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## EVALUATION OF BAITS FOR ORAL RABIES VACCINATION OF MONGOOSES: PILOT FIELD TRIALS IN ANTIGUA, WEST INDIES

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**ABSTRACT:** A field study was conducted on the island of Antigua, West Indies, to evaluate baits for delivering an oral rabies vaccine to the small Indian mongoose (*Herpestes auro-punctatus*). Tracking tiles were used to determine that mongooses were nonselective and took both egg-flavored polyurethane baits and fish-flavored polymer baits containing several different food materials. A high proportion of baits were taken the day of placement with minimal disturbance by nontarget species. DuPont Oil Blue A® dye was an effective short-term biomarker for use in baits; based on its subsequent detection in mongooses, some of the population had consumed and not cached or discarded baits. Central point baiting stations showed promise as an alternative delivery technique.

**Key words:** *Herpestes auro-punctatus*, small Indian mongoose, oral vaccination, rabies, bait evaluation.

### INTRODUCTION

The small Indian mongoose (*Herpestes auro-punctatus*) was introduced into many of the Caribbean Islands during the period between 1866 and 1900 (Nellis, 1989); the animal now is an important reservoir of rabies in Cuba, the Dominican Republic, Grenada, Haiti, and Puerto Rico (Everard and Everard, 1988). Possible introduction of the disease into rabies-free mongoose populations on other islands of the West Indies also is of concern. The extent of rabies in mongooses is not known, but reported cases most certainly represent only a small proportion of the actual cases. Attempts to eliminate rabies by reducing mongoose numbers with toxic baits have been unsuccessful, even on small islands (Everard and Everard, 1988).

Immunization of terrestrial wildlife with baits containing an oral rabies vaccine may be a feasible alternative to population reduction programs or trap-vaccinate-release control methods (Baer, 1988; Blancou et al., 1988; Perry, 1989; Wandeler, 1991). However, this approach for controlling mongoose rabies has received little attention; an effective oral vaccine has not been developed. M. C. Vargas (pers. comm.) has evaluated the bait preferences of captive mongooses with the objective of delivering

an oral rabies vaccine to this species. Keith et al. (1990) has reported on a baiting system for delivering a toxicant, diphacinone, to mongooses preying upon endangered birds in Hawaii.

In our pilot field trials on Antigua, we evaluated use of tracking tiles to record bait discovery and removal; the disturbance rates of experimental baits by mongooses and nontarget species; mongoose bait consumption, both with a new oral biomarker and by direct observation; and the efficacy of central point bait stations for delivering baits to mongooses.

### MATERIALS AND METHODS

Antigua is a 280-km<sup>2</sup> island located on the outer edge of the Leeward Islands chain in the West Indies (17°05'N, 61°50'W). Mean annual rainfall and temperature are 110 cm and 26.7 C, respectively, with a distinct dry season during the winter and early spring months. Vegetation is primarily mixed grass-woody thicket comprised of acacia (*Acacia* sp.), mesquite (*Prosopis chilensis*), and logwood (*Haematoxylum campechianum*) (Harris, 1965). We conducted our field tests in Antigua from 6 to 23 February 1991. Baits, tracking tiles, and live-traps were placed along interior dirt roads or power line right-of-ways accessible by pickup truck. We selected roads characterized by 3 to 5-m high dense thicket and trees because of the higher mongoose densities present in such areas.

Two bait types were tested. The first was a

1.7- × 3.0-cm cylindrical-shaped polyurethane sleeve bait similar to that developed for raccoons (Linhart et al., 1991). It was saturated with a 50:50 homogenate of corn oil (Best Foods, CPC International Incorporated, Englewood Cliffs, New Jersey, USA) and whole fresh chicken eggs; the sleeve bait then was coated with fish meal (Faithway Feed Company, Incorporated, Gunterville, Alabama, USA). The second bait type (1.2 × 2.5 cm) was an experimental polymer bait obtained from E. I. DuPont de Nemours and Company (Sabine River Laboratory, Orange, Texas, USA; Smith and Daigle, 1988) that was custom formulated for our specific mongoose field tests. The four polymer bait formulations we tested were intended to represent different basic food groups: egg, grain, poultry, and fish products. They consisted of an extruded cylindrical bait formulated with various dry meals (obtained from an Athens, Georgia, feed mill), soybean oil as a lubricant, and a DuPont polymer (Aquaseal®) to bind and waterproof the bait (Smith and Daigle, 1988). Baits A, B, and C all contained 12% polymer, 10% soybean oil, and 20% fish meal. In addition, bait A also contained 57% powdered whole eggs (Dried Egg Product, PYA/Monarch Food Services, Greenville, South Carolina, USA); bait B contained 57% broiler starter (Poultry Science Feed Mill, The University of Georgia, Athens, Georgia, USA); and bait C contained 57% poultry by-products (North Georgia Rendering Company, Cumming, Georgia), respectively. Bait D, a proprietary bait, contained unspecified amounts of polymer, soybean oil, fish meal, and fish sludge, and except for size, was nearly identical to the raccoon bait described by Hanlon et al. (1989).

We inserted 1- × 3-cm paraffin wax ampules (W&F Manufacturing Company, Inc., Buffalo, New York, USA) containing 1 ml of food dye colored water into polyurethane baits. These baits were placed in 15- × 15- × 61-cm live traps (Tomahawk Live Trap Company, Tomahawk, Wisconsin, USA) containing captured mongooses to observe their feeding behavior. Other polyurethane baits were modified by mixing 2 g of an experimental short-term dye marker, DuPont Oil Blue A® (DuPont Chemicals, Wilmington, Delaware, USA), into 1 liter of the corn oil/whole egg mix. Baits were saturated with the mixture and each contained about 7.5 mg of dye. Assuming mean mongoose weights of 465 g for females and 710 g for males from previous work (Linhart, unpubl.) and that all bait material would be consumed, doses of dye per bait per animal averaged about 16.1 and 10.6 mg/kg of body weight, respectively.

One hundred tracking tiles (Lord et al., 1970; West et al., 1976; Clark and Campbell, 1983) were placed about 20 m apart along infrequent-

ly traveled rural roads. Black printers ink (Superior Printing Inks, New York, New York, USA) was dissolved in mineral oil (150 g ink/1 liter oil) and painted in a 5-cm-wide strip along the edges of 30- × 30- × 0.23-cm white fiberglass tiles that were cut from fiberglass sheets purchased at a building supply store. A 20- × 20-cm square of white tracking paper was positioned in the center of each tile. A 3.0- × 4.5-cm paper condiment cup was placed in the center of the paper. A nail was pushed through the cup, paper, and a hole in the center of the tile to secure tracking tile components together and the tile to the ground. A single test bait was placed in the cup. We recorded both the number of baits taken and the species visiting the tracking tiles by checking them daily and comparing animal tracks with those shown in Murie (1954) and Faaborg and Arendt (1985). Tiles were rendered inoperable only infrequently by rural people, their livestock, or rain and wind.

The four experimental DuPont baits were placed one per station in series of four, along a line of 100 stations consisting of 25 series of four baits each. The sequence of baits within each series was randomized using a table with random numbers. Baits were placed out for the initial test between 15:00 and 18:30 hr, left overnight, and checked the following morning for disturbance by mongooses and nontarget species. Initially there was considerable bait loss to nocturnally active mice (*Mus musculus*); all subsequent tests were conducted by placing baits or traps out in the morning and checking them in late afternoon of the same day.

We sought to determine whether mongooses were visually attracted to the conspicuous tracking tiles used for our test, and whether baits placed directly on the ground would be discovered and taken at the same or a different frequency as those put on tracking tiles. This question had important practical implications because the widespread distribution of vaccine baits obviously would not involve use of tracking tiles, and baits distributed from aircraft or broadcast on the ground might not be readily discovered by mongooses. On 18 February 1991, we placed 100 polyurethane baits (bait D) along a line in a new location where baits at even-numbered sites were placed on tracking tiles and baits at odd numbered sites were placed directly on the ground. Baits were placed in the morning (08:30 to 11:15 hr) and checked later the same day (15:45 to 16:30 hr) to determine bait disappearance rates.

To determine whether baits actually were eaten by mongooses, we distributed 100 polyurethane baits containing DuPont Oil Blue A dye along both sides of a 1-km power line right-of-way by throwing them from a pickup truck

at about 20-m intervals into the thicket on either side of the road. To capture mongooses, 30 Tomahawk live traps baited with raw chicken were set along the road at about 40-m intervals the morning after bait distribution and checked later the same day. Captured mongooses were sedated with 4.5 mg/kg ketamine (Veterinary Products, Bristol Laboratories, Syracuse, New York, USA) and xylazine (The Butler Company, Columbus, Ohio, USA) administered intramuscularly in the hip. They then were euthanized by intracardiac injection of 0.2 mg/kg sodium pentobarbital (Veterinary Laboratories, Incorporated, Lenexa, Kansas, USA). Their abdominal fat and lower intestine were exposed and visually checked for the presence of dye. We also checked the bone marrow in femurs to determine if the dye had been deposited in the marrow fat.

We observed captive mongooses kept in live traps and fed polyurethane baits to determine their feeding behavior and the fate of paraffin wax ampules contained within the baits. This was carried out by live-trapping five mongooses in the early morning and transporting them to a shaded cement-floored building. One bait was placed with each of five mongoose traps and a hidden observer watched the animals for several hours to determine if and how baits and ampules were eaten.

Use of central point bait stations involves placing multiple baits at several widely spaced central locations with the expectation that visits by different animals will result in a high proportion of the population locating and consuming baits (Keith et al., 1990). If effective, such a technique might be less costly and labor intensive than the distribution of individual baits throughout an entire treatment area. Such stations also might reduce nontarget species interference. We selected five central baiting stations approximately 400 m apart, cleared a 45-cm circular site of leaves and debris, and placed 20 DuPont D baits within each cleared site. Baits were placed out from 09:15 to 11:15 hr and each station checked at 17:00 to 17:40 hr on the same day to see how many baits had been taken.

## RESULTS

When DuPont baits A, B, C, and D were placed on 100 tracking tiles and left until the following morning, there was considerable bait loss to nocturnally active house mice. Mongoose tracks were found on 21 of 100 tiles where they had removed 20 baits (20%). House mice visited 37 stations and took 32 baits (32%). Both mouse and mongoose tracks were found on 41 addi-

tional tiles with missing baits (41%), thereby making it impossible to determine which animal was responsible for bait removal. A rat (*Rattus* sp.) removed a single bait, one of only two stations visited by this species during the entire period of bait evaluation in Antigua.

A subsequent test of the same four DuPont baits, but placed out in the morning and checked for disturbance rates in late afternoon of the same day, resulted in removal of baits from 82 (87%) of 94 operable stations within a few hours. Mongooses visited and took baits from 73 (78%) stations; of these 19 (26%) were A baits, 18 (25%) were B baits, 17 (23%) were C baits, and 19 (26%) were D baits. Based on a G-statistic with Williams correction indicated that bait uptake was independent ( $P > 0.10$ ) of bait type (Sokal and Rohlf, 1981). Mice visited only four stations and took three baits (3%). Tracks of both species were found at only six additional stations from which all baits (6%) had been removed.

Mongooses took 44 (88%) of 50 available baits on tracking tiles and 37 (76%) of 49 placed directly on the ground; the difference in bait uptake was not significant ( $P > 0.10$ ). The percentage of baits removed from the ground by mongooses (76%) was nearly the same as that reported earlier for the bait disturbance test (78%). Based on these data, we believe that mongooses would most likely locate and take inconspicuous baits dropped directly on the ground in the manner which would occur in large scale bait applications.

Based on examination of trapped mongooses following distribution of baits with DuPont Oil A dye, seven of ten animals had both abdominal fat and lower intestines colored a vivid robin's egg blue. No dye was found in the femur bone marrow. Thus some mongooses along the baited road consumed one or more baits.

During our observations of captive mongooses held in live traps and given baits, four of five mongooses ate baits; however, varying amounts of colored water leaked

from all four ampules as they ruptured during bait consumption. The dye and bait material passed rapidly through the gastrointestinal tract of the mongoose; one animal ingested bait and dye water at 08:30 hr and excreted green-colored feces 8 hr later.

DuPont baits were rapidly taken at all five central baiting sites. We found that 93 of the 100 baits placed out at 09:15 to 10:15 hr had been removed by late afternoon of the same day. However, no data were available to indicate either the number of animals involved or the proportion of the local mongoose population that took baits.

#### DISCUSSION

Results of our field trials on Antigua, while preliminary in nature, revealed that mongooses removed virtually all test baits and showed no apparent bait preferences. We found that baits left overnight were taken by nocturnally active mice but when placed out in the morning they were taken almost exclusively by mongooses during the same day. Mongooses readily located baits and odor enhancement of baits with a supplemental olfactory attractant was not considered necessary. Baits placed at central point bait stations also disappeared within a few hours. However, observation of captive mongooses that consumed placebo vaccine baits indicated that liquid loss from paraffin wax ampules could reduce the efficacy of vaccine delivery. The use of a new biomarker, DuPont Oil Blue A dye, showed that an unknown percent of mongooses ate the baits they encountered; we saw no evidence that mongooses discarded or cached the baits they removed from stations. Tracking tiles were extremely effective for evaluating mongoose and nontarget species bait disturbance. The mongooses had a much smaller body size (465 to 710 g) and oral cavity as compared to other carnivores for which vaccine baits have been developed; this difference in size will likely necessitate the use of smaller baits, vaccine containers, and doses of vaccine.

From our data, it appears that bait ingredients were not critical and that discovery and ingestion rates were high. However, much more research is needed to effectively vaccinate mongooses. For example, the persistence of DuPont Oil Blue A<sup>®</sup> and other candidate biomarkers need to be determined so that placebo baiting field trials can be conducted to ascertain seasonal and geographic variations in efficacy and selectivity. Smaller baits and vaccine containers or an alternate method of vaccine delivery are needed. Ant damage to baits can be seasonally severe (M. C. Vargas and J. O. Keith, pers. comm.) and the incorporation of an insect repellent such as dimethyl phthalate (Aldridge Chemical Company, Milwaukee, Wisconsin, USA; Roese, 1984) into baits needs investigation. No data are available on such basic information as the minimum number of baits required per unit of land area to reach 70% or more of the mongoose population. An accurate and easily applied technique is required to assess relative mongoose densities so as to eventually establish the relationship between bait density, mongoose densities, and the proportion of the mongoose population that can be reached by baits. Finally, once an orally effective rabies vaccine for mongooses becomes available, vaccine bait field trials should be undertaken concurrently with active rabies surveillance efforts to determine to what extent the control technique can suppress or eradicate the disease.

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