

same combination as that in monoferric phytate. This conclusion concerning the nature of most of the endogenous iron in wheat bran explains the finding that the iron in bran and monoferric phytate was equally bioavailable (Morris and Ellis, 1976).

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## Laboratory Comparisons of Polyphenols and Their Repellent Characteristics in Bird-Resistant Sorghum Grains

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Laboratory evaluations of repellency and polyphenol composition were conducted for 15 varieties of bird-resistant (BR) sorghums. Tests involved the weaver finch (*Quelea quelea*) and the red-winged blackbird (*Agelaius phoeniceus*). We compared BR varieties with a bird-susceptible (Martin X) sorghum by using a two-choice paired preference test under light and reduced lighting (near darkness) conditions. The most important observation of this study was recognition of the diversity of polyphenolic properties among BR sorghums. The 15 varieties were separated into three groups (seven least, seven intermediate, and one most preferred) based on preference response. In most instances, polyphenol values from modified vanillin-HCl and Sephadex LH-20 gel permeation chromatography analyses of these varieties could be placed in similar groups. The seven least preferred sorghums, with the exception of WGF, were uniform in polyphenol properties whereas substantial variation occurred among the remaining eight varieties. Several observations involving test conditions, bird species, and sorghum properties were also discussed.

Bird damage to sorghum crops [*Sorghum bicolor* (L.) Moench] is so severe in many parts of the world that control measures must be taken or most of the crop will be lost. The most common practice is to grow sorghums which are astringent during the immature stages when bird damage is normally the highest (Doggett, 1957; McMillian et al., 1972). In many varieties, however, the polyphenolic tannins which impart bird-repellent properties are also present in the mature grain and lower their palatability and nutritional qualities for the consumer (Harris, 1969; Mabbayah and Tipton, 1975). The result is that these high-tannin "bird-resistant" (BR) sorghums have less value on the export market (Price et al., 1979) and farmers that produce them are at an economic disadvantage. Many farmers in this country have need to include sorghum in their crop rotation program, and the economic disadvantage is especially serious for farmers in arid regions of the

world. Many cannot otherwise protect their sorghum crops and cannot grow wheat, corn, or rice as an alternative.

Currently, the repellent characteristics of BR sorghums are attributed to the group of polyphenolics known as "condensed tannins". Specifically, the references are usually to proanthocyanidins which are a series of condensed flavon-3-ol and flavan-3,4-diol molecules of increasing complexity. The term "tannin" is generally reserved for those polymers having molecular weights between 500 and 3000 which form stable complexes with proteins (Ribereau-Gayon, 1972). In the human mouth, these tannins elicit an "astringent response"—a contracting or drying feeling caused by the precipitation of proteins in saliva and on mucousal surfaces (Joslyn and Goldstein, 1964; Singleton and Noble, 1976). Astringency generally increases with increased polymerization up to an intermediate molecular weight (e.g., hexamer or heptamer) and then decreases as the molecule becomes insoluble and too large to effectively bind with proteins (Goldstein and Swain, 1963; Ribereau-Gayon, 1972). Apparently, any characteristic that influences the protein binding properties of a molecule also influences its activity in other biochemical processes such as leathering of hides (Gus-

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tavson, 1956), inhibition of enzymes (Byrde et al., 1960), weathering of grains (Mabbayad and Tipton, 1975), and reduction of protein digestion (McGinty, 1969; Oswalt, 1975).

In beginning this study we believed that a better understanding of the chemical and biochemical properties of sorghum polyphenolics would help in the genetic selection of varieties which would be satisfactory to both the producer and consumer. One hindrance to this selection had been the lack of definitive laboratory investigations of repellency associated with BR sorghums. Almost every publication has involved field investigations where a multitude of other variables (configuration, alternate food supply, nesting behavior, etc.) may have influenced the bird's response to the sorghum grains. Thus, our first objective was to conduct a laboratory evaluation of wild bird repellency where factors other than taste preference could be minimized (Bullard and Shumake, 1979). Second, we wanted to determine the relation, if any, between the repellency and the composition of polyphenolics in BR sorghums.

## EXPERIMENTAL SECTION

**Birds.** Two species of granivorous birds representing the Ploceid and Icterid families were tested: quelea (*Quelea quelea*) and red-winged blackbirds (*Agelaius phoeniceus*). Quelea were wild-trapped in Tanzania, flown to the Denver Wildlife Research Center, and held for a 90-day quarantine and acclimatization period before the preference tests. Similarly, blackbirds were trapped in the Platte River Valley of Colorado and held for at least 90 days before testing. Throughout these 90-day periods, all birds were presented with ad libitum water, grit, and a maintenance ration of whole grain Martin X sorghum or cracked corn, proso millet, and Purina Game Bird Startena in large ( $2.4 \times 4.8 \times 2.1$  m for quelea and  $3\text{ m}^3$  for blackbirds) communal cages.

**Sorghum Grains.** Cooperators supplied 1 kg or more of mature sorghum from 15 BR varieties. A wide range of physical and chemical characteristics were represented. The varieties were as follows: BR 54, WGF, RA Bird Go 68, Funks G516-BR, AR 3005, IS 2403c, AKS 614, IS 3063c, Hegari, ROKY 78, TAM 2566, IS 2801c, IS 2266c, AR 3009, and BR-ATG-051.

All test sorghums were ground to 40 mesh and analyzed immediately by the respective chemical assay. For preference tests, they were ground to 20 mesh and pressed into 2-mm diameter  $\times$  4-mm long pellets for quelea and 3.5-mm diameter  $\times$  8-mm long pellets for redwings the day before each test series and stored in capped bottles at  $-10^\circ\text{C}$  until used. A low-tannin variety with red pericarp (Martin X; 0.67 catechin equivalent total polyphenols by method 1, discussed later) was selected as the control to minimize the differences in color between it and the 15 BR varieties.

The different sorghum pellets varied markedly in coloration; pellets of BR sorghum ranged from light yellow to dark brown. In earlier tests, visual cues from food color differences influenced the food preference response (Bullard and Shumake, 1979). Also, a search of the literature revealed substantial differences in opinion as to the association between pigmentation and biochemical activity of sorghums. Thus, we had 10 individuals order the pellets of BR sorghums in terms of increasing coloration intensity (mean rank) in an attempt to assess this property.

**Preference Test Procedure.** Details of the preference test procedure used in this study are reported elsewhere (Bullard and Shumake, 1979). Briefly, test birds were adapted to Martin X pellets in communal cages (8–10

birds/cage), weighed, and transferred to individual test cages on a five-tier rodent rack of  $44 \times 25 \times 20$  cm cages. For quelea, the cages were divided by wire mesh for testing pairs of birds (one bird on each side of the divider); we tested blackbirds individually by using these cages but without dividers.

Reduced lighting (darkened) minimized the influence of food coloration on the food preference response of quelea in earlier tests (Bullard and Shumake, 1979). Consequently, separate groups of quelea and blackbirds were tested under fluorescent light (day) and darkened (1.8 and  $-1.3 \log \text{ft L}$ , respectively, at 30 cm from the food cups) conditions for the daily "on" light period.

In the preference tests, five male birds were each offered a choice between pelleted Martin X and one of the BR varieties for 4 consecutive days. The two foods were placed in separate cups 5 cm apart at the front of each cage, and cup positions were alternated daily to decrease the effects of learned position habits. Weighed amounts of food (5 g for quelea and 10 g for redwings) were placed in each cage at the beginning of each test day (12 h of fluorescent lighting and then 12 h off), and the unconsumed remainder was weighed the following morning. The difference, corrected for spillage, was recorded as the quantity consumed. BR preference measures were computed as percentages of total grain consumption across the 4 days of tests:

% preference =

$$[[\text{BR sorghum consumption (g)}]/[\text{BR sorghum + Martin X sorghum consumption (g)}]] \times 100$$

**Chemical-Analytical Procedures.** Total polyphenolics were determined by two methods: the modified vanillin–hydrochloric acid method (method 1) of Maxson and Rooney (1972) and their method as modified (method 2) by Price et al. (1978). Samples were extracted with 1% concentrated HCl in methanol for both methods. The primary changes in the modification of Price et al. (1978) were a shorter extraction time and the subtraction of blank absorbance from sample absorbance ( $\Delta A_{500}$ ) at 500 nm on a Bausch and Lomb Spectronic 20 or Beckman DK-2A spectrophotometer.

The molecular characteristics of tannins for each respective variety were determined by a slight modification of the Sephadex LH-20 gel permeation chromatography procedure of Strumeyer and Malin (1975). Sephadex LH-20 separates compounds on the basis of their molecular weight and adsorptive characteristics. Both size (Goldstein and Swain, 1963; Ribereau-Gayon, 1972) and shape (Quesnel, 1968) influence the ability of condensed tannin molecules to interact with protein molecules. Since tannin properties are manifested in this interaction, a chromatographic separation which involves both size and shape can be useful in evaluating the tannin characteristics of a given variety.

A ground 30-g sample was defatted by shaking it with 100 mL of diethyl ether (2 $\times$ ), and the extract was discarded. The defatted sample was then extracted for 1 h with 100 mL of acetone (2 $\times$ ) and then methanol (2 $\times$ ). These extracts were combined, concentrated to 10 mL by rotary evaporation, and held at  $-10^\circ\text{C}$  until the day of analysis. Then a 50- $\mu\text{L}$  aliquot (equivalent to 150 mg of sample) was injected on a 1-cm diameter  $\times$  30 cm Sephadex LH-20 column and eluted with 75 mL of absolute ethanol, followed by 75 mL of 95% ethanol and finally 90 mL of 1:1 acetone–water for elution of the tannins. Fractions (3 mL) of eluate were read at 340 nm on a Bausch and Lomb Spectronic 20 and plotted according to absorbance. The areas of nontannin (alcohol eluate) and tannin (50% acetone eluate) regions of the chromatogram

Table I. Mean Percent Preferences<sup>a</sup> of *Q. quelea* and *A. phoeniceus* (Red-Winged Blackbirds) for 15 Pelleted BR Sorghums

variety	mean % preference of <i>Q. quelea</i>		mean % preference of <i>A. phoeniceus</i>		overall mean % preference <sup>b</sup>
	darkened	day	darkened	day	
BR-54	3.8	17.4	7.3	26.2	13.7 <sup>e</sup>
WGF	0.6	17.8	1.1	37.2	14.2 <sup>e</sup>
RA Bird Go 68	2.6	4.3	10.7	43.0	15.1 <sup>e</sup>
Funks G516-BR	7.0	13.9	6.2	34.3	15.4 <sup>e</sup>
AR 3005	10.8	21.1	2.9	27.7	15.6 <sup>e</sup>
IS 2403c	19.3	24.3	11.5	29.7	18.7 <sup>de</sup>
AKS 614	4.8	26.1	11.1	40.3	20.5 <sup>cde</sup>
IS 3063c	29.0	38.6	7.0	38.2	28.2 <sup>bcd</sup>
Hegari	36.5	46.1	2.1	38.7	29.4 <sup>bcd</sup>
ROKY 78	26.5	44.1	4.3	49.0	31.0 <sup>bc</sup>
TAM 2566	51.8	31.7	14.6	34.4	32.5 <sup>bc</sup>
IS 2801c	36.6	34.6	11.0	52.3	33.4 <sup>ab</sup>
IS 2266c	26.4	50.4	14.8	45.4	34.2 <sup>ab</sup>
AR 3009	47.6	29.5	23.2	38.3	34.6 <sup>ab</sup>
BR-ATG-051	25.6	53.5	51.3	51.6	45.5 <sup>a</sup>

<sup>a</sup> Individual cage tests, 5 birds/treatment, 4 days. The preference response of quelea or red-winged blackbirds when given a choice between the test food and pelleted Martin X control was % preference = [(test food consumed)/(test + control food consumed)] × 100. <sup>b</sup> Values followed by the same superscript letter(s) are not significantly different at the 0.05 level of Duncan's Multiple Range Test.

were determined in square centimeters by planimetry.

**Statistical Analyses.** We analyzed percentage measures of BR sorghums ingested relative to the Martin X variety by using a three-way complete factorial (15 BR sorghums × 2 bird species × 2 light levels) analysis of variance (ANOVA) (Winer, 1971). Significant treatment effects were further analyzed by the Duncan Multiple Range Test (Duncan, 1955).

The mean percentage preference relationship to mean catechin equivalents (CE) was determined by regression analyses. Linear regressions were computed between preference measures for quelea-day, quelea-darkened, blackbird-day, and blackbird-darkened feeding periods and each of the two modified vanillin-HCl (MV-HCl) methods. In addition, similar regressions were computed between the overall mean percentage preference of each BR variety (i.e., measurements combined across bird species and light levels) and total polyphenolic estimates obtained by MV-HCl methods 1 and 2, pellet coloration, and chromatogram values for tannins and nontannins.

The 0.05 level of significance was used with the statistical tests.

## RESULTS

Overall, an important consideration concerning the bird response to BR sorghums was the reduced preference (<50%) for candidate BR varieties in all but six of the tests. The BR candidate varieties were generally less preferred than the Martin X control. Analysis of variance revealed significant differences in BR sorghum preferences for two main and two interaction effects: BR sorghums ( $F = 7.46$ ;  $df = 14/240$ ), light levels ( $F = 89.90$ ;  $df = 1/240$ ), BR sorghums × bird species interaction ( $F = 3.13$ ;  $df = 14/240$ ), and bird species × light levels interactions ( $F = 21.90$ ;  $df = 1/240$ ).

Regarding the main effect of BR sorghums, an overlapping pattern of mean differences for Duncan's Multiple Range Tests was obtained (Table I). This pattern can be summarized based on three main points: (a) mean percentage preference values for BR-54, WGF, RA Bird Go

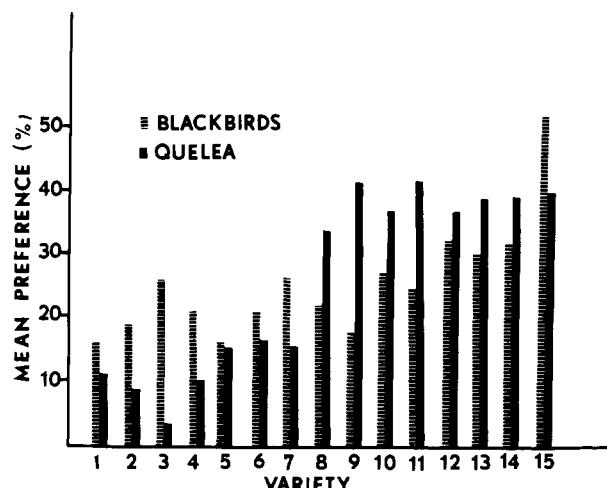


Figure 1. Interaction between bird species and sorghum varieties for a three-factor complete factorial (15 BR sorghums × 2 bird species × 2 light levels) analysis of variance. Varietal identification: 1 = BR-54, 2 = WGF, 3 = RA Bird Go 68, 4 = Funks G516-BR, 5 = AR 3005, 6 = IS 2403c, 7 = AKS 614, 8 = IS 3063c, 9 = Hegari, 10 = ROKY 78, 11 = TAM 2566, 12 = IS 2801c, 13 = IS 2266c, 14 = AR 3009, and 15 = BR-ATG-051.

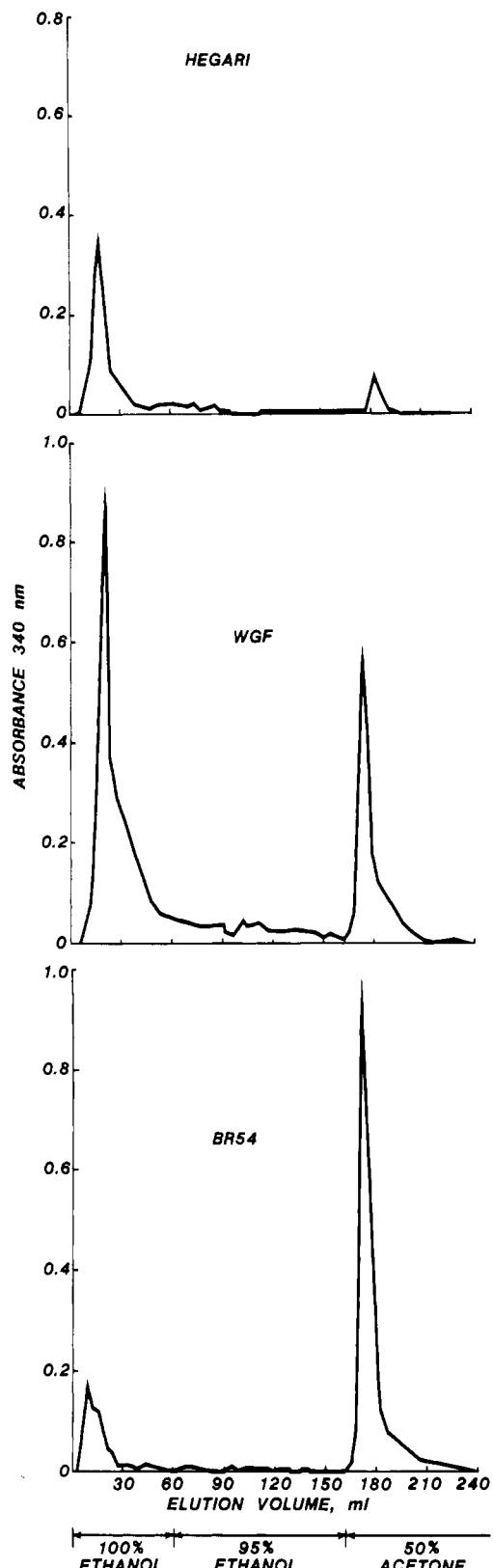
68, Funks G516-BR, AR 3005, IS 2403c, and AKS 614 were significantly less than those of the remaining varieties (i.e., mean preferences ranged between 13.7% for BR-54 and 20.5% for AKS 614) but not essentially different from each other; (b) mean percentage preferences for IS 3063c, Hegari, ROKY 78, TAM 2566, IS 2801c, IS 2266c, and AR 3009 were intermediate in magnitude (i.e., mean preferences ranged between 28.2% for IS 3063c and 34.6% for AR 3009), not differing greatly among each other but being more preferred than the previous seven varieties; (c) varieties in both of the foregoing sets were significantly less preferred than BR-ATG-051 (i.e., 45.5% mean).

The main effect of light levels revealed that lighting significantly influenced the preference response of all the birds. The mean preference response under day conditions was 16.9% compared with 34.6% for the darkened conditions.

The pattern of differences in mean preferences for the BR sorghum × bird species interaction was complex. Results are illustrated in Figure 2. As shown, all preference means for quelea were either below 16% or above 32% whereas 13 of the 15 redwing means ranged between these two values. The seven least preferred BR varieties (i.e., BR-54, WGF, RA Bird Go 68, Funks G516-BR, AR 3005, IS 2403c, and AKS 614) had lower percentage preference values (except for AR 3005) for quelea than for blackbirds whereas the reverse occurred for the remaining varieties (except for BR-ATG-051). The only significant differences in mean preferences for quelea and redwings occurred with RA Bird Go 68 (3.5% for quelea and 26.9% for blackbirds) and WGF (9.2% for quelea and 19.2% for blackbirds).

Mean comparisons for the bird species × light level interaction (blackbird-day = 11.9%, quelea-day = 21.2%, quelea-darkened = 30.2%, and blackbird-darkened = 38.4%) were all significant. Note that the mean preferences for both species are lower under day conditions—a result previously confirmed for the light levels main effect. However, decreased illumination elicited a much greater shift in mean preference for blackbirds than for quelea; compared to quelea, blackbirds appear to be more reliant on visual cues when feeding on somewhat repellent foods.

Significant negative correlations were found between polyphenol content and BR sorghum preferences for all



**Figure 2.** Sephadex LH-20 gel permeation chromatograms of extracts from three bird-resistant sorghums. Nontannins were removed with ethanol and 95% ethanol followed by tannins which eluted with 50% acetone.

analytical methods and preference tests with quelea. Only one significant correlation between polyphenols and preference was noted for blackbirds; the polyphenol content of BR sorghums as determined by method 1 was negatively correlated with preference under day conditions ( $r = -0.60$ ). The overall mean percentage preference for

both species under both conditions of illumination correlated significantly with both methods of polyphenol analysis (method 1,  $r = -0.83$ ; method 2,  $r = -0.69$ ). Thus, high polyphenol content appears to be negatively associated with lower preferences for BR sorghums relative to Martin X.

Pellet coloration was not significantly correlated with overall preference ( $r = -0.38$ ) or with either polyphenol assay (method 1,  $r = 0.21$ ; method 2,  $r = 0.24$ ).

When peak areas from chromatograms of BR sorghum extracts were compared with the overall mean preference values (Table II), a significant negative correlation was found between tannins and preference ( $r = -0.74$ ) but not between nontannins and preference ( $r = -0.39$ ). A correlation was also found between catechin equivalents (CE) of total polyphenols for both methods of analysis and the overall mean percentage preferences of birds for BR sorghums (method 1,  $r = -0.83$ ; method 2,  $r = -0.69$ ).

#### DISCUSSION

Overall the most important observations in this study were (1) the laboratory confirmation of "taste"-repellent properties of BR sorghums to wild birds and (2) delineation of large variations in polyphenol properties among the 15 BR sorghums. Also important was the recognition that varieties with poor nutritional reputations were among the seven less preferred BR sorghums and this group yielded the most uniform chemical and behavioral effects.

Chromatograms in Figure 1 illustrate the variations in polyphenolic properties among BR sorghums. Differences in polyphenol analyses and in varietal preference further highlight the variability in constituents of BR sorghums. Based on the current data, we believe that these 15 varieties were clearly separable into two groups according to chemical and preference data. The first set consisted of the seven least preferred varieties (Table I). This group was fairly uniform (WGF was the most variant for all chemical and behavioral measurements) and contrasted sharply with the second set of the remaining eight varieties.

An interesting observation was that varieties which have been associated with deleterious nutritional properties were all in the first set of sorghums. All had method 1 MV-HCl values above 3 CE, whereas none of the remaining sorghums were that high (Table II). When blank corrections ( $\Delta A_{500}$ ) were made to correct for nontanning pigments (Price et al., 1978) in method 2, all varieties in the least preferred (first) set had at least 35% of the CE of method 1, compared with only four of the sorghums in the second set (i.e., ROKY 78, IS 2801c, AR 3009, and BR-ATG-051). Additionally, all seven of the least preferred set had a large tannin peak ( $>200 \text{ cm}^2$ ) compared with the remaining varieties (except IS 2801c which had  $314 \text{ cm}^2$  in set 2) and all had greater than 50% (except WGF) relative tannin area with a mean of 65.7% compared with 30.5% for the remaining eight varieties.

The literature clearly cautions us against attempts to relate seed color to tannin concentration (Thayer et al., 1957; Harris, 1969; Maxson et al., 1972; Mabbayah and Tipton, 1975; Nelson et al., 1975). We found that pellet coloration was not significantly correlated with tannin content by either of the MV-HCl methods. Since preference data do not indicate a significant sorghum-illumination interaction ( $F = 1.6$ ;  $df = 14/240$ ), we can assume that pigmentation differences did not have any overall effect on bird preference. The significant illumination-bird species interaction might suggest that the influence of decreased illumination is greater for blackbirds than for quelea. However, because blackbird cage sizes were not proportionate to those of quelea and birds were not paired

Table II. Polyphenolic Composition of Bird-Resistant Sorghums

variety	total polyphenols, CE		pellet coloration <sup>c</sup>	peak area, cm <sup>2</sup> <sup>d</sup>		relative area of tannins, %
	method 1 <sup>a</sup>	method 2 <sup>b</sup>		nontannins	tannins	
BR-54	3.59	2.45	5	109	289	72.6
WGF	4.67	1.95	15	530	234	30.6
RA Bird Go 68	4.61	4.05	3	231	278	54.6
Funks G516-BR	3.79	2.25	9	150	488	76.5
AR 3005	3.21	3.50	13	163	345	67.9
IS 2403c	4.30	1.50	12	94	256	73.1
AKS 614	3.07	2.65	14	84	454	84.4
IS 3063c	2.03	0.45	10	92	90	49.4
Hegari	2.62	0.10	1	232	15	6.1
ROKY 78	2.74	1.80	7	141	128	47.6
TAM 2566	1.96	0.05	4	86	8	8.5
IS 2801c	2.35	1.70	11	147	314	68.1
IS 2266c	2.77	0.05	8	101	58	36.5
AR 3009	3.47	1.91	2	169	21	11.0
BR-ATG-051	0.54	0.55	6	80	16	16.7
<i>r</i> <sup>e</sup>	-0.83	-0.69	-0.38	-0.39	-0.74	

<sup>a</sup> Method 1 is the modified vanillin-hydrochloric acid method of Maxson and Rooney (1972). <sup>b</sup> Method 2 is the modified vanillin-hydrochloric acid method of Price et al. (1978). <sup>c</sup> Ranking which increases numerically with the darkness of the pelleted test food. <sup>d</sup> Peak area of gel permeation chromatogram as measured by planimetry. <sup>e</sup> Correlation coefficient (*r*) for relationship between the overall mean percent preference (Table I) and the respective independent variable (i.e., total phenolics, pellet coloration, and chromatogram areas).

as were quelea, we remain cautious in speculations about species differences. The high incidence of brown color among these varieties is undoubtedly the reason why it is associated with deleterious nutritional properties. The color of the whole seed for these seven was dark brown (red tints for WGF and AR 3005) whereas a wide range of combinations including white, red, brown, and purple colors were found in the remaining varieties. Unfortunately, the term "brown sorghums" has been extended to include all sorghums with a pigmented testa, possibly giving some varieties an undeserved "bad" reputation.

The second set of BR varieties (i.e., IS 3063c, Hegari, ROKY 78, TAM 2566, IS 2801c, IS 2266c, AR 3009, and BR-ATG-051) had a comparatively large diversity of chemical and morphological properties.

Varieties like IS 2266c, BR-ATG-051, and IS 3063c are red but have low concentrations of tannin when pigment absorption is subtracted by method 2 MV-HCl analysis. Hegari, AR 3009, and IS 2801c have a white pericarp with a tannin-containing testa underneath. (The pellets were darker than the seeds.) Hegari, TAM 2566, and IS 2266c had the most striking differences in CE values for the two methods of analysis. All three are group II sorghums by the classification method of Price et al. (1978), and this group is recognized as having good nutritional properties (Bullard & Elias, 1980). This would indicate that much of their color is due to nonflavan phenolics. AR 3009 had a low relative tannin chromatogram area (11%), but CE differences in methods 1 and 2 were more comparable to those of the least preferred set. Presumably, the non-tannins measured in this variety were relatively higher in flavanoid compounds which have a phloroglucinol nucleus (Ribereau-Gayon, 1972). BR-ATG-051, which was significantly less preferred than any of the other 14 varieties, exhibited chromatogram properties similar to those of AR 3009 with the exception of being much lower in polyphenolics.

Chromatographic data indicated huge variation among the nontannins. Some could influence the BR properties of select varieties. For example, the chromatographic properties of WGF were much different than those of the other six varieties in the least preferred set. We have since found that a nontannin fraction from Sephadex LH-20 separations of WGF extract was more repellent to quelea

than an equal quantity of the tannin fraction (unpublished data).

In fruits there appears to be a loss of astringency (i.e., tannin activity) as large inactive molecules form from smaller ones during ripening (Goldstein and Swain, 1963). An analogous process probably occurs in sorghums. Our findings indicate that the polymerization process varies considerably among certain varieties. When the 15 varieties were grouped into two sets, we could account for characteristics that have given BR sorghums a bad nutritional reputation in the seven least preferred varieties. On the other hand, the remaining varieties exhibited such a diversity that we were encouraged as to the prospects of plant geneticists being able to selectively develop varieties having improved palatability and BR properties. If BR sorghums are similar to fruits in being most astringent before physiological maturity, some varieties may be developed which have BR qualities in immature stages but a ripened grain of good taste and nutritional quality.

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## Interactions between Agricultural Chemicals and Soil Microflora and Their Effects on the Degradation of [<sup>14</sup>C]Parathion in a Cranberry Soil

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The interaction of selected fungicides, herbicides, and N-fertilizers with microorganisms in cranberry soils and their effects on the degradation of [*phenyl*-<sup>14</sup>C]parathion were investigated. Soil microorganisms were responsible for the oxidative as well as reductive degradation of the insecticide. Incubation of soils with parathion or *p*-nitrophenol for 4 days followed by the addition of [<sup>14</sup>C]parathion resulted after 24 h in an enhanced degradation of the insecticide to <sup>14</sup>CO<sub>2</sub> (34-39% of the applied radiocarbon as opposed to 2% in controls) and also in an increased binding of <sup>14</sup>C to the soil. The fungicide captan inhibited the degradation of soil-applied [<sup>14</sup>C]parathion as evidenced by a reduction of both <sup>14</sup>CO<sub>2</sub> evolution and <sup>14</sup>C-bound residues. Maneb and benomyl suppressed the degradation of [<sup>14</sup>C]parathion to <sup>14</sup>CO<sub>2</sub> but not the formation of bound residues. Pentachloronitrobenzene had no effect. Addition of (2,4-dichlorophenoxy)acetic acid to [<sup>14</sup>C]parathion-treated soil also resulted in an increased persistence of the insecticide. Studies conducted with the insecticide and (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, NH<sub>4</sub>NO<sub>3</sub>, KNO<sub>3</sub>, or urea showed that under all experimental conditions the total amounts of <sup>14</sup>C recovered were similar, yet the distribution of <sup>14</sup>C-labeled compounds into benzene-soluble, water-soluble, and bound residues was not. This possibly indicated a change in the pathway of [<sup>14</sup>C]parathion degradation. The insecticide was most persistent in soils containing (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, as demonstrated by a recovery of 29% of the applied radiocarbon in the benzene-soluble form. Analyses by TLC of this benzene extraction phase revealed the presence of [<sup>14</sup>C]parathion, *p*-amino[<sup>14</sup>C]phenol, and amino[<sup>14</sup>C]parathion. It appears that the form of the N-soil amendment and not the N amendment as such affected the degradation of [<sup>14</sup>C]parathion. Results reported here stress the importance of investigating the environmental fate of a particular pesticide in relation to the presence of other agricultural chemicals.

Cranberries (*Vaccinium macrocarpon* Ait.) are an important fruit crop in the State of Wisconsin. A variety of pesticides and fertilizers are used each year to obtain high yields. Parathion is one of the major insecticides used, being applied at rates ranging from 0.5 to 5 lb/(acre/year). Since during part of the year cranberry bogs are flooded, concern has been expressed about the potential transport of parathion residues by water draining from cranberry bogs. Since fungicides, herbicides, and fertilizers are ap-

plied to cranberry soils, in addition to insecticides, we were interested in the potential interactions of some of these chemicals with the soil microflora and their effects on the degradation of parathion. Previous studies in our laboratory indicated that some herbicides synergized insecticides (Lichtenstein et al., 1973a; Liang and Lichtenstein, 1974) and that the herbicide Eptam affected the uptake and metabolism of [<sup>14</sup>C]phorate in corn plants (Schulz et al., 1976). Gorder and Lichtenstein (1980) demonstrated parathion degradation in soil-free culture media inoculated with microorganisms obtained from cranberry soils. These microorganisms also grew in basal salt media utilizing parathion as the sole carbon source. Addition of 0.05% glucose to the basal salt media inhibited the degradation of parathion.

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