Research Note Behavioral Responses of European Blackbirds and Australasian Silvereyes to Varying Acid and Sugar Levels in Artificial Grapes

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Abstract: Diminishing acid concentrations have long been thought to be one of the effects of ripening grapes that leads to increased bird pressure approaching harvest. Blackbirds (*Turdus merula*) and silvereyes (*Zosterops lateralis*) were offered in a field context varying concentrations of tartaric and malic acids in artificial grapes, where sugar and all other ripening grape parameters were controlled. No linear response of consumption to varying acid concentrations was found for either species. A response to rising sugar was confirmed, but diminishing acid concentrations in ripening grapes appear not to be a contributing factor to increasing bird pressure approaching harvest.

Key words: acid, artificial grape, blackbirds, silvereyes

In most grapegrowing regions bird pressure on ripening grapes begins just after veraison, some two months before winemaking ripeness, and increases dramatically close to harvest. Apart from loss of yield, which in unprotected vineyards can be total, bird peck often results in sour rot, acetobacter, and other molds or insect damage that impact on wine quality. Bird control is a major cost to vineyards growing high-quality grapes in many regions of the world. The hypothesis is widely accepted that high acid content typical of less ripe grapes is not palatable to birds (Boudreau 1972), which may be why bird pressure is lighter on less ripe or high-acid grapes. There is little robust data to support this hypothesis since most research on frugivorous birds has explored either the role of color and sugar in fruit pulp as avian attractants or the effects of seed load on ingestion. Little research has investigated chemosensory cues to birds (Espaillat and Mason 1990). As part of a larger investigation into cues that attract birds to grapes in vineyards (Saxton 2004), the experiments reported here were designed to examine the behavioral responses to various acid concentrations of two bird species that forage intensively on grapes.

Tartaric and malic acids constitute almost all total organic acids in grapes (Coombe 1992, Beriashvili and Beriashvili 1996). Eighty percent of the acid content of grapes is tartaric acid (Lavee and Nir 1986, Hunter et al. 1991), with ~10% malic acid, and lesser amounts of *p*-coumaric and other acids. Typical titratable acidity (tartaric acid equivalents) of unripe grapes can reach 40 g/L and in the course of ripening reduce to less than 10 g/L in ripe grapes. In the course of ripening, glucose and fructose concentrations in the grape rise while acid concentrations concurrently fall. In previous experiments (Saxton et al. 2004c), it was established that sugar was an important cue to grape-eating birds in the vineyard and that preferred concentrations of sugar varied between bird species, which agreed with Boudreau (1972).

Tartaric acid content does not fall much between veraison, when it is at its highest, and harvest, but concentration of tartaric acid is reduced because of enlargement of the grape, which doubles in size (Coombe 1992), and some leakage through breakdown of tonoplast (Terrier et al. 2001). Malic acid in the grape is reduced after veraison through metabolism in the Calvin and Krebs cycles of the photosynthetic process (Doneche et al. 1985), but less so in a cool climate or a cooler season (Lavee and Nir 1986). It is the reduction of malic acid concentration that correlates well with falling titratable acidity as grapes ripen (Barbeau et al. 2004, Doneche et al. 1985, Lavee and Nir 1986, Coombe 1992, Robredo et al. 1991, Terrier et al. 2001).

That chemosensory cues might be important to birds was explored by Espaillat and Mason (1990) who included citric acid in their experiments. For both starlings (*Sturnus vulgaris*) and red-winged blackbirds (*Agelaius phoenicius*), citric acid was tolerated in conjunction with

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The experiments reported here investigated bird responses to the major acid, tartaric; the possibility that reducing malic acid might be an important cue to birds was also explored. Tartaric and malic acids were offered in separate experiments to blackbirds (Turdus merula) and silvereyes (Zosterops lateralis) in a field situation where birds had free choice. Other ripening parameters such as sugar and color were controlled for by using an artificial grape. Because it was thought that the sugar concentration might mask a response to the acid, sugar levels were also manipulated to discover at what concentration rising sugar might drive acid perception down to tolerable levels (Boudreau 1972) or whether there might be an enhancing effect of acid with sugar, which might increase palatability of unripe grapes with low sugar levels. The hypothesis tested was that there would be a significant behavioral response to a level of acid and that this response might be moderated by increasing sugar levels.

Materials and Methods

Sites for blackbird study were at the Lincoln University vineyard. As silvereyes were not present in the university vineyard, experiments were also conducted in a pear orchard ~4 km from the vineyard (silvereyes). Experiments were run in December 2002 with tartaric acid and in May 2003 with malic acid. An initial attempt to study birds during grape ripening showed that birds could not be enticed out of the vines at this time. The artificial grapes were offered on a bird feeder table (Figure 1) on a tripod at eye level, positioned ~8 m from trees that the birds appeared to use for cover.

Twenty grapes were set out on two levels, 10 on the top of the table and 10 on the lower level using a progression design based on a Latin square so that no two

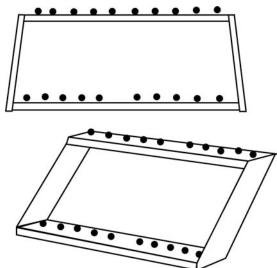


Figure 1 Bird feeder table.

adjacent grapes were the same, nor were any grapes in the same position twice consecutively. For each experiment two each of natural grapes were positioned with two of each of four acid concentrations of artificial grapes to validate the experiment as relevant to the natural environment. The natural grapes were mostly taken first and then the artificial grapes. The natural grapes were excluded from statistical analysis since the controlled level of acid was the factor of interest. Birds were videoed for three hours in the morning, beginning shortly after sunrise. Cameras (TC395X; Burle Industries, Lancaster, PA) with TS6ZME-5 6.3-38 mm lenses (Cosmicar/Pentax, Tokyo, Japan) were positioned ~1.5 m from and focused on the bird feeder table. A time-lapse VCR (Panasonic, Tokyo, Japan) recorded bird feeding at a slow speed approximating 7–8 frames per second. The bird table filled the monitor screen, allowing clear vision of bird activity. Other bird species did not alight on the table. Blackbirds did appear twice in the silvereye experiment in the orchard and took grapes. Their choices were analyzed with the blackbird data. We considered the possibility that silvereye choices were then limited, but this supposition was discounted.

Artificial grapes of 5-6 mm diam were made from agar and gelatine and colored purple (Saxton et al. 2004a). Varying concentrations of hexose sugars (50%) D-glucose and 50% L-fructose) were added to the mixture. Experiments were run with grapes of sugar concentrations of 10, 15, 20, and 25% for blackbirds, and 5, 10, and 15% for silvereyes. Tartaric or malic acids were also added at 0, 10, 15, and 20 g/L. In earlier experiments blackbirds had exhibited preference for a higher sugar concentration (>20%) than silvereyes (10-15%) (Saxton et al. 2004c). The sugar concentrations chosen reflected these preferences to maximize the effect that sugar may have had on responses to acid in order to gain comparable results between species. The experiments were run as an exploration beginning with tartaric levels as high as in unripe grapes, and with malic acid levels at the same concentration, and in one experiment twice as high to elicit a response.

Video footage was viewed and details of bird behavior recorded. The number of bird visits to the bird table was recorded. For blackbirds, grape preference was recorded as to which grapes were eaten or taken away (accepted) or dropped after handling (rejected). Accepted grapes were analyzed as a percentage of total grapes handled. Grapes that were not handled were not included in the data set. For silvereyes the number of consecutive pecks at one grape was recorded as one bird visit, and data presented as mean pecks per visit to each type of grape. A silvereye would often then move to another grape, which was recorded as another bird visit. Data were not normally distributed so were analyzed by non-parametric Kruskall-Wallis ANOVA (H) and Mann-Whitney U test (U) using GenStat 6 software (Siegel and Castellan 1988).

Results

Blackbirds. At 10% hexose sugar concentration there was a significant preference (p = 0.05) for no acid in tartaric acid experiments (Table 1; H = 5.91), where 0 g/L was significantly preferred to 10 g/L and all other tartaric levels (U = 76.5), but there were no significant preferences shown for malic acid. At 15% sugar and tartaric acid there was no significant difference among treatments. A wider range of malic acid concentrations (0, 10, 20, and 40 g/L) were offered in grapes with 15% sugar, but still no significant differences found. At both 20% and 25% sugar there were no significant differences for both tartaric and malic acids. There was a significant preference (p = 0.05)

for high sugar over all acid treatments (Table 1; H = 12.4) with tartaric acid at 20% and malic acid at 25% preferred to 10% (U = 1153 and 1140, respectively). Tartaric acid at 20% and malic acid at 25% were also significantly preferred to 15% (U = 1136.5 and 1103, respectively).

Silvereyes. With silvereyes no significant differences were shown at any level of acid concentration, and no preferences were detectable. A significant preference among sugar levels was shown over all acid concentrations (Table 2; H = 30), with malic acid for 10% sugar preferred over 5% (U = 174.5) and with tartaric acid for 15% preferred over 5% (U=212.1). Between 10 and 15% sugar there was no significant preference.

Table 1 Blackbird response to tartaric and malic acid concentration in artificial grapes at varying sugar level	els:
mean percentage grapes taken (SE) for number of sessions (N) and number of bird visits (n). (Note: In a similar e	xperiment
with blackbirds with 40 g/L malic acid, no significant response of birds to the acid was detected.)	

	Blackbird response according to acid concentration [mean (SE)]								
	0 g/L	10 g/L	15 g/L	20 g/L	Total	Mean			
Tartaric acid									
10% sugar (N = 16, n = 63)	90.6 (3.6)**a	68.7 (7.9)	71.9 (7.3)	73.9 (7.6)	305.1	76.2			
15% sugar (N = 7, n = 45)	95.8 (4.7)	81.9 (8.7)	54.1 (13.5)	88.9 (11.1)	320.7	80.2			
20% sugar (N = 6, n = 26)	100.0 (0)	81.0 (10.3)	83.3 (7.45)	80.9 (14.3)	345.2	86.3*			
25% sugar (N = 6, n = 32)	78.3 (9.7)	55.0 (9.3)	70.0 (20.0)	100.0 (0.0)	303.3	75.8			
Malic acid									
10% sugar (N = 9, n = 46)	87.0 (5.2)	63.9 (11.9)	77.9 (11.3)	87.9 (4.8)	316.7	79.1			
15% sugar (N = 3, n = 20)	38.6 (19.8)	36.0 (21.7)		58.0 (12.7)	221.2	55.3			
20% sugar (N = 7, n = 61)	92.9 (4.6)	100.0 (0)	85.7 (6.7)	92.8 (4.6)	371.4	92.8			
25% sugar (N = 10, n = 7)	97.2 (2.7)	93.5 (4.34)	94.4 (4.6)	97.2 (2.7)	382.1	95.5*			
Totals over both acids	680.4	979.8	537.3	679.6					
Means over both acids	85.0	72.47	67.1	84.95					

^{a*} and ^{**} indicate significance at p = 0.05 for sugar concentration and at p = 0.05 for acid concentration, respectively.

 Table 2
 Silvereye response to tartaric and malic acid concentration in artificial grapes at varying sugar levels:

 mean pecks per visit (SE), for number of sessions (N) and number of bird visits (n).

	Silvereye response according to acid concentration [mean (SE)]								
	0 g/L	10 g/L	15 g/L	20 g/L	Total	Mean			
Tartaric acid									
5% sugar (N = 4, n = 57)	1.16 (0.27)	1.00 (0.32)	1.23 (0.29)	1.35 (0.28)	4.74	1.18			
10% sugar (N = 6, n = 97)	2.52 (0.52)	1.36 (0.46)	1.39 (0.55)	2.43 (0.50)	7.70	1.92			
15% sugar (N = 15, n = 88)	3.10 (0.63)	4.50 (1.84)	2.99 (0.51)	2.58 (0.41)	13.17	3.29*ª			
Malic acid									
5% sugar (N = 5, n = 37)	2.78 (0.69)	3.01 (0.85)	1.78 (0.44)	1.32 (0.29)	8.89	2.22			
10% sugar (N = 5, n = 12)	5.09 (1.17)	2.93 (0.15)	3.20 (0.98)	3.08 (0.24)	14.30	3.57*			
15% sugar (N = 6, n = 36)	3.25 (0.62)	2.32 (0.31)	1.83 (0.33)	2.72 (0.34)	10.12	2.53			
Totals over both acids	17.9	15.12	12.42	13.48					
Mean over both acids	2.98	2.52	2.03	2.24					

^{a*} indicates significance at p = 0.05 for sugar concentration.

Discussion

Results suggest that birds do not respond in any particular way to the levels of the acids used in this study (and found in natural grapes). The results did support the results from previous sugar experiments (Saxton et al. 2004c). The acid concentrations used were similar to those found in ripening grapes from the period of veraison onward (~10 g/L by harvest). The greatest bird damage to grapes occurs shortly before harvest (Boudreau 1972). Acid concentrations higher than 20 g/L are recorded in unripe preveraison grapes. Bird depredation at this stage is minimal.

The birds in this study appeared insensitive to acids, which concurs with Fuerst and Kare (1962, cited in Mason and Clark 2000), who noted that finches were tolerant of acidic and alkaline solutions and even preferred water with acid to tap water. This tolerance to acid may be due to lack of physiological mechanisms used to detect acids. For example, birds have fewer taste buds than mammals (King and McLelland 1984, Mason and Clark 2000). Blackbirds and silvereyes are sensitive to tannin concentration (V. Saxton, author's unpublished data), which has a bitter taste sensation, and to sugar (Saxton et al. 2004c), which has a sweet taste sensation, indicating that these birds can detect tastes. Taste perception is highly variable in humans (Bartoshuk et al. 2005), and although sour taste forms part of the avian taste spectrum, in one study it did not affect preference choices of starlings (Sturnus vulgaris) and red-winged blackbirds (Agelaius phoenicius) (Espaillat and Mason 1990). High acid in most fruit is repellent to mammals, but there is little evidence for this reaction for birds (King and McLelland 1984, Mason and Clark 2000). The highest concentration of acid in grapes is in the pulp (Ruffner 1982), which is the part that is most attractive to birds (Martinez Del Rio et al. 1992, Sallabanks 1993). Mason and Clark (2000) suggest that lack of sensitivity to acid is important to starlings, as juvenile starlings use unripe fruit as a food source, possibly because they are closed out of preferred food sources by more dominant adult birds (Feare 1984). Espaillat and Mason (1990) found that starlings and red-winged blackbirds were not averse to citric acid in combination with fructose and noted that most fruit pulp contains both. They also noted that tannic acid invoked an aversive response from both species, which concurs with our findings using tannins with European blackbirds and silvereyes (V. Saxton, author's unpublished data) where birds were sensitive to tannin. From the experiments reported here, the hypothesis that decreasing concentrations of acid increases grape attraction to birds or explains lesser bird pressure on some

cultivars such as Riesling (a high-acid and late-ripening grape variety) is not upheld. It is also not proven that sugar increases tolerance of birds to acids or that acid enhances grape attraction at lower levels of sugar.

Other cues such as color or aroma (Saxton et al. 2004b) may well override completely any deterrent that acid perception may offer to these bird species. High acid in fruit is often associated with green color, but green color is not a deterrent to birds in vineyards. The reason that cultivars such as Riesling are not attacked preferentially by birds is more likely to be lower sugar levels than other nearby grape varieties.

Conclusion

Results here cast considerable doubt on the theory that reducing acids in grapes renders them more attractive to birds. These results concur with anecdotal perceptions of blackbirds eating high-acid preveraison grapes. In this experiment sugar cues clearly overrode acid, although it remains unknown the degree to which these species can detect acid. Cues such as sugar, color, and aroma appear to override any effect that acid may have on avian perception of grapes. Decreasing acid levels are not important to birds, suggesting that vineyard managers can ignore acid level when evaluating which varieties will be under most pressure and at what stage of ripening.

Literature Cited

- Barbeau, G., S. Bournand, R. Champenois, M.H. Bouvet, A. Blin, and M. Cosneau. 2004. The behaviour of four red grapevine varieties of Val de Loire according to climatic variables. J. Int. Sci. Vigne Vin 38:35-40.
- Bartoshuk, L., D.J. Snyder, and V.B Duffy. 2005. The research challenge. *In* Proceedings of the Twelfth Australian Wine Industry Technical Conference, 24–29 July 2004. R. Blair et al. (eds.), pp. 20-24. AWITC, Glen Osmond.
- Beriashvili, T.V., and L.T. Beriashvili. 1996. Metabolism of malic and tartaric acids in grape berries. Biochemistry 61:1316-1321.
- Boudreau, G.W. 1972. Factors related to bird depredations in vineyards. Am. J. Enol. Vitic. 23:50-53.
- Coombe, B.G. 1992. Research on development and ripening of the grape berry. Am. J. Enol. Vitic. 43:101-110.
- Doneche, B., F. Roux, and P. Ribereau-Gayon. 1985. Degradation de l'acide malique par *Botrytis cinerea*. Can. J. Bot. 63:1820-1824.
- Espaillat, J.E., and J.R. Mason. 1990. Differences in taste preference between red-winged blackbirds and European starlings. Wilson Bull. 102:292-299.
- Feare, C. 1984. The Starling. Oxford University Press, Oxford, UK.
- Hunter, J.J., J.H. Visser, and O.T. De Villiers. 1991. Preparation of grapes and extraction of sugars and organic acids for determination by high performance liquid chromatography. Am. J. Enol. Vitic. 42:237-244.
- King, A.S., and J. McLelland. 1984. Birds, Their Structure and Function. Bailliere Tindall, London.
- Lavee, S., and G. Nir. 1986. Grape. *In* CRC Handbook of Fruitset and Development. S. Monselire (ed.), pp. 167-191. CRC Press, Boca Raton, FL.

- Martinez Del Rio, C., H.G. Baker, and I. Baker. 1992. Ecological and evolutionary implications of digestive processes: Bird preferences and the sugar constituents of floral nectar and fruit pulp. Experientia 48:544-550.
- Mason, J.R., and L. Clark. 2000. The chemical senses in birds. *In* Sturkie's Avian Physiology. 5th ed. G.C. Whittow (ed.), pp. 39-56. Academic Press, San Diego.
- Robredo, L.M., B. Junquera, M.L. Gonzales-Sanjose, and L.J.R. Barron. 1991. Biochemical events during ripening of grape berries. Ital. J. Food Sci. 3:173-180.
- Ruffner, H.P. 1982. Metabolism of tartaric and malic acids in *Vitis*: A review. Vitis 21:247-259, 346-358.
- Sallabanks, R. 1993. Hierarchical mechanisms of fruit selection by an avian frugivore. Ecology 74:1326-1336.
- Saxton, V.P. 2004. Influence of grape ripening compounds on behavioural responses of birds. Thesis, Lincoln University, Christchurch, New Zealand.

- Saxton, V.P., G.L. Creasy, A.M. Paterson, and M.C.T. Trought. 2004a. Experimental method to investigate and monitor bird behavior and damage in vineyards. Am. J. Enol. Vitic. 55:288-291.
- Saxton, V.P., G.L. Creasy, A.M. Paterson, and M.C.T. Trought. 2004b. Response of blackbirds (*Turdus merula*) and silvereyes (*Zosterops lateralis*) to geraniol and 2-methoxy-3-isobutylpyrazine. Am. J. Enol. Vitic. 55:292-294.
- Saxton, V.P., G.J. Hickling, M.C.T. Trought, and G.L. Creasy. 2004c. Comparative behavior of free-ranging blackbirds (*Turdus merula*) and silvereyes (*Zosterops lateralis*) with hexose sugars in artificial grapes. Appl. Animal Behav. Sci. 85:157-166.
- Siegel, S., and N.J. Castellan. 1988. Nonparametric Statistics for the Behavioral Sciences. McGraw-Hill, NY.
- Terrier, N., F.X. Sauvage, A. Ageorges, and C. Romieu. 2001. Changes in acidity and in proton transport at the tonoplast of grape berries during development. Planta 213:20-28.