METHYL ANTHRANILATE AS A RICE SEED TREATMENT TO DETER BIRDS

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Abstract: Red-winged blackbirds (*Agelaius phoeniceus*) and related species cause millions of dollars of damage annually to sprouting rice in Louisiana and Texas. Seed treatments that deter birds offer an approach to managing this problem, so we evaluated a formulation (ReJeX-iT AG-36) of methyl anthranilate (MA) in aviary and field tests to assess its potential as an avian feeding deterrent for rice seed. In a feeding trial with an untreated commercial ration as the alternative food, MA suppressed ($P < 0.05$) rice consumption at 2.5% (g/g) but not at lower rates. With untreated rice as the alternative food, however, repellency occurred at 1.0% MA ($P < 0.05$). Controlled field trials showed that seed loss from plots having a 1.7% MA treatment averaged 27 and 34% compared with control plot losses of 52 and 73%. We conclude that MA has potential in the management of blackbird damage to rice, particularly if MA residues on rice seed can be prolonged.

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The bird repellent properties of methyl anthranilate (MA) have been known since the 1960s (Kare 1961). From the early 1980s, the feeding deterrence of MA has been applied to various bird management situations (Mason and Clark 1992). Proposed uses for MA and closely related compounds include reducing depredations in feed lots (Mason et al. 1983, Glahn et al. 1989), protecting fruit crops (Askham 1992, Avery 1992), deterring grazing waterfowl (Cummings et al. 1991, 1992, 1995), decreasing exposure to contaminants (Clark and Shah 1993), and discouraging bird use of ponds at airports (Dolbeer et al. 1992).

Seed depredation by blackbirds in rice fields of Louisiana and Texas causes millions of dollars in damage annually (Wilson et al. 1989, Decker et al. 1990). Although research has identified compounds that effectively reduce consumption of rice seed by captive blackbirds (Avery and Decker 1991, Avery et al. 1993), none was registered (by Sep 1994) as a bird repellent seed treatment. Because MA reduced red-winged blackbird food consumption in other trials (Mason et al. 1991), and because MA-based products are registered for other bird management uses (P. F. Vogt, PMC Spec., Cincinnati, Oh., pers. commun.), we evaluated ReJeX-iT for bird repellency on rice seed. Our objectives were to measure captive red-winged blackbirds' responses to treated rice in feeding tests with 2 alternative foods, and then to assess repellency in controlled tests under field conditions.

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METHODS

Cage Tests

We trapped male red-winged blackbirds in Alachua County, Florida, and held them outdoors in group cages (1.2 × 1.2 × 1.7 m) with access to F-R-M Game Bird Starter (Flint River Mills, Bainbridge, Ga.) and water for 1–3 months prior to testing. We captured, maintained, and tested birds following animal care procedures approved by the Animal Care and Use Committee of the Denver Wildlife Research Center. Four days before the start of the pretreatment period, we removed birds from their group holding cages, determined their mass, and assigned them to individual test cages (45 × 45 × 45 cm) in an outdoor aviary. We formed treatment groups of 6 birds each by randomly assigning treatments to cages. During the acclimation period, we provided birds with 2 clear-plastic food cups (8.2-cm diam, 3.2 cm high, with a 3.1 cm opening in the top), each of which contained a mixture of untreated rice and Game Bird Starter.

We conducted 2 2-cup tests, each involving different sets of birds. Test procedures were identical except that in test 1, the alternative food was Game Bird Starter and in test 2 the alternative food was untreated rice. Following acclimation, there was a 5-day pretreatment period, a 2-day break, and a 5-day treatment period. Daily during pretreatment and treatment periods, we randomly assigned 1 cup in each cage as the treated cup. During pretreatment, the treated and untreated cup each contained 30 g of untreated food. In the treatment phase, the designated treated cup contained 30 g of MA-treated rice seed and the alternative cup held 30 g of Game Bird Starter (test 1) or untreated rice (test 2).

The density of MA is 1.168 g/mL (Windholz 1983). We used MA that was formulated as ReJeX-iT AG-36 (PMC Spec., Cincinnati, Oh.) containing 14.5% MA (vol/vol) (P. F. Vogt, pers. commun.). We calculated the volume of ReJeX-iT needed for seed treatment rates of 0.0, 0.1, 0.5, 1.0, and 2.5% MA (g/g). Throughout, we express concentration levels in terms of the active ingredient, MA, and not the formulated product, ReJeX-iT. We prepared seed in 5-kg batches by adding the appropriate volume of ReJeX-iT to rice in the receptacle of a rotating mixer. Mixing continued for ≥15 minutes to ensure coverage. Then we airdried each batch.

Throughout pretreatment and treatment periods, we removed maintenance food cups at 0700, and 1 hour later administered test food cups. We exposed 2 food cups holding the test seed and alternative food to ambient conditions to determine moisture gain or loss. After 3 hours, we removed the test food and provided maintenance food again. Each day, we measured test food spilled on aluminum trays beneath each cage. After the study, we banded and released all birds.

For each bird, we estimated daily consumption by subtracting the food in each cup after the trial from 30 g, corrected for moisture gain or loss, and then adding the mass of spilled food. Because we detected no pretreatment differences in consumption among groups or between cups, we analyzed the 5-day treatment periods only in repeated measures analysis of variance (ANOVA) with MA level as the independent factor and day and cup as repeated factors. Tukey's HSD (Steel and Torrie 1980) isolated differences among means ($P < 0.05$). Power analysis (Steel and Torrie 1980:113) revealed that at $\alpha = 0.05$, we could detect differences in consumption among MA levels ≥0.5 SD with a Type II error of 0.29.

Field Trials

We conducted field trials at the Louisiana State University Rice Experiment Station in Crowley, Louisiana, and at the Texas A&M University Agricultural Research and Extension Center in Beaumont, Texas. At each location, we prepared a 0.2-ha site according to standard practices (Tex. Agric. Ext. Serv. 1993) and divided each into 3 9- × 44-m plots. We prepared MA-treated rice using ReJeX-iT AG-36 as before. We selected MA treatment levels (0.7 and 1.7%) intermediate to those tested in the cage trials to extend our knowledge of blackbird responses to various levels of MA, and we used 1 high level (6.7%) that we felt would ensure effective repellency. In Louisiana, 1 test plot received MA-treated rice at 1.7% (g/g) and another received rice treated at 6.7% MA. The third plot received untreated rice. Levels tested in Texas were 0.7 and 1.7% MA. We applied seed with a hand-operated centrifugal spreader calibrated to deliver 113 kg/ha.

One day after seeding, we erected 3 netted enclosures (2.4 × 6.1 × 1.8 m) on each of the 9- × 44-m plots. We provided each enclosure with a shaded, centrally located perch and waterer, and within each enclosure, we established 4 pairs of 0.19-m^2 sampling quadrats. One randomly selected quadrat of each pair was protected with a welded-wire bird exclosure. The other quadrats were open. We erected an electric fence around each site to discourage mammalian predators.

On the second day after seeding, we released 3 locally trapped male red-winged blackbirds into each enclosure, and we collected a water sample for residue analysis. In the test enclosures, the birds received a bowl with 200 g of rice as an alternative to the rice seed in the plot. On day 4, we counted rice sprouts remaining in each of the open and protected sampling quadrats, and we measured the mass of rice seed left in the alternative food bowl. We collected a composite 20-g seed sample from each treatment level for residue analysis. We then moved the enclosures to new locations within the study plots and repeated the test procedure (including seed collection for residue analysis) during days 5–8 and 9–12 after seeding. We released birds after the last test day.

We estimated sprout loss by subtracting the number of sprouts in each open sampling quadrat from that in the paired protected quadrat and expressed the difference as a percent of the protected quadrat count. Because percent data were not normally distributed (MINITAB 1989), we applied arcsine transformation (Steel and Torrie 1980). Transformed data were normally distributed (MINITAB 1989), and we examined effects of MA level and time in a 2-way repeated measures ANOVA. We assessed alternative food consumption in a 2-way (MA level and test period) ANOVA.

Methyl Anthranilate Residue Determination

We analyzed only Louisiana samples because Texas samples were lost during shipment. The analytical technique we used determines only the MA bound to the seed surface. Free MA is not determined.

We dried rice seeds in high vacuum (oil pump) for 30 minutes prior to analysis. Then we placed a measured amount (approx 50 g) of dried seed into the glass chromatographic column (35-mm inner diam, 50-cm length). We filled the column with distilled water, connected it to the peristaltic pump, and passed water through the column at 5 mL/minute. We collected fractions of outcoming solution every 45 minutes. We determined MA concentration in each fraction by high performance liquid chromatography (HPLC; Zorbax ODS 4.6 × 250 mm, acetonitrile-water 1:1; 1 mL/min, detection at 330 nm) and calculated the initial concentration of bound MA in the seed sample. We analyzed aqueous samples directly by HPLC under the same conditions. We estimated the error rate of HPLC readings at ±3%.

The extraction process was approximated by the equation $CV = C_{s0}e^{-At}$, where C was the concentration of MA (mg/mL) in outcoming water at time t (min), V was the flow rate of water through the column (mL/min), C_{s0} was the concentration of MA in the sample of seeds at the beginning of the process (mg/g of seed), and A was a constant incorporating mass transfer coefficient of the process, partition coefficient of MA between phases, and size of microcapsules and seeds. Thus, logarithm of CV is a linear function of time, and values of A and C_{s0} can be determined by least squares method from the plot of $\ln(CV)$ versus time.

RESULTS

Cage Tests

Game Bird Starter as Alternative Food.—Total food consumption did not differ ($F = 2.05$; 4, 25 df; $P = 0.118$) among MA levels. Total consumption increased ($F = 19.74$; 4, 100 df; $P < 0.001$) from 1.44 g/cup (SE = 0.15) on day 1 to 1.82 g/cup (SE = 0.22) on day 5. Overall, consumption of rice (2.31 g/bird, SE = 0.12) exceeded ($F = 16.95$; 1, 25 df; $P < 0.001$) that of Game Bird Starter (0.95 g/bird, SE = 0.08).

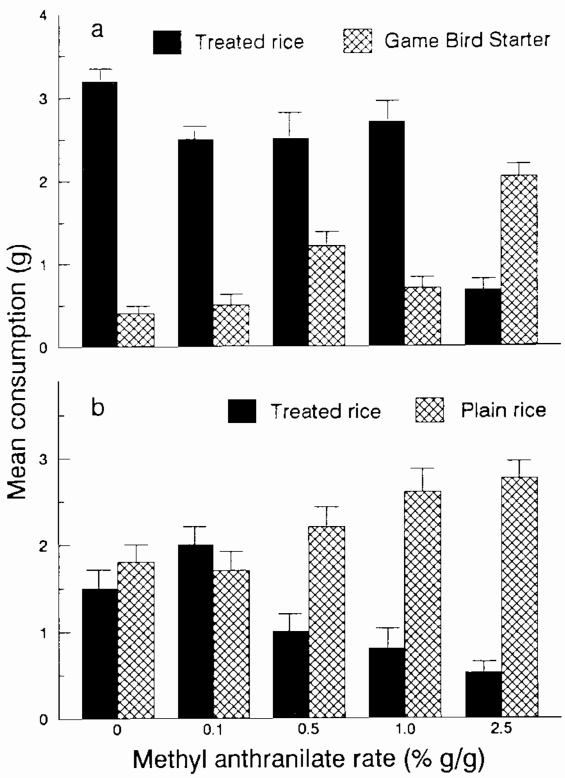

Fig. 1. Mean daily consumption of methyl anthranilate-treated rice by captive red-winged blackbirds in feeding trials when the alternative food was (a) Game Bird Starter or (b) untreated rice, Gainesville, Florida, January–March 1993. Capped bars denote 1 SE.

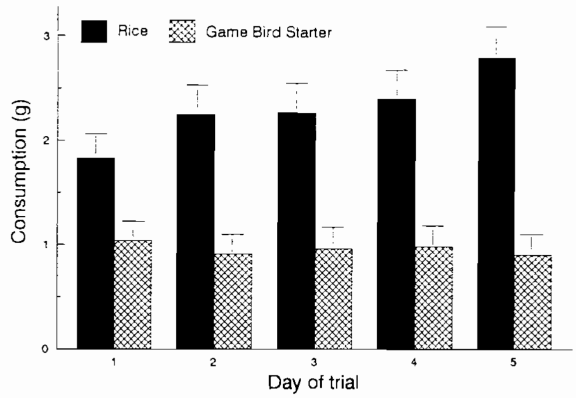

Fig. 2. Mean daily consumption by red-winged blackbirds of methyl anthranilate-treated rice and Game Bird Starter during a 5-day feeding trial, in Gainesville, Florida, 1993. Capped bars denote 1 SE.

The interaction (F = 3.40; 16, 100 df; P < 0.001) between group and day reflected daily variation in total consumption among treatment groups. In particular, consumption by the 2.5% group was lowest on day 5 whereas for each of the other groups consumption was highest on day 5.

The group-cup interaction (F = 4.83; 4, 25 df; P = 0.005) resulted from the preference for Game Bird Starter exhibited by the 2.5% MA group, whereas the other groups consumed rice preferentially (Fig. 1a). The day-cup interaction (F = 7.44; 4, 100 df; P < 0.001) reflected increasing disparity in consumption between rice and Game Bird Starter over days (Fig. 2). There was no 3-way interaction (P = 0.204).

Rice as Alternative Food.—Consumption did not vary (F = 0.35; 4, 25 df; P = 0.840) among groups. Over days, consumption increased (F = 11.24; 4, 100 df; P < 0.001) from 1.41 g/cup (SE = 0.15) on day 1 to 1.82 g/cup (SE = 0.18) on day 5. Consumption from the untreated cup (2.21 g/bird, SE = 0.10) exceeded (F = 37.39; 1, 25 df; P < 0.001) that from the treated cup (1.17 g/bird, SE = 0.10).

The group-cup interaction (F = 7.88; 4, 25 df; P < 0.001) reflected increasing disparity in consumption of MA-treated and untreated rice with increased treatment levels (Fig. 1b). Consumption of treated and untreated rice differed (P < 0.05) in the 1.0 and 2.5% MA groups. There was no 3-way interaction (P = 0.223).

Field Trials

Seed Loss.—In Louisiana, seed loss varied among treatment levels (F = 21.57; 2, 6 df; P = 0.002). Four days after seeding, loss of seed from control pens was 39% and averaged 52.3% throughout the trial (Fig. 3). In contrast, estimated loss of seed treated with 1.7 and 6.7% MA was 11 and 6% after 4 days, and overall averaged 26.7 and 17.7%, respectively (Fig. 3).

Across all treatments, seed loss increased (F = 10.89; 2, 93 df; P < 0.001) from 18.7% 4 days after seeding to 40.7 and 37.3% 8 and 12 days after seeding, respectively. The interaction (F = 3.41; 4, 93 df; P = 0.012) between treatment level and day reflected the increase in seed loss from MA-treated pens with time compared with an increase and then a decrease in loss from control pens. Rain flooded the alternative food bowls on several occasions and prevented assessment of alternative food consumption.

In Texas, seed loss varied among MA treatment levels (F = 9.69; 2, 6 df; P = 0.013). After 4 days, seed loss was 58% (SE = 7) in the control plot, 55% (SE = 4) in the 0.7% plot, and 32% (SE = 5) in the 1.7% plot. Overall, seed loss in

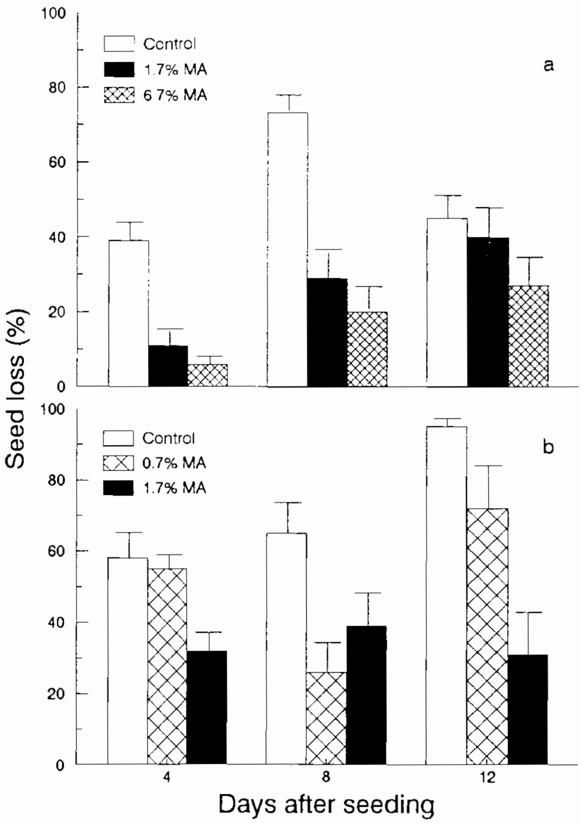

Fig. 3. Removal of methyl anthranilate-treated rice seed from sampling quadrats within test enclosures at (a) Crowley, Louisiana, and (b) Beaumont, Texas, April–May 1993. Each pen held 3 red-winged blackbirds. Capped bars denote 1 SE.

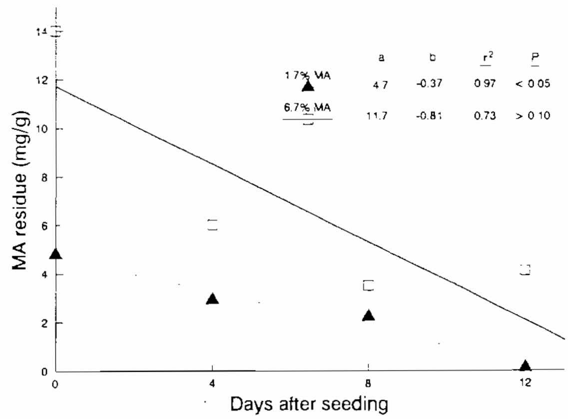

Fig. 4. Residues of methyl anthranilate (MA) from rice seeds treated at 1.7 and 6.7% (g/g) and planted in flooded test plots in Crowley, Louisiana, May 1993. Linear regression lines are described by the equation Y = aX + b, where Y is the MA residue measured on the sample and X is the days after seeding.

control plots was 74% (SE = 4) and 51% (SE = 6) in the 0.7% MA plots, and 34% (SE = 5) in the 1.7% MA plots.

Across all groups, seed loss increased (F = 13.42; 2, 93 df; $P < 0.001$) from 48% (SE = 4) 4 days after seeding to 66% (SE = 7) 12 days after seeding. The interaction (F = 4.77; 4, 93 df; $P = 0.002$) between treatment level and day reflected the increase in seed loss by the control and 0.7% groups over time compared with the consistent level of seed loss in the 1.7% MA groups (Fig. 3). Rice consumption from the alternative food bowl was greater (F = 21.17; 2, 18 df; $P < 0.001$) in the 1.7% MA groups (80.1 g/pen, SE = 5.1) than in the control (39.9 g/pen, SE = 4.7) or 0.7% MA groups (32.7 g/pen, SE = 6.7).

Methyl Anthranilate Residues.—We recovered no MA from the control seed samples or from the water samples taken from the plots in Louisiana. In Louisiana, residues on the 1.7% MA-treated seed declined linearly ($P < 0.05$) with days after seeding (Fig. 4). Residues on the 6.7% MA seed appeared to decline more rapidly, but the relationship was not significant ($P > 0.05$).

DISCUSSION

The feeding trials demonstrated the importance of alternative food in assessing blackbird responses to MA-treated seed. The birds' preference for rice over Game Bird Starter resulted in persistent feeding on treated rice up to the 2.5% MA level. These results are similar to those obtained by Mason et al. (1991) in which food treated with 1.0% MA with no alternative only temporarily repelled red-winged blackbirds. Apparently, red-winged blackbirds will tolerate relatively high levels of MA on dry food if alternative food is absent or not highly preferred. Redwings readily discriminated and were deterred by MA levels as low as 0.5% when the alternative to treated rice was untreated rice.

Our findings suggest that one of the principal factors affecting repellency of MA-treated rice seed to red-winged blackbirds is the nature and availability of alternative foods. If newly sown rice is all that is available, then redwings may not be deterred by MA treatment.

Apparently, however, other foods are available to redwings during the March–April rice depredation period (Meanley 1971). Wilson (1985) found that rice composed just 20–28% (by vol) of the food eaten by redwings in southwestern Louisiana during March and April. Weed seeds constituted 50% of the birds' diet. Wilson (1985:63) noted that in March and April rice represented a smaller portion of the birds' diet than it did during fall and winter, possibly due to "increased availability of weed seeds and insects that accompany the preparation of land for planting."

MANAGEMENT IMPLICATIONS

Whereas MA might make rice seed less palatable to blackbirds, it may not be realistic to view MA-treated seed as the solution to bird depredations to rice. Instead, MA treatment should be viewed as 1 potential component of blackbird management strategies that could involve other tactics such as selective lethal control (Glahn and Wilson 1992) and alternative planting practices (Wilson et al. 1989). Also, MA might prove more effective combined with other types of repellent seed treatments (Avery and Decker 1991, Avery et al. 1993) than as a seed treatment deterrent by itself.

A bird's handling of a rice seed could affect its exposure to a chemical treatment. For example, in feeding trials of an insecticidal rice seed treatment (Avery et al. 1994), approximately 85% of the chemical originally applied to the seed remained on the discarded hull after red-winged blackbirds fed on the treated seed. Although MA is a contact irritant and thus does not need to be ingested, the birds' handling of the seed in our trials may have exposed them to only a fraction of the applied chemical. The MA formulation that we used was not designed specifically for rice seed treatment and MA did not persist on rice. Effective bird depredation management in seeded rice will depend upon extending MA activity to cover the 2–3 week damage period and also increasing the exposure of the bird to the repellent, perhaps by prolonging seed handling time (Daneke and Decker 1988).

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