AVOIDANCE OF METHIOCARB-POISONED APPLES BY RED-WINGED BLACKBIRDS

J. RUSSELL MASON, U.S. Department of Agriculture, Animal and Plant Health Inspection Service, Denver Wildlife Research Center, % Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104-3308

Abstract: I tested the relative effectiveness of chemical (methyl anthranilate) and visual (calcium carbonate [$CaCO_3$]) stimuli as cues to enhance methiocarb-induced food avoidance in red-winged blackbirds (*Agelaius phoeniceus*). Methyl anthranilate was superior to $CaCO_3$, but a combination of both cues elicited stronger learning than either cue alone. I speculate that specific chemical cues or a combination of chemical and visual cues could permit reductions in the rate of methiocarb application without negative impacts on effectiveness, thus reducing the costs of crop protection from birds.

J. WILDL. MANAGE. 53(3):836–840

Methiocarb is the most effective broad-spectrum bird repellent currently registered for use in the United States. It acts by eliciting conditioned (i.e., learned) avoidance of treated foods via post-ingestional malaise (Rogers 1974, Conover 1984). Unfortunately, the cost of methiocarb ($54.00/kg) is prohibitively high for use in many agricultural situations.

Previous investigations have suggested that methiocarb-induced avoidance of crops might be enhanced by the addition of visual, olfactory, or tactile cues (Mason and Reidinger 1982, 1983*a*; Bullard et al. 1983; Avery 1984). The addition of cues could permit reductions in the rate of methiocarb application (Mason and Reidinger 1983*a*, Avery 1984), without significant changes in effectiveness, thereby reducing the cost of crop protection.

To date, only visual enhancement of methiocarb has been systematically investigated (Mason and Reidinger 1982, 1983*a*,*b*; Avery 1984; Tobin 1985*a*,*b*). While laboratory findings have generally supported the notion that visual markers increase methiocarb effectiveness (Mason and Reidinger 1982, 1983*a*), field test results have been ambiguous (Bullard et al. 1983). One factor that may contribute to the lack of positive results in some field studies is the harmless (i.e., nonaposematic) quality of the visual cue (white) employed. The present experiment was designed to assess whether the use of an aversive sensory cue might elicit greater avoidance of methiocarb-treated food than methiocarb alone, or methiocarb in combination with a neutral color cue.

I thank M. L. Avery, R. A. Dolbeer, J. F. Glahn, and D. L. Otis for critical reviews of earlier manuscript drafts. S. Lewis provided valuable technical assistance. Funding was provided by the Animal and Plant Health Inspection Service, U.S. Department of Agriculture.

METHODS

Fifty-six male red-winged blackbirds were mist netted during March 1988, and transported to the Monell Chemical Senses Center (Philadelphia, Pa.). The birds were individually caged (61 × 36 × 41 cm) under a 6:18 hour light: dark cycle that maximized feeding without reducing the total quantity of food consumed (Rogers 1974, Mason and Reidinger 1983*a*). Use of this feeding regime helped to assure regular measurable consumption during the experiment. Water was freely available, and before the experiment began, birds were permitted free access to Purina Flight Bird Conditioner (PFBC) (Purina Mills Inc., St. Louis, Mo.) and crushed shell grit. Birds were not presented with fruit during pretreatment, because apple consumption was to be used as the dependent variable during the treatment period. Pre-exposure to stimulus foods prior to methiocarb treatment can decrease the ability of birds to acquire avoidance in the laboratory (Mason and Reidinger 1983*b*) and in the field (Dolbeer 1980).

Technical grade methiocarb (CAS No. 2032-65-7, Mobay Chem. Co., Kansas City, Mo.) dissolved in propylene glycol to form a 0.4% weight/weight (w/w) solution served as the unconditioned stimulus. Methyl anthranilate (CAS No. 134-20-3, Int. Flavors and Fragrances, Union Beach, N.J.) and $CaCO_3$ (CAS No. 471-34-1, Aldrich, St. Louis, Mo.) dissolved in propylene glycol to form a 25% (w/w) solution served as the conditioned stimuli. Methyl anthranilate is a harmless flavoring that is repulsive to birds (Avery et al. 1988, Mason et al. 1989). Calcium carbonate is a white pigment that has been used in laboratory and field studies of cue-enhanced methiocarb efficacy (Bullard et al. 1983).

During pretreatment (day 1–7), birds were adapted to a food deprivation regime that continued throughout the experiment. Deprivation involved removing PFBC and grit from the cages just before dark. Thirty minutes before first light on the following day, each bird was presented with a cup containing 20 g of PFBC. After 2 hours, consumption was measured. Birds were then left undisturbed with free access to PFBC and grit for the remaining hours of light.

On day 8, the birds were assigned to 8 groups (6 birds/group) on the basis of mean pretreatment consumption. The bird with the highest pretreatment (feeding trial) consumption was assigned to the first group, that with the next highest consumption was assigned to the second group, and so on. The next 8 birds were assigned to groups in the opposite order. This assignment regime assured that the groups were balanced with respect to consumption of PFBC prior to treatment. I assumed a priori that consumption of PFBC reflected eating behavior in general, and that the groups would be approximately equal with respect to consumption of other foods including apples.

Table 1. Treatments presented to each group of red-winged blackbirds to examine chemically and visually enhanced avoidance of methiocarb-poisoned apples.

Group	Treatment
M	Methiocarb
C–M	Calcium carbonate ($CaCO_3$) and methiocarb
MA	Methyl anthranilate
M–MA	Methiocarb and methyl anthranilate
C–MA	$CaCO_3$ and methyl anthranilate
C–MA–M	$CaCO_3$, methyl anthranilate and methiocarb
C	$CaCO_3$
P	Propylene glycol

During the treatment period (day 9–18), each bird was presented daily with a quarter of a Mackintosh apple suspended 4 cm above the floor in the center of the front of each cage. Red-winged blackbirds do not damage apples in the wild, but they readily consume them in the laboratory, and apple slices provided a convenient food to which stimulus solutions could be applied.

Apple slices were freshly prepared each morning, and were weighed just before and just following each diel test period. Five additional slices were prepared, weighed, and placed in an empty cage during each test period. The mean weight loss of these apple slices was taken to reflect evaporation, and was subtracted from measurements of consumption.

A 5-mL syringe fitted with a 14-gauge needle was used to inject slices (Table 1). Group M received apple slices injected with methiocarb solution (0.2 mL/kg of apple). Group C–M received slices injected with methiocarb solution (0.2 mg/kg) and dipped in $CaCO_3$. Group MA was presented with slices injected with methyl anthranilate (0.4% w/w). Slices given to group M–MA were injected with methiocarb (0.2 mL/

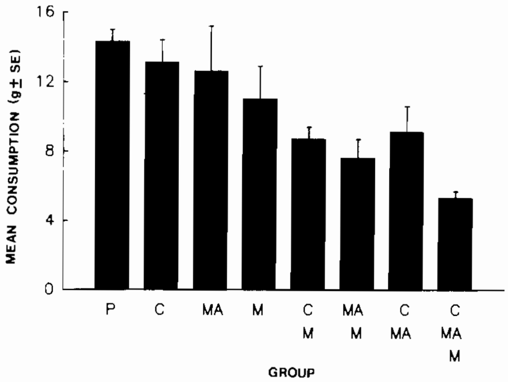

Fig. 1. Consumption of stimulus apple slices by all groups of red-winged blackbirds, collapsed over treatment and post-treatment sessions. Group P was presented with untreated apples. Group C was presented with apples dipped in calcium carbonate ($CaCO_3$). Group MA was presented with apples injected with methyl anthranilate. Group M was presented with apples injected with methiocarb. Group C–M was presented with apples injected with methiocarb and dipped in $CaCO_3$. Group MA–M was presented with apples injected with both methyl anthranilate and methiocarb. Group C–MA was presented with apples injected with methiocarb and dipped in $CaCO_3$. Group C–MA–M was presented with apples injected with methyl anthranilate and methiocarb, and dipped in $CaCO_3$.

kg) and methyl anthranilate (0.4% w/w). Group C–MA was given slices injected with methyl anthranilate (0.4% w/w) and then dipped in $CaCO_3$. Group C–MA–M was given slices injected with methiocarb (0.2 mL/kg), methyl anthranilate (0.4% w/w), and dipped in $CaCO_3$. Group C was given slices dipped in $CaCO_3$ only, and group P received slices injected with propylene glycol (0.2 mL/kg apple).

To assure an even distribution of methiocarb, methyl anthranilate, or propylene glycol in apple slices, I injected a minimum of 20 sites on each apple slice. On all treatment days, birds were observed by video camera for symptoms of malaise (i.e., regurgitation, bill-wiping, immobility, feather erection).

The concentrations of methiocarb, methyl anthranilate, and $CaCO_3$ applied to apple slices were chosen on the basis of previous experimentation. The level of methiocarb was suggested by pilot work as the lowest dose to reliably elicit symptoms of malaise in red-winged blackbirds eating apples under laboratory conditions. The level of methyl anthranilate (0.4%) was chosen as the lowest concentration that reliably elicits food avoidance in the laboratory (Mason et al. 1989). The level of $CaCO_3$ (25% in aqueous solution) was identical to that used in a prior field test of this cue (P. P. Woronecki and R. W. Bullard, Denver Wildl. Res. Cent., Denver, Co., unpubl. data). Our aim in choosing marginally effective methiocarb and methyl anthranilate concentrations was to provide an experimental setting in which any increase in stimulus-elicited avoidance could be readily observed.

Following treatment, all birds were permitted access to fresh, untreated apple slices on each of 10 post-treatment days. Consumption of apple during each 2-hour feeding period was measured, and measurements were adjusted for evaporation, as previously described.

An analysis of variance (ANOVA) was used to assess the results. The factors in this analysis were group, treatment, and session. Subsequent to the omnibus procedure, Tukey's Honestly Significant Difference tests were used to isolate significant differences among means ($P < 0.05$).

RESULTS

Video observations revealed that red-winged blackbirds in groups offered methiocarb poisoned slices (i.e., M, C–M, MA–M, C–MA–M) regularly exhibited symptoms of malaise. Red-winged blackbirds in groups P, C, MA, and C–MA showed no signs of discomfort.

Group C–MA–M ate least overall ($F = 4.5$; 7, 40 df; $P < 0.001$) (Fig. 1). Groups C–M and MA–M ate slightly more ($P < 0.05$), followed by groups C–MA and M ($P < 0.05$). The most consumption was exhibited by groups C, MA, and P ($P < 0.05$).

Overall, treatment consumption ($\bar{x} = 8.0 \pm 0.6$ [SE]) was lower than post-treatment consumption (12.4 ± 0.8), but not all groups contributed to this effect ($F = 2.7$; 7, 40 df; $P < 0.02$) (Fig. 2). Among those groups that exhibited differential consumption, the greatest differences were shown by group MA, followed by group M ($P < 0.05$). Differences observed for groups MA–M, C–M, and C–MA–M were not so large, albeit statistically significant ($P < 0.05$). Birds in groups C–M, C, and P exhibited similar levels of consumption during treatment and post-treatment.

Within treatment and post-treatment, there were overall increases in consumption during later sessions ($F = 5.4$; 9, 360 df; $P < 0.00001$). Such increases were most pronounced during treatment ($F = 5.4$; 9, 360 df; $P < 0.00001$), and largest for groups C, MA, and M ($F = 1.9$; 63, 360 df; $P < 0.0003$).

The following pattern emerged when all factors were considered ($F = 1.5$; 63, 360 df; $P < 0.011$). Group C–MA–M showed consistently low levels of consumption during treatment and post-treatment (relative to the other groups $P < 0.05$), although post-treatment consumption was higher than treatment consumption. For groups C–M, MA–M, and C–MA, treatment consumption was higher than that of group C–MA–M ($P < 0.05$) and tended to increase over days: post-treatment consumption was equivalent to that of C–MA–M birds. Groups M and MA showed treatment consumption similar to that of groups C–M, MA–M, and C–MA, but post-treatment consumption by M and MA birds was significantly higher than that exhibited by birds in these other groups ($P < 0.05$). Groups C and P showed significantly higher consumption during treatment and post-treatment than did any of the other groups ($P < 0.05$) and failed to show lower consumption during treatment than during post-treatment.

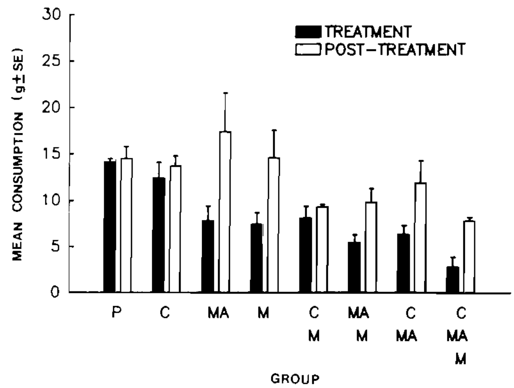

Fig. 2. Treatment and post-treatment consumption by all groups of red-winged blackbirds, collapsed over sessions. Group C was presented with apples dipped in calcium carbonate ($CaCO_3$). Group MA was presented with apples injected with methyl anthranilate. Group M was presented with apples injected with methiocarb. Group C–M was presented with apples injected with methiocarb and dipped in $CaCO_3$. Group MA–M was presented with apples injected with both methyl anthranilate and methiocarb. Group C–MA was presented with apples injected with methylanthranilate and dipped in $CaCO_3$. Group C–MA–M was presented with apples injected with methyl anthranilate and methiocarb, and dipped in $CaCO_3$.

DISCUSSION

The results of my experiment are consistent with previous laboratory findings (Rooke 1983). First, combinations of methiocarb and sensory cues were generally more effective than methiocarb alone. Also, redundant (visual and chemical) cues were relatively more effective than either cue alone paired with the repellent (Mason 1988).

Ecologically, the importance of cues to food avoidance learning and the superiority of redundant cues are predictable from studies of predator-prey interactions. Toxic prey often use aposematic (i.e., warning) signals to advertise unpalatability to potential predators (Wickler 1968). Frequently, multiple signals are used, perhaps to decrease ambiguity, or perhaps to affect different sensory capacities in different types of predators (Mason 1988; D. H. Janzen, Univ. Pennsylvania, Philadelphia, pers. commun.).

My data suggest that an aversive chemical (methyl anthranilate), when paired with toxicant (methiocarb), is relatively more effective than pairing of methiocarb with an essentially neutral (i.e., non-aposematic) visual cue ($CaCO_3$). As for mammals (Garcia and Hankins 1977), it may be that when appropriate flavors are used, red-winged blackbirds show stronger learning of taste- than vision-sickness associations. The discrepancy between this notion and previous conclusions on food avoidance learning in red-winged blackbirds (i.e., that they show stronger color- than taste-sickness associations) (Mason and Reidinger 1983*a*) reflects the pervasive lack of chemosensory data on passerines and other birds in general (Kare and Mason 1986).

Differences among groups in treatment versus post-treatment consumption provide further evidence that acquisition and/or maintenance of avoidance differed among groups, or that the absence of some treatments was more easily detected. I favor the former explanation, because groups that showed the greatest differences in consumption between treatment and post-treatment (e.g., groups MA and M) were among those groups that showed the greatest habituation (i.e., increases in consumption over sessions) during treatment. These increases suggested that treatment conditions were not strongly repellent and that birds may have become accustomed to them.

MANAGEMENT IMPLICATIONS

The results of the present experiment suggest that the effectiveness of methiocarb applications might be enhanced through the use of aversive flavor cues, or aversive flavor cues in combination with visual cues. Substantial reductions in consumption during treatment and post-treatment were obtained when redundant cues were used even though the concentration of methiocarb was the minimum dose required to elicit behavioral symptoms of malaise. I speculate that the use of aversive flavors or flavor-visual cue combinations might permit reduced levels of methiocarb application in the field.

Apart from methiocarb effectiveness, the results also suggest that the effectiveness of aversive flavors can be improved by the addition of visual cues. For example, anthranilate derivatives can be used as feed additives to reduce bird depredations in feedlots (Mason et al. 1985). I propose that the use of anthranilates as feed additives might be improved by the addition of a distinctive color.

Although $CaCO_3$ was used as the visual cue in the present work because of its prior use in field evaluations, it is clear from an extensive body of laboratory and field research (Wickler 1968, Mason and Reidinger 1983*b*) that aposematic colors (e.g., orange or red) are likely to be more effective cues than colors that infrequently serve aposematic purposes. Perhaps the redundant use of aposematic colors and aversive flavor cues will elicit even stronger avoidance when paired with methiocarb than the avoidance observed when $CaCO_3$ was paired with anthranilate and methiocarb in the present study.

Finally, I caution that repellents are frequently applied to crops only after damage by birds is observed. This differs from the procedures of the present experiment, in which birds received no pretreatment exposure to apples. Pre-exposure to stimulus foods can weaken avoidance learning (Mason and Reidinger 1983*b*), and if implemented here, could have affected responses by birds.

LITERATURE CITED

AVERY, M. L. 1984. Relative importance of taste and vision in reducing bird damage to crops with methiocarb, a chemical repellent. Agric. Ecosystems Environ. 11:299–308.

———, R. E. MATTESON, AND C. O. NELMS. 1988. Repellency of methyl anthranilate and dimethyl anthranilate to caged red-winged blackbirds and European starlings. Denver Wildl. Res. Cent. Bird Sect. Res. Rep. 418. 14pp.

BULLARD, R. W., R. L. BRUGGERS, S. A. KILBURN, AND L. A. FIELDER. 1983. Sensory-cue enhancement of the bird repellency of methiocarb. Crop Prot. 2:387–398.

CONOVER, M. R. 1984. Responses of birds to different types of food repellents. J. Appl. Ecol. 21: 437–443.

DOLBEER, R. A. 1980. Blackbirds and corn in Ohio. U.S. Fish Wildl. Serv. Resour. Publ. 136. 18pp.

GARCIA, J., AND W. G. HANKINS. 1977. On the origin of food aversion paradigms. Pages 3–22 *in* L. M. Barker, M. R. Best, and M. Domjan, eds. Learning mechanisms in food selection. Baylor Univ. Press, Waco, Tex.

KARE, M. R., AND J. R. MASON. 1986. The chemical senses in birds. Pages 59–73 *in* P. D. Sturkie, ed. Avian physiology. Springer-Verlag, New York, N.Y.

MASON, J. R. 1988. Direct and observational learning by red-winged blackbirds (*Agelaius phoeniceus*). the importance of complex visual stimuli. Pages 99–116 *in* T. R. Zentall and B. G. Galef, eds. Social learning: psychological and biological perspectives. Lawrence Erlbaum Assoc., Hillsdale, N.J.

———, L. CLARK, AND M. A. ADAMS. 1989. Anthranilate repellency to starlings: chemical correlates and sensory perception. J. Wildl. Manage. 53:55–64.

———, J. F. GLAHN, R. A. DOLBEER, AND R. F. REIDINGER. 1985. Field evaluation of dimethyl anthranilate as a bird repellent livestock feed additive. J. Wildl. Manage. 49:636–642.

———, AND R. F. REIDINGER. 1982. Observational learning of food aversions in red-winged blackbirds (*Agelaius phoeniceus*). Auk 99:548–554.

———, AND ———. 1983*a*. Importance of color for methiocarb-induced food aversions in red-winged blackbirds. J. Wildl. Manage. 47:383–393.

———, AND ———. 1983*b*. Generalization and effects of pre-exposure on color-avoidance learning by red-winged blackbirds (*Agelaius phoeniceus*). Auk 100:461–468.

ROGERS, J. G. 1974. Responses of caged red-winged blackbirds to two types of repellents. J. Wildl. Manage. 38:418–423.

ROOKE, I. J. 1983. Conditioned aversion by silver-eyes *Zosterops lateralis* to food treated with methiocarb. Bird Behav. 4:86–89.

TOBIN, M. E. 1985*a*. Cues used by house finches for detecting methiocarb-treated grapes. Crop Prot. 4:111–119.

———. 1985*b*. Cues used by European starlings for detecting methiocarb-treated grapes. J. Wildl. Manage. 49:1102–1108.

WICKLER, W. 1968. Mimicry in plants and animals. McGraw-Hill Publ. Co., New York, N.Y. 255pp.

Received 3 October 1988.
Accepted 6 January 1989.