits); and 2) describe skunk consumption behavior (posture, mastication, and handling of baits and VCs). Examination of baits was qualified by a direct sniff or lick, bitten but not broken, or picked up or fumbled. Selection was based on biting into (either breaking or consuming) the bait. Also noted was the handling style and angle used in manipulating each bait type, and any difficulties in manipulation or ingestion. Although difficult to quantify, these behaviors were important in the interpretation of the efficiency with which a skunk could manipulate the bait, and whether the bait performed as it should while being handled. Seven hundred twenty hours of video footage were viewed, and technicians who were trained to use the same methods, criteria, and vocabulary recorded data. Excel spreadsheets were used to generate and graph descriptive statistics.

Consumption trials

For each trial, we measured selection frequency of bait types, puncture frequency of VCs within bait types, and the proportion of bait matrix consumed. Bait selection frequency and VC puncture frequency were based on binomial data (yes or no). Selection of the bait was indicated by bite marks or partial consumption. To determine consumption, we measured the amount (weight) of bait matrix consumed as a proportion of the original (pretrial) mass (not all baits were of equal mass). In order to remove any weight associated with the VC (which could not always be separated from the remaining bait matrix), VC remains were classified as full, three-quarters full, half full, one-quarter full, or empty, and assigned a corresponding weight that was subtracted from the amount of the remaining bait matrix.

In Trials 1 and 2, we offered four flavors of polymer baits in various shapes. Polymer baits were the only baits that could be made in various shapes, and thus, were the only baits used in these trials (Table 1). In Trial 3, we offered only fish-flavored baits in each of the bait types (Table 1). In Trial 4, we presented identically shaped Ontario slim baits in various flavors (Table 1).

Statistical analyses

Each trial was considered a separate experiment. For the consumption trials, a chi-

TABLE 1. Experimental design of oral rabies vaccine (ORV) placebo bait trials performed in captive striped skunks.

Trial	Bait type	Flavor	Shape
1	Polymer meal	Fish	Standard rectangle
		Cat food	Short rectangle
		Alligator bait	Cylinder
		Chicken	Cylinder
2	Polymer meal	Fish	Cylinder
		Cat food	Cylinder
		Alligator bait	Short rectangle
		Chicken	Short rectangle
3	Polymer meal	Fish	Standard rectangle
	Polymer meal	Fish	Cylinder
	Coated sachet	Fish	Standard
	Ontario slim	Seafood	Standard
4	Ontario slim	Seafood	Standard
		Apple	Standard
		Cherry	Standard
		Chicken	Standard
		Sugar	Standard

square test for equal proportions was conducted for selected baits without punctured VCs to determine if this outcome was a function of bait type ($\alpha=0.05$). In each trial, 120 baits of each type were offered. In a few cases, some VCs were not recovered after a test. In trials 1, 2, 3, and 4, 118, 120, 117, and 118 VCs were recovered, respectively. Missing data did not affect the chi-square analyses, and Type III sums of squares were used to account for these missing data.

Differences in VC puncture rates by trial day were analyzed by chi-square analysis. A $5\times2\times4$ cross tabulation table was created to represent day of trial (1–5), bait selection (yes or no), and bait type (four baits/trial except in Trial 4, when five baits were evaluated). A Cochran-Mantel Haenszel (CMH; SAS, 2002) statistic was used to measure associations between VC puncture and day of trial. Significant associations ($P\leq0.05$) were further investigated by examining VC puncture $\times$ day tables across bait types. To minimize error, a decision criterion of $P\leq0.012$ was used for evaluating multiple tables within a trial (Bonferonni adjustment). Differences in VC puncture due to bait type were also analyzed by chi-square analysis.

For each trial, the highest proportion of bait consumed was given a rank of 1 for a given skunk on a given day (skunk $\times$ day). A Kruskal-Wallis test was done on ranked data (with fixed effects of bait, day, and bait $\times$ day). Mean rankings for a given bait were compared by using Fisher's least significant difference (SAS, 2002).

RESULTS

Bait manipulation

Examination and selection: Occurrences of first-examined and first-selected baits were influenced by two factors, the direction from which a skunk approached the plate and the attractiveness of bait characteristics (odor, color, shape, texture, size). Routine approaches by skunks did not necessarily result in routine examination and selection relative to specific locations of the plate. In all trials, baits on the right side of the plate were most often the first to be examined; however, first-selected baits were selected relatively evenly from around the plate.

In both Trials 1 and 2, the fish standard rectangle and chicken cylinder polymer baits were most often the first to be examined and selected by skunks (Fig. 2). In Trial 3, the fish standard rectangle polymer bait was selected first most often and the seafood Ontario slims were most often the first to be examined (Fig. 2). Selection frequencies for the fish-coated sachet and the fish cylinder were similar. In Trial 4, there was very little interest in the Ontario slim baits overall; both examination and selection were low relative to Trials 1–3 (Fig. 2). Interestingly, the percentage of times in Trial 4 that the seafood slim was the first to be examined dropped by half from Trial 3.

Consumption behavior: Skunks typically held polymer baits vertically and ate them from the outer edges. During consumption, the baits often crumbled into smaller pieces. Rectangle polymer baits were typically consumed beginning at a corner, whereas the entire circumference of cylinder polymer baits was placed in the mouth (Fig. 3). During consumption, skunks either pulled out VCs or they fell out as

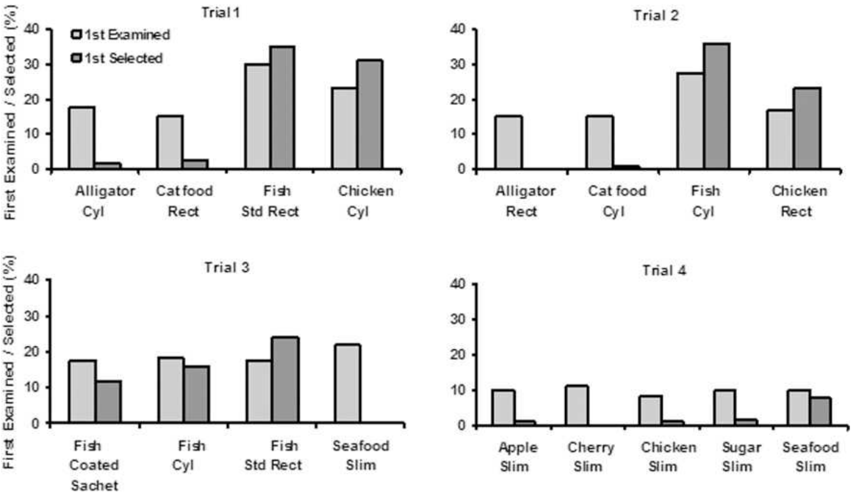

FIGURE 2. The percentage of occasions a bait type was the first to be examined and the first to be selected out of all the bait types offered, as observed from video recordings of consumption trials in four independent trials.

the bait matrix was eaten. There was no video evidence that showed an interest by skunks in the VC once it was separated from the bait matrix.

Skunks picked up fish-coated sachets, held them vertically, and bit into their ends, puncturing VCs, often biting off the ends. Coated sachets were either chewed thoroughly before being discarded or the entire sachet was completely ingested. Sometimes coated sachets were pinned to the ground with a forepaw and licked

FIGURE 3. Typical skunk feeding posture where the cylinder polymer bait is held downward and bitten into with the molars.

rather than bitten and VCs were not punctured. The skunks did not seem to have a problem passing the VC through their gastrointestinal tracts.

Ontario slims were often held vertically and bitten at the corners with the teeth penetrating the VC from the top and bottom of the thin bait. In some cases, skunks held the slim baits flat against the ground and used their incisors to bite across the surface of bait matrix.

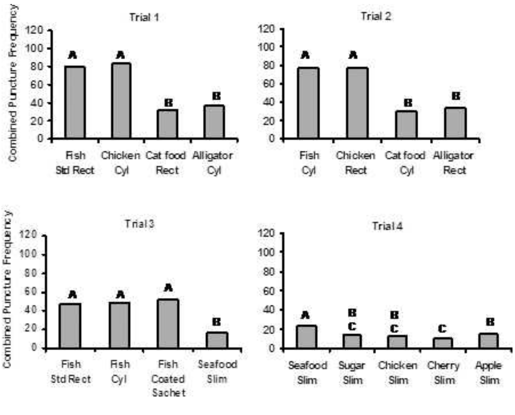

FIGURE 4. Combined frequency of punctured vaccine containers (N=120/bait type/trial) based on four independent consumption trials. Letters indicate statistical differences determined from Kruskal-Wallis tests. Each trial lasted 5 days. Twenty-four striped skunks of mixed sex and age class were the test subjects from 22 September to 17 October 2003.

Consumption trials

Puncture of VCs was contingent upon bait selection, but bait selection does not necessarily result in skunks puncturing the VC. Chi-square results indicated that nonpuncture of selected VCs did not differ among bait types in Trials 1–3 (α=0.05). Data collected in Trial 4 could not be analyzed because of insufficient observations for a chi-square test. The data indicated that VC puncture frequency was a function of bait type. Additionally, both selection frequency and VC puncture frequency responses yielded similar results (i.e., if a bait was bitten into, the VC was likely to be punctured), which made it unnecessary to discuss them separately. Therefore, we discuss our results in terms of VC puncture frequency.

When controlling for bait, day was significantly associated with VC puncture in Trials 1–3 ($P<0.01$), but not in trial 4 ($P=0.13$). Although VC puncture frequencies differed among days, no significant (α=0.01) associations were observed between day and VC puncture for any of the baits in any of the trials. In other words, VC puncture frequencies differed among days, but daily variation was not influenced by bait type. Therefore, VC puncture frequencies among baits were examined by combining results from all days of a trial.

Trial 1: Bait type and VC puncture of polymer baits were significantly associated ($P<0.01$). The VCs in fish standard rectangle and the chicken cylinder baits were punctured more frequently than VCs in the alligator bait cylinder and cat food rectangle baits (Fig. 4). There was also a significant bait effect ($P<0.01$) observed in the rank consumption data (Fig. 4). Consumption was greatest for the fish standard rectangle and chicken cylinder baits, which was significantly greater than both the alligator bait cylinder and cat food rectangle baits.

Trial 2: There was a significant association between polymer bait type and VC puncture ($P<0.01$). The VCs in fish cylinder and chicken rectangle baits were punctured more frequently than in the alligator bait rectangle and cat food cylinder baits (Fig. 4). There was also a significant bait effect ($P<0.01$) in the rank consumption data (Fig. 4). Consumption was greatest for the chicken rectangle and fish cylinder baits versus the alligator bait rectangle and cat food cylinder baits.

Trial 3: There was a significant association between the fish-flavored baits and VC puncture ($P<0.01$). Puncture frequency of VCs in the seafood Ontario slim was low, whereas VC puncture of the fish cylinder polymer, fish standard rectangle polymer, and fish-coated sachet were similar (Fig. 4). There was a significant bait effect ($P<0.01$) and day effect ($P=0.02$) in the rank consumption data (Fig. 4). Consumption was similar among the fish standard rectangle polymer, fish cylinder polymer, and fish-coated sachet baits, which were all greater than the seafood Ontario slim bait.

Trial 4: There was no significant association between the four flavors of Ontario slim baits and VC puncture ($P=0.13$). Puncture frequencies of VCs were highest for seafood followed by apple, sugar, chicken, and cherry (Fig. 4). Trial 4 did not result in a significant consumption model ($P=0.10$), indicating that the variation observed in this trial could not be attributed to day or bait, or the interaction of bait $\times$ day.

DISCUSSION

Video data of bait examination and selections suggested that skunks selected for bait characteristics. Although skunks approached and/or examined first the right side of the plate in all trials, this consistent approach did not influence bait selection. In Trials 1 and 2, skunks selected first fish and chicken flavors more frequently than alligator bait or cat food flavors, regardless of shape. Moreover, fish and chicken baits were first selected more often than first examined. This suggested that, following examination of available baits, skunks preferred to initiate consumption with these meat-flavored baits over alligator bait or cat food baits.

In Trial 3, all fish-flavored baits were first examined similarly, but seafood slims were never first selected. Novelty may have contributed to the occurrence, but the fish-coated sachets were also novel and their examination and selection frequencies were comparable to polymer baits. Nonselection of seafood slims first indicated that following examination of available baits, the seafood slims did not stimulate a consumption response in the skunks.

In Trial 4, the familiar seafood slims were most often the first-selected bait; but in general, the slim baits appeared to be unattractive, as indicated by the low occurrence of examination and selection. The low attraction might have been due to skunks becoming accustomed to testing. Another possibility is that experience in consumption of slim baits reduced attractiveness: video data of consumption behavior suggested that low attractiveness was most likely due to bait texture. The waxy texture of slim baits appeared to make chewing difficult. The matrix would often stick to skunks' teeth or the roof of the mouth, stimulating them to gag and spit out pieces of the bait, which sometimes led to rejection. In a field setting where an option for various ORV baits for skunks is not likely, distribution of bait that skunks readily select (and consume), such as the fish and chicken polymer baits or coated sachets, would be most efficient for ORV campaigns.

Video data of consumption behavior indicated that large baits seemed difficult to manipulate and inhibited contact with VCs. Due to skunks' significantly smaller jaw size and oral cavity than other species targeted by current ORV baits, all trial baits were too large to be fully inserted into their mouths and chewed. Only the coated sachet was inserted far enough into the mouth to enable a bite through the VC.

Typical skunk feeding posture was to tilt the head to one side, biting into the bait with the molars. The posture was ideal because it may facilitate a more direct splash of vaccine on the oropharyngeal mucosa. The thickness of the bait wall and breakage of rectangle polymer baits resulted in minimal VC contact; the baits were seldom held at an angle to facilitate delivery of liquid to the mouth. These baits tended to break apart quickly and either exposed the VC, making it easy for skunks to select against the synthetic material, or caused the VC to separate from the matrix, and thereby minimizing availability for puncture. Additionally, larger baits were sometimes held against the ground and consumed with the skunks' mouths downward. With their mouths downward, skunks might have repeatedly punctured VCs but most of the liquid contents dripped or spilled to the ground rather than being directed in the mouth. The thinner walls, smaller circumference, and continuous shape of cylinder polymer baits enabled skunks to insert an entire end into their mouths and bite through the cylinder, which aided VC puncture. The direct coating on coated sachets and Ontario slims decreased the likelihood of the VC becoming separated from the bait matrix and, thereby maximizing its availability for puncture.

The VCs were folded in half inside the hollow core of polymer baits. This made them slightly pressurized and probably aided in delivery of liquid contents. Vaccine containers in slims were not under pressure and most liquid dripped out of puncture holes on the ground. The VCs in coated sachets were not pressurized, but insertion of the coated sachet into the mouth positioned it such that a puncture would more likely direct liquid into the oral cavity rather than drip to the floor.

In consumption trials, some baits may be selected and its VC not punctured. We examined whether this was influenced by bait type, and results indicated that VCs of selected baits were likely to be punctured, regardless of bait type. From an applied standpoint, these results offered no indication that a specific bait type might be selected but the VC not punctured in a field situation.

The day effect observed in VC puncture $\times$ day data for Trials 1–3 generally followed a downward trend. This daily variance was likely due to initiation of trials on Mondays (Day 1); on weekends, a diet of only maintenance feed was offered to skunks prior to the start of a subsequent trial. Although VC puncture frequency was generally highest on Mondays, VC puncture frequency by bait type was generally consistent through Fridays.

Puncture frequencies of VCs consistently decreased over the course of the four trials. The decrease may have been due to skunks becoming accustomed to testing, or it may have been a response to bait types. Fish-flavored polymer baits were offered in all but Trial 4 and consistently were one of the baits with the highest consumption and VC puncture frequency. Thus, fish-flavored polymer baits could be viewed as the standard of interest by skunks across trials.

In Trials 1 and 2, consumption and VC puncture frequency were highest for chicken- and fish-flavored baits, regardless of bait shape. The observation strongly suggested that flavor influenced VC puncture frequency. However, in Trial 4, chicken and seafood flavors of Ontario slim baits did not have consumption or VC puncture frequency similar to those observed in Trials 1 and 2. The lower consumption and VC puncture frequencies of chicken and fish flavors in Trial 4 as compared to Trials 1 and 2 may be due to the different flavor additives used between bait companies, or to the bait texture of Ontario slims (based on video footage of consumption behavior).

In Trial 3, two novel fish-flavored baits (coated sachet and Ontario slim) were offered. The novelty of the baits may have contributed to the overall decrease in VC puncture frequency from Trials 1 and 2; however, VC puncture frequency was also low for familiar baits. Regardless, VC puncture frequencies among fish-coated sachet baits and the fish polymer baits were comparable, whereas VC puncture was low for seafood slim baits. Video data of consumption behavior for Trial 3 indicated that although the odor of seafood Ontario slim baits was attractive, low VC puncture frequency was likely due to bait texture, as was observed in Trial 4.

In Trial 4, VC puncture frequency, in general, was lower than all other trials. Video data of consumption behavior suggested that the odor of Ontario slim baits was sufficiently attractive; however, texture was likely the cause of the low selection and subsequent low VC puncture frequency.

Future versions of ORV baits can be made more effective and can improve vaccine delivery by tailoring bait preferences and consumption behaviors of target species. Skunks were discriminatory in their selection. Fish- and chicken-flavored baits were most often selected and consumed, and most often had punctured VCs. Baits having parts that could be entirely inserted into a skunk's mouth maximized contact with the VC and the likelihood of vaccine ingestion. Although polymer baits had a high VC puncture frequency, the amount of liquid ingested was variable. The texture of Ontario slims may have limited selection by skunks. Baits with a direct coating on the VC, such as the coated sachets and Ontario slim baits, and with a slightly pressurized VC were more conducive to adequate vaccine delivery. Further consideration should be given to methods and materials used to directly coat VCs, such as adhesives. Additionally, flavoring the vaccine without compromising its stability and efficacy, and development of a homogeneous bait and vaccine in a solid form could effectively bypass most of the weaknesses we found with existing VCs. Vaccine container development should consider volume delivery. Based on our trials, the fish-coated sachets were attractive to skunks, easily manipulated, and facilitated the best likelihood of adequate liquid ingestion. The fish-coated sachet may be an ideal ORV bait for use in field trials for skunks.

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