

**Influence of ripening grape compounds on  
behavioural responses of birds**

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submitted in partial fulfilment

of the requirements for the Degree of

Doctor of Philosophy

By

V.P.Saxton

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## **Influence of ripening grape compounds on behavioural responses of birds**

**By V.P.Saxton**

**Keywords: vineyards, bird damage, artificial grape, blackbirds, silvereyes, behaviour, hexose sugar, aroma, acid, colour, tannin, bird management**

Vineyards in New Zealand suffer bird damage caused by several avian species, including blackbirds and silvereyes. The introduced European blackbird takes whole grapes which reduces yield. The self-introduced Australasian silvereye pecks on grapes, leaving them on the vine to be further attacked by fungi and bacteria, and the subsequent off-odours can cause grapes to be refused by the winery or to suffer a price-reduction. Bird control methods remain primitive and largely ineffective during the long ripening period of wine grapes. An ecologically sound method to manage and reduce bird pressure requires deeper understanding of why some birds eat grapes, especially since grapes are not particularly nutritious.

This work investigated the extent to which blackbirds and silvereyes are attracted by various compounds in ripening grapes. Since in natural grapes these compounds develop and change simultaneously, I developed an artificial grape in which a single parameter could be investigated. Artificial grapes (and sometimes nectar) were presented on a bird feeder table and the responses of birds to hexose sugars, the aromas 2-3-isobutylmethoxypyrazine and geraniol, tartaric and malic acids, grape tannins, and purple and green colour were recorded on timelapse video and analysed.

Blackbirds preferred high concentrations of hexose sugar, were responsive to the aroma of ripe grapes, not deterred by high acid concentrations, sensitive to grape tannin concentrations lower than occur in grape seeds, and showed no preference for purple or green in summer, but in winter took only purple grapes. Silvereyes preferred a hexose sugar concentration between 10% and 15%, did not respond to the aroma of ripe grapes, showed no significant distaste for acid, were not so sensitive to tannins, showed no colour preference in summer but in winter showed a significant preference for green grapes.

The responses of the two species varied considerably, possibly reflecting physiological differences that would explain the behaviours. Blackbirds have an inefficient glucose assimilation digestive system that requires regular foraging to replenish the gut. They remove whole grapes at regular intervals and take time to digest them. This enables them to watch for predators, as they are solitary territorial birds. Silvereyes may have a passive glucose absorption digestive system, which is up to 95% efficient, and therefore assimilation rate controls the feeding rate. This fits in with the observed preference for lower sugar concentration and the constant pecking. Because they must consume sugar constantly and slowly they have no time to watch for predators and need the protection of a flock. Both species attack both colour grapes before harvest, and colour preferences may be subject to other factors such as sugar concentration or availability. The winter preference of blackbirds for purple may reflect a metabolic process similar to that of migratory birds to gain fat for winter. Silvereyes preference for green may be to avoid the toxicity of purple-black fruits to which they may be vulnerable through their passive system of assimilation.

The differences between the two species suggest that bird management practices should be species-specific, and that an ecological solution requires such understanding of why the birds eat grapes.

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## Preface

This project was conceived by Drs. Mike Trought and Graham Hickling to investigate what in grapes was more important to birds – sugar or colour. Some previous surveys (Watkins, 1999; Davies 2000) used timelapse video methodology to view bird behaviour. Avery's (1995) experimental method using artificial food was the springboard for the idea of an artificial grape bunch where sugar, colour and other grape compounds could be manipulated while all others remained controlled.

At the beginning of this project there were many uncertainties about whether birds would visit or take artificial grapes at all. Development of the grape itself took several months of experimentation, and a reliable repeatable recipe was a matter of trial and error for many more months, with many intended grapes being thrown out (mostly because they did not set well). The order of adding ingredients and the temperature at which gelatine was added proved critical (Chapter 3).

The original intention had been to hang a 'surrogate bunch' in the canopy. Creating a bunch proved too difficult so it became necessary to present the grapes to birds in some other way. In addition bringing the bird to where it could be caught on video in a large grape canopy proved problematic. After some video sessions where grapes had been placed on a trellis endstay, the bird feeder table was developed to make site choice more flexible, and the double deck was developed to get more data from each video session (see Chapter 3). Operation of the video equipment required patience and involved many unsuccessful sessions. Difficulties included electronic failure (batteries or fuses failing, inverters breaking), weather conditions (misting of lens, sunstrike, rain, camera or table

blowing over), no bird visits (many sites produced no data), human error (forgetting to put in a tape or to turn everything on, failing to focus the lens). Development of the experimental method took some eighteen months.

Thanks to my external adviser Dr Mike Trought of the Wine Research Centre in Marlborough, who kept the project firmly on the applied track and assisted with contacts for the Marlborough survey (Saxton, 2002). Very sincere thanks go to my supervisors Dr Glen Creasy and Dr Adrian Paterson, who were diligent and responsive, encouraging and supportive at all times. Thanks also to Dr Jim Coleman of Landcare Research who was interested and commented in detail on all the manuscripts. I have learned much from all these experts in areas of perspective, writing skills, communication and application. Their expertise and willingness to share and mentor are gratefully recognised.

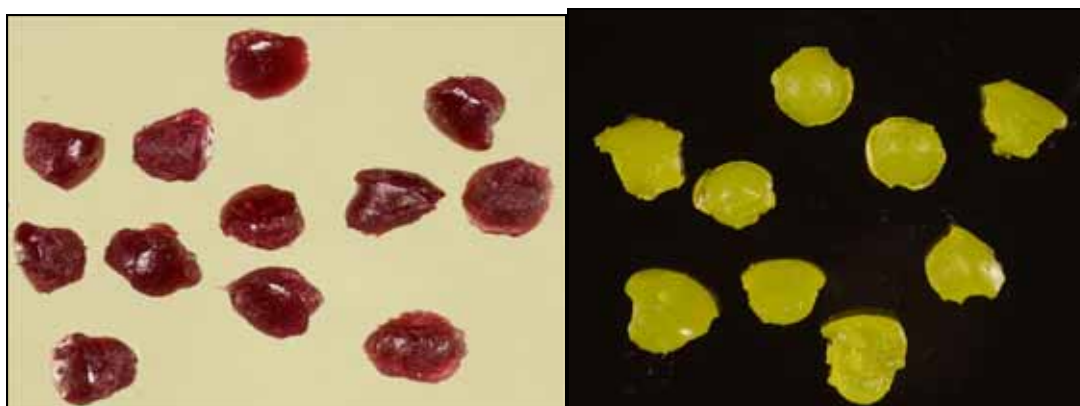
Sincere thanks for technical support go to Lewis Jennings, who mended everything and solved all problems, gave sage advice and instructions on the operation of and technical limitations of the equipment. Heartfelt thanks go also to a colleague, Dr Wendy Parr, who befriended, inspired and encouraged me, and to my two children, James and Ellie, for real moral support, albeit of a casual nature.

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Damage to Shiraz and to Chardonnay grapes in Marlborough, 2002



Artificial grapes made from agar and gelatine, piped into a mould 11mm in diameter.

# Chapter one

## General introduction

### 1.1 The problem

The New Zealand winegrowing industry suffers problems of bird damage in vineyards. This is not a uniquely New Zealand problem (Nelson, 1990, 1991; Coleman 2001), nor is it confined to winegrapes. Some parts of the world experience severe pressure, especially at locations that are on starling migratory routes, and in some countries growers receive compensation for damage sustained (Coleman, 2001). Damage is suffered by many horticultural and agricultural crops in New Zealand (Porter *et al.*, 1994). With economic control of pathogens and insects available to growers, birds now rank as a primary cause of loss and damage to grapes in New Zealand (Porter, 1992; Boyce *et al.*, 1999; Fukuda, 1999; Watkins, 1999).

New Zealand's temperate climate results in a long two month ripening season between colour change (or véraison) and harvest, during which time grapes are subject to attack from birds. There are many vineyard areas of less than 10Ha, small on an international scale, often surrounded by trees or other crops. The adaptation of introduced birds alongside horticultural developments has resulted in avian species that have established in New Zealand almost exclusively in conjunction with crops. Intensification of cropping has enabled populations of these birds to increase, while milder winters have enabled the survival of greater numbers of birds than in their original habitat where more severe winters are a natural population control. All are elements that compound the problem of bird damage to grapes.

Economically the small croploads necessary to achieve quality in a marginal temperate climate, lack of cheap labour and high costs of export all contribute to the need to maximise profits from New Zealand grapes. Losses of 10-15% in grape yield that are commonly reported represent substantial economic loss when extrapolated to wine lost from high quality production.

Many avian species are involved in crop damage generally. Some species damage only specific crops, and some are general foragers (O'Connor and Shrubbs, 1986; Boutin *et al.*, 1999). There is no universal control method for birds (Nelson, 1991; Hickling, 1995), and it seems likely that each pest species will have to be controlled independently, at least in the short term. Research into bird damage to crops appears to be unintegrated and sporadic, due perhaps to the inconsistent levels and locations of damage sustained from year to year (Tracey and Saunders, 2001). Annual fluctuations of bird populations (Somers and Morris, 2002), of bird behaviour, of weather conditions (Saxton, 2002), or perhaps just grower perceptions (Saxton, unpublished data), has led to an uneven level of urgency, especially if growers are compensated for losses sustained (as in Germany, Coleman, 2001). Lack of data on economic losses (Boyce *et al.*, 1999; Sinclair, 2000, 2001; Coleman, 2001; Tracey and Saunders 2001) means that the overarching agencies such as governments consider bird damage to be an industry problem that should be solved at industry level with industry funds. The difficulty with this assessment is that, contrary to many areas where industry does fund product development, in the area of bird behaviour the underlying research has not been done, so that the principles needed to define and underpin solutions are not in place. One serious difficulty is that birds represent a highly mobile and intermittent population of research subjects (Burton, 1996) and consistency is needed for experimental research. This causes problems with control of variables, which leads to lack of robust analyses.



Bird damage to crops, though generally inconsistent, can be heavy and consistent (Jensen, 1974; Taber, 2002; Saltzl, J. pers. comm. 2003), and in New Zealand there are many smaller (less than 10Ha) vineyards where unprotected grapes can be totally devastated by birds (Porter and McLennan, 1988). Damage appears to be greater in smaller vineyards (Somers and Morris, 2002), while larger vineyards (more than 10Ha.), or vineyards surrounded by other vineyards, tend not to sustain high levels of bird-related damage, at least in the interior of the crop (Tracey and Saunders, 2001; Somers and Morris 2002). There has been little research beyond the evaluation of some crop protection methods (hawk kites, eye-spot balloons, mirrors, acoustic alarm calls etc.), although those tested scientifically have been found to be largely ineffective (Hickling, 1995; Fukuda, 1999; Tracey and Saunders, 2001; Saxton, unpublished).

## **1.2 The ripening grape**

The New Zealand Wine industry is based on cultivars to make single varietal wines. Most of the cultivars are typical of northern European regions, the most planted being Sauvignon Blanc (Loire and Bordeaux), Chardonnay (Champagne and Burgundy), and Riesling (Alsace, Mosel, Saar), to make white wines, and Pinot Noir (Burgundy) Merlot and Cabernet Sauvignon (Bordeaux) for red wines (Jackson and Schuster, 1994). There are lesser plantings of many other varieties such as Gewürztraminer, Müller-Thurgau, Pinot Gris and Pinot Blanc, Chenin Blanc, Semillon, Reichensteiner, Pinotage, Malbec, Syrah, and Cabernet Franc (NZ Winegrowers Annual Report, 2003).

Seeds in the grapes are mature when the grape reaches *véraison*, or colour change. At this stage berries of 'red' varieties turn from green to red and 'white' varieties become a translucent green. In all varieties the berries soften suddenly some six days before enlarging

in volume (depending on water availability) and the skin becomes noticeably waxy (Coombe, 1992). These changes vary slightly from cluster to cluster and from berry to berry, due to the differing dates of anthesis (fruitset), as each berry is independent of its neighbours (Lang and Caspari 1999). Hexose sugars (mainly glucose and fructose) concentrate in the berry. The two main organic acids, tartaric and malic, decrease (Rebucci *et al.*, 1997). Secondary metabolites (also known as phenolic compounds) also increase in concentration (Coombe, 1992; Davies *et al.*, 1996), and the grapes become more aromatic.

Varieties vary in their speed and time of ripening, but most of the above were planted because they will ripen, albeit slowly, in the maritime climate of New Zealand, which is buffered by the Pacific Ocean. Most of the cultivars experience early budburst and ripening. Ripening is slow compared to Australian regions or to continental European climates, but the length of the New Zealand season compensates. As a consequence, however, the grapes are a long time ripening on the vine, up to 10 weeks from véraison to harvest, and a long time exposed to potential bird depredation.

### **1.3 Birds in vineyards**

There are probably many factors that contribute to an individual bird's decision to attack a grape. These may be broadly divided into endogenous motivation (such as hunger), physiological or nutritional needs, cultural behaviour (such as copying other birds), exogenous factors such as availability and abundance of grapes at the time, and environmental factors that account for the bird's presence in the vineyard at all. The nutritive value of grapes to birds is low, for instance Herrera (1981) summarises overall profitability of grapes (*Vitis vinifera silvestris*) at 0.9, whereas wild olives (*Olea europaea var. silvestris*) is 7.3 and the pistachio tree (*Pistacia lentiscus*) is 18.2 (in a formula based on the value of dry weight) .

### **1.3.1 Bird pressure**

Depredation pressure on a crop will depend to some extent on the size of the bird population relative to the area of grapes, i.e. birds per hectare. It is difficult to estimate small bird numbers in any situation. Three banding programs that were run with silvereyes (*Zosterops lateralis*) in Canterbury and Wellington returned few birds probably due to individual death, relocation of flocks, possibly invisibility of such small birds to the public, and the data remain unpublished (Reese, P. pers. comm. 2004). A Department of Conservation project aiming to trap the estimated one hundred magpies in the Te Matai treatment block in the Bay of Plenty was still trapping 1800 birds later (Waikato University Magpie Project, December 2002 Magpie Newsletter), and these are relatively large, conspicuous, non-flocking birds. It is difficult to reduce bird populations due to reinvasion, as birds are highly mobile, and even a robust assessment may not be valid for long. Thus, rather than attempting a population survey, the most practical way to measure bird pressure on vineyards may be to assess damage done, as an indirect measure of bird pressure. As the type of damage may also indicate bird species, some information on populations of the specific species involved could be gathered in this way.

### **1.3.2 Environmental factors**

Environmental characteristics such as type of cover, density, height, and proximity to vines, are possibly significant to foraging birds. In the predator-prey, risk-reward models of bird behaviour (Carpenter, 1987; Stephens, 1990; Krebs and Kacelnik, 1991; Maurer, 1996, Abreu and Kacelnik, 1999), cover is an essential element that contributes to risk minimisation. Different bird species adopt different types of cover (Krebs and Kacelnik, 1991; Isenmann and Debout, 2000; Somers and Morris, 2002), and the type preferred may be linked to their flight characteristics. For example blackbirds (*Turdus merula*), which are often seen on the

ground, use low cover such as matagouri and dense bushes (Jensen, 1974; Porter *et al.*, 1994; King, G. pers. comm., 2002). Silvereyes, which forage higher in the grape canopy (Saxton, 2002) and are seldom seen near the ground, prefer willows and similar mid-height leafy trees (Stanley and Lill, 2002). Starlings are often seen perching on power lines, or in tall open canopies such as gum trees (Porter *et al.*, 1994). The presence of these cover characteristics is probably significant in an individual bird's choice to stay at the foraging site (Somers and Morris, 2002; Taber, 2002). Environmental factors are likely to have overriding importance to birds. A survey conducted for NZ Winegrowers of vineyard surroundings that correlate with increased damage suggested several factors of importance. Rated from most likely to increase damage to least likely: open land, fruit orchards, roads, trees, cropping farms, buildings, streams, power lines and more vineyards next door (Saxton, unpublished data). This is a large topic and could not be addressed in this project.

### **1.3.3 Exogenous motivators.**

Bird species that attack grapes in New Zealand fall into three main groupings: residential species, itinerant flocks and migrating flocks. European blackbirds and thrushes (*Turdus philomelos*) are resident birds, and are territorial (but probably only to their con-specifics, Orians and Willson, 1964; Ford, 1989; McKean 1990). They defend their territory at nesting time but probably not at times of grape ripening since they gain nothing from defending a food resource that is so abundant (Carpenter, 1987). Both species are ground hoppers, and for most of the year eat insects and worms, which they find on the ground. They will eat grapes when they are ripe and available, but whether it is grapes that bring the birds to the vineyard is not known. Simple availability of grapes may be the reason these birds eat them, although it is possible that this patch has been selected by the bird because grapes are available at a certain season that complements the bird's endogenous nutritional needs (Bairlein, 2003).

Silvereyes and the common myna (*Acridotheris tristis*) frequent vineyards in small flocks (five to fifteen birds, Sinclair and Porter, 1994). They are present in some vineyards while grapes are ripe, but not at other times. Silvereyes may nest in the vicinity, but it is noted that they often arrive from elsewhere (Sinclair and Porter, 1994), anecdotally from the hills after the first frost. In New Zealand silvereye populations are frequently itinerant (Reese, P. pers. comm., 2004). Although their movements are seasonal, they are probably governed more by environmental factors such as drought, cold, and food availability.

Starlings are the last of the five main bird pests in New Zealand vineyards. These birds do fly long distances, and are known to migrate in both Europe, where they migrate from Siberia to Africa, and in North America, where they migrate from Canada to Mexico. They stop off at sites, including vineyards, en route, which are then heavily damaged as thousands of birds refuel (Somers and Morris, 2002; Taber, 2002, Salzl, J., pers. comm., 2003). Starlings in New Zealand do not obviously migrate, but do appear to roost in a particularly suitable group of trees, and then maraud looking for food during the day. If grapes are available in the vicinity then these will be subject to attack.

For birds that target grapes, although abundance itself may be a significant cue, there are probably intrinsic aspects of grapes that attract these birds. There are many species of birds that turn from an insect or arthropod dominated to fruit -in autumn (Afik and Karasov, 1995). It has been assumed that this change is prompted by the seasonal disappearance of caterpillars (Herrera, 1982). Recent research, however, has shown that for some bird species, a diet of fruit alone can reduce metabolic rate, and then stimulate over-eating (hyperphagia) with the aim of increasing fat deposits very rapidly in preparation for migration (Bairlein and Totzke,

1992; Bairlein, 2002). Birds that have to survive a long cold winter exhibit a similar process (Totzke *et al.*, 2000).

#### **1.3.4 Nutritional factors**

Nutritional needs of birds centre on carbohydrates for energy, protein for growth of feathers, and for amino acids and enzymes to drive their digestion, calcium for egg making, lipids for fat, and water (Herrera, 1982). At all times they must maintain a balance between energy expended searching for food, and the gain from that food (Ricklefs, 1996; Bautista *et al.*, 1998). Energy budgets have been modelled for several species (Moermond and Denslow, 1983; Ford *et al.*, 1990; Abreu and Kacelnik, 1999). Risk-reward and predator-prey models are a more sophisticated version where risk of predation is built into the energy model. With this risk built in, it is often more worth-while for a bird to forego a high energy reward in favour of a lesser reward that is more safely and more frequently gained (Herrera, 1982; Morse, 1990; Giles and Lill, 1999). This is probably very relevant to the vineyard situation, because it is known that grapes are a low reward for birds (Herrera, 1981).

### **1.4. Bird-fruit interactions.**

Analysis of bird motivation when choosing a grape suggests that endogenous, environmental and cultural factors, ontogenetic learning and individual choice all contribute to decision-making processes. A brief summary might read as follows: perception, recognition, sampling, repetition. Intermediary processes contribute by stimulating or rewarding the behaviour.

#### **1.4.1 Stages of decision-making for an individual:**

1. Preceding the initial perception of fruit by the bird, endogenous motivation such as hunger, energy to search, and development of a search image for cryptically displayed fruit would be

present. Cues available to the bird at this stage include conspicuousness, colour, and aroma, which stimulate perception.

2. Perception, recognition as a food source from prior feeding, and/or genetic recognition of shape or colour. Cues here may include size, colour, shape, abundance, position and other display characteristics.

3. Recognition and sampling may influence further behaviour: habit, culture, imitation, genetic programming, and memory.

4. With sampling and repetition come reward factors, digestive, nutritive, immediate, ongoing or delayed, and cues to the bird may include size and positioning of the fruit, nutritive value as perceived by the bird's senses or delayed feedback from the gut.

5. The final stage of repetitive continued feeding probably follows feedback from the bird's brain and body regarding the satisfaction of needs and accompanying this feedback will be ontogenetic learning. All this occurs within a framework of environmental constraints such as predator risk assessment, type of cover, and social needs of the individual bird.

Factors that contribute to the size of the foraging population would thus begin with environmental factors, cover, food resources, nesting opportunities and territory size, predator presence, and intra- and inter-specific competition for resources. When these needs are met then the attractiveness of the crop to the particular bird species will be the next factor. Given that the above process is satisfactorily resolved in nutritive reward, the behaviour is reinforced and the birds remain.

The research presented here compares two species that are common vineyard pests, blackbirds and silvereyes, and attempts to discover from their foraging behaviour the relative importance of grape ripening characteristics as cues to each species. Endogenous, exogenous and

environmental factors that also contribute in the decision-making process of grape foraging are discussed in relation to the meaning of the cues that grapes appear to give the birds.

## **1.5 Research to date**

Investigating influences of fruit on the frugivory of birds is a wide brief. There are several areas of research that are divided approximately into ultimate (why) and proximate (how) questions. Co-evolutionary aspects of birds and fruit (e.g. Reid, 1991), the ontogenetic learning of individuals (McKean, 1990; Sherry, 1990), and studies of the adaptiveness of behaviour (defined as ethology) are based on comparative behavioural experiments with individual captive birds and address the ultimate questions. Proximate questions of how morphological or physiological constraints (such as digestive capacity) influence foraging behaviour, and the function of frugivory in bird foraging (Afik and Karasov, 1995) is a third field, while the incorporation of environmental factors has led to the development of models to describe bird behaviour, such as the predator-prey, risk-reward, and similar models, that analyse and balance the bird's priorities in foraging. Questions addressed include: how important is the initial stimulus of availability and attractiveness of the fruit, and the reward after eating it, in basic stimulus-response-reward behavioural conditioning process (Herrera, 1981, 1982; Moermond and Denslow 1983; Willson, 1986; Tiebout, 1991; Martinez Del Rio *et al.*, 1992; Stiles, 1993; Puckey *et al.*, 1996; Stanley and Lill, 2001)? How do environmental factors such as cover, nesting sites, presence of other birds or predators, contribute to the overall picture? (Hinde, 1982; Willson, 1986; Ford *et al.*, 1990; Hutto, 1990; Maurer, 1996; Morse, 1990; Stephens, 1990; Krebs and Kacelnik, 1991; Murphy, 1994)? The approach of all these authors is highly theoretical, testing basic concepts rigorously using a few individual birds, usually in captivity. The application of basic conceptual findings to field conditions requires an understanding of the interaction of bird behaviour with the environment.



Correlation of environmental factors with bird behaviour, of which frugivory is a part, forms a large body of data. This more applied research field covers topics such as avian colour choices, size and position of fruit, seed content, and other factors that impact on individual bird-fruit interactions (Gaze and Clout, 1983; Moermond and Denslow, 1983; Gautier-Hion *et al.*, 1985; Wheelwright, 1985; Davidar and Morton, 1986; Jordano, 1987; Hutto, 1990; Place and Stiles, 1992; Sallabanks, 1993; Stiles, 1993; Fuentes, 1994,1995; Murphy, 1994; Williams and Karl, 1996; Bautista *et al.*, 1998; Giles and Lill, 1999; Stanley and Lill, 2001). A wide range of ecological factors are involved. The relevance of co-evolutionary patterns between plants and seed dispersing bird species underpins this research, and may have relevance to some of the bird-fruit interactions investigated in the research reported here. There is a large body of data from surveys of what is happening, and some basic concepts about motivation have been developed. But bird species differ in many aspects of frugivory (Fuentes, 1994) and within the same species individuals differ in foraging choices depending on age, dominance, sex and seasonal requirements (Morse, 1990). It is accepted that much research may apply only to the species investigated.

There are very few data on vineyard birds, and the reasons why vineyard bird populations suddenly increase, some would say explode, over the two months of ripening, remain unclear. This research addresses the questions of what aspects of grapes attract birds, and the experimental results are discussed in the framework of cues and rewards to the birds that draw largely on the literature cited above. Further gaps in our knowledge, but not addressed here, include the perception of risk (many vineyard managers shoot birds) and the balancing of energy budgets, as many grape harvests occur as winter approaches.

## **1.6 Watkins' experimental work**

The research presented in this thesis grew out of a project that showed that silvereyes were attracted first to purple grapes until they learned that neighbouring green grapes were of higher degrees Brix (sugar concentration), and that blackbirds seemed to prefer purple grapes (Watkins, 1999). Proceeding further with natural grapes proved difficult, as it was not possible to control the confounding parameters of sugar concentrations, colour, aroma and acid levels in individual grapes. This led to the creation of an artificial grape. Many of the experiments presented here use the artificial grape in various configurations, and some use artificial nectar. Field surveys and some further environmental experiments with novel food sources completed the experimental work.

## **1.7 Where from and where to from here?**

### **1.7.1 Origin of the problem**

There is a close link between the diversification of landscape (or the reverse) and the bird populations (Gates and Donald, 2000; Munck, 2003). The close link between horticulture and the advent and increase of the bird damage problem in New Zealand is often not well appreciated. Apart from landscape diversification and increase in horticultural crops which favour avian species that use these crops as a food source, the damaging bird species have deliberately been introduced to New Zealand in tandem with farming and horticultural activity, often as control methods for other biological problems. Starlings were introduced as a natural predator of grassgrubs, blackbirds and thrushes for slugs and snails, and mynas to reduce insect pests. Science has provided chemical controls for many of these invertebrate pests, so that the presence of exotic bird species for these purposes is now somewhat historical. However, as pesticides become less acceptable and many become less effective, due to overuse inducing resistance in the target species or toxicity in the soil, using birds as a

biocontrol agent of insects may well be reconsidered. Meanwhile, due to their boldness and high adaptability, compared to native birds (Greenberg, 1990; Huber *et al.*, 2001), introduced species populations have increased in New Zealand, and they have become adept at foraging in farmland. Given the intensification of horticulture that has taken place over recent years, combined with a move to higher value crops and more stringent control of economic budgets, losses due to bird damage are both more extensive and less tolerated than in the past.

### **1.7.2 Bird control methods**

The demand for effective bird control has increased, but the effectiveness of most of the existing methods used to protect grape crops, apart from exclusion netting, is seriously questioned. Shooting together with gasguns is the most widely used method, with netting the second most widely used (Boyce *et al.*, 1999). Both of these methods are costly. There is an increase in humidity within the net, which may result in higher pathogen pressure, and, due to inhibition of photosynthesis, the accumulation of photosynthates in the grape berry can be reduced by up to 10% (Trought *et al.*, 1997; Creasy and Trought, 2001). Many high value vineyards elect not to net, preferring to sustain losses rather than reduce the quality of the grapes (Wood, G. pers. comm. 2001).

An acoustic soundnet system developed in Australia and aggressively marketed over the last few years in both Australia and New Zealand is unpopular with neighbouring residents, as are gasguns, and local byelaws may limit the allowed use of acoustically based methods. Some ancillary scaring devices such as eyespot balloons, glitter tapes, hawk kites and mirror devices are often used along with one or more of the above three main control methods.

Growers ignore birds for most of the year. An educational program supported by NZ Winegrowers (Saxton, 2003) has drawn attention to options for bird control outside the grape

ripening season, such as in late winter to reduce springtime numbers, or at nesting time for blackbirds and thrushes, with the aim of reducing pressure later in that season. However, the need for an ecologically based strategy is clear. Little is known about what attracts itinerant silvereyes, starlings and mynas to vineyards, or what makes grapes a preferred food source for the five species involved.

## **1.8 Research Aim**

The aim of the research presented here is to discover which grape ripening parameters might be significant in influencing bird foraging on grapes. This work was experimental and academic, but conducted in the framework of a need to understand the motivations of birds attacking grapes. Many other factors are undoubtedly involved and these are discussed in the conclusion. The main body of the research reported here focussed on the experimental testing.

### **1.8.1 Questions addressed**

Each experiment addressed one of five major grape-ripening parameters (colour, sugar, acid, tannin and aroma) and tested the responses of blackbirds and silvereyes to the hypotheses that there would be no significant responses. Multichoice tests (except for colour which was a binary choice test) presented the birds with four concentrations of the selected compound in an attempt to discover not only whether the compound was significant to the birds, but at what threshold level their responses were statistically significant.

### **1.8.2 Compounds**

While there are aspects of grapes such as size, shape, availability, abundance, position on the vine, translucency, UV reflection and proximity to bird cover (to mention a few), this researched focussed on the major contents of the grape itself which, besides water, are hexose

sugars, organic acids, secondary metabolites including anthocyanins that give colour and tannins, and aroma. Artificial grapes or nectar were used as a controlled food source. The manipulated compounds were:

1. 50% each glucose/fructose in 0%, 5%, 10%, 20% and 40% concentrations
2. geraniol and 2-3isobutylmethoxypyrazine aromas
3. tartaric and malic acids in 0%, 5%, 15% and 20% concentrations
4. grape tannins in 0%, 2.5%, 5%,7.5% and 10% concentrations
5. purple and green colour

### **1.8.3 Method**

Bird activities with the grapes and nectar were videoed and tapes viewed to analyse bird responses. An ethogram was developed for each species separately, as they displayed different behavioural characteristics, which were quantified for statistical analysis.

### **1.8.4 Bird species**

Initial experiments focussed on the birds in the Lincoln University Vineyard, which happened to be blackbirds. The experiments were tried at a different location with silvereyes, and the very different behaviour of the two species enabled a comparative analysis of two of the most common vineyard pest species. There were no starlings, mynas or thrushes at the locations of the experiments, though these species are responsible for damage to many vineyards.

### **1.8.5 Location**

Initial experiments were at several locations in the Lincoln University Vineyard, where blackbirds were present. A second location with several sites was established in a pear orchard 4km from the university, where silvereyes were present throughout the year.

Silvereye experiments were conducted in the pear orchard because there was no population of silvereyes in the vineyard for most of the year. These birds are often itinerant, and move in small flocks. The population in the pear orchard appeared resident at least for the duration of the experiments. Whether results were impacted by the fact that this is not typical behaviour of vineyard populations was questioned and the results appear in Appendix I. Vineyard populations appear to come to specific vineyards annually, about which they may have some prior knowledge or else there is something about grapes or the vineyard that cues them. The orchard birds appeared to be present in the pear orchard throughout the three-year period of the experiments, so they were unlikely to have been influenced by previous learning about grapes. Their behaviour with artificial grapes could be taken to be inherent and genetically programmed, and the results likely to be relevant.

#### **1.8.6 Design of experimental timeline**

Because the birds were present all year round experiments took place in all seasons. Midwinter was difficult due to fogging of lenses and unreliability of recording equipment in cold temperatures (<5°C) so little was done in midwinter. Most of the experiments were conducted in summer. The experiments began in May 2000 and finished in November 2003.

#### **1.9 Structure of the thesis**

The structure of the thesis is as follows:

**Chapter one:** Introduction, research and questions.

**Chapter two:** Bird control methods and limitations.

**Chapter three:** An experimental method to investigate and monitor bird behaviour and damage in vineyards. This chapter has been published in the American Journal of Enology

and Viticulture, 55:288-292. The investigation was: how valid are the results from an experimental investigative method with free- ranging birds in a real vineyard situation?

**Chapter four:** Comparative behaviour of free-ranging blackbirds (*Turdus merula*) and silvereyes (*Zosterops lateralis*) with hexose sugars in artificial grapes. This chapter has been published in Applied Animal Behaviour Science 85:157-166. The investigation was: which concentrations of hexose sugars were preferred by blackbirds and silvereyes?

**Chapter five:** Response of blackbirds and silvereyes to some grape aromas. This chapter has been published in the American Journal of Enology and Viticulture 55: 292-295. The investigation was on preferences of blackbirds and silvereyes for the grape aromas geraniol and 2-3-isobutylmethoxypyrazine.

**Chapter six:** Behavioural responses of two species of birds to varying levels of tartaric and malic acids in artificial grapes. This investigation attempted to determine the importance and threshold levels to birds of the two major organic acids in grapes.

**Chapter seven:** Comparative behavioural responses of two species of birds to colour and other secondary metabolites in grapes (*Vitis vinifera*). This chapter reports an investigation into the secondary metabolites of grapes by testing colour preferences and tannin threshold levels of the two bird species.

**Chapter eight:** Conclusions and general discussion.

**Epilogue:** Suggestions for further research

**Appendix I:** Comparison of a vineyard silvereye population with that in a nearby orchard.

**Appendix II:** QuikCARD survey card for grapegrowers.

This research was conducted in response to an industry problem, and the approach has been broad rather than an in-depth analysis of a more limited topic, as there was very little prior knowledge available on which to base experiments. Many of the concepts in the discussions

have been researched on other bird species, the aim was to deduce from observed behaviour of blackbirds and silvereyes whether these concepts might apply to them. Three of the chapters have been accepted for publication (Chapters 3, 4 and 5). Complementary work: A review of bird control methods (Watkins *et al.*, 2000), surveys of bird behaviour in Marlborough vineyards in 2000 and 2002 (Davies *et al.*, 2000; Saxton, 2002), and an investigation into avian populations of native bush remnants near vineyards in Marlborough (McEntee *et al.*, 2004) have all been published separately and are not included in the work presented here, though they are cited in the text. A grapegrowers survey of environmental correlates with bird damage (QuikCARD) which was presented at a workshop at the Romeo Bragato National Grapegrowers Conference, Wellington, 2003, remains unpublished.

I am responsible for the design and analysis of the experiments (analysis conducted with assistance from statistician Dr. Andrew McLachlan). As a viticulturist, and the problem addressed is that of grape deredation, the natural beginning was the grape itself. In the course of the study other aspects of avian foraging came into focus and will need further investigation. This is the first attempt to discover the mechanisms underlying the foraging behaviour of these two species in vineyards in New Zealand, and will hopefully provide a platform for further investigations of elements that encourage some bird species to attack fruit, and how this may be prevented.

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## **Chapter Two**

### **Bird management in vineyards**

#### **2.1 – Seasonality**

##### **2.1.1 Vineyard seasons**

In cool climates grapes are seasonal, experiencing dormancy in winter, and ripening from late summer through autumn. In May leaves fall, and from May to September in New Zealand the vines are bare. Winter pruning removes up to 90% of dry matter from the vines. From September to December green growth is vigorous, with flowering, pollination and fertilisation, followed by fruitset in November or early December, whereupon the berries begin to grow in size through cell division, but are still hard and green. Grapes pass through véraison (colour change) when seeds are mature in late summer, and thereafter increase in size due to water accumulation, while sugar content increases and acidity decreases (Lavee and Nir, 1989; Coombe, 1992). Growers frequently report an increase in bird predation on grapes, and an increase of bird populations, as soon as the grapes turn colour. Whether it is the bird populations or the depredation that increases has not been tested. It is common in New Zealand to see blackbirds, starlings, sparrows, finches, thrushes and mynas in vineyards throughout the year. It seems that silvereyes appear later in the season, and are not so frequently seen at other times of the year in vineyards (Stanley and Lill, 2001, 2002). Personal observation suggests that many bird species nest in shelterbelts and tree assemblages near vines, and that these birds frequent the vineyard even when there are no grapes. They can be seen hopping on the wires, probing the ground, and birdsong indicates that they are present in the trees.

### 2.1.2 Seasonality of bird frugivory

Seasonal variation in diets of birds is well documented (Verner, 1984; Sherry, 1990; Stanley and Lill 2001; Bairlein, 2002). Birds that frequent vineyards throughout the year forage differently when grapes are absent, according to the season. Blackbirds, starlings, mynas and thrushes take arthropods, worms, snails, and insects. Finches and sparrows are seed eaters (Sherry, 1990; Bautista *et al.*, 1998; Jordano, 2000). Most generalist frugivores move from an insect-based diet to frugivory in autumn (Stiles, 1980; Levey and Karasov, 1992; Afik and Karasov, 1995; Bairlein, 2002). The reason for this shift was thought to be a lack of available insects, due to stages of insect development (Johnson *et al.*, 1985).

Recent literature suggests that the natural scheme is more complex, and that birds actually need to eat fruit in autumn to induce metabolic changes and over-eating (hyperphagia) in preparation for winter (Jenni-Eiermann and Jenni, 1996; Bairlein, 2002), and do not need the protein rich diet of insects at this time of year. Whether the needs of birds have exerted a selective pressure on both plants and insects to be available when birds need them is an intriguing point, but it is more likely that birds have learned to maximise the food substrates that are available at certain times of year (Bairlein, 2002), i.e. the selective process has acted on the birds. Whether the autumnal needs of birds for fruit (*ibid.*) and not for insects, has influenced the life-cycle of insects is doubtful, but the possibility must not be overlooked.

In tropical regions there are many specialist frugivores that find fruit throughout the year (Herrera, 1985; Gautier-Hion *et al.*, 1985; Stanley and Lill, 2002), but all avian species that occur in New Zealand vineyards are only seasonally frugivorous. Availability of the fruit is probably a significant factor for birds that are permanent residents of the vineyard.

Therefore, the relative weighting of grape availability versus endogenous bird drivers

needs to be investigated in order to better understand how to manipulate their behaviour to reduce grape attacks. For birds that enter the vineyard at grape ripening the attraction of the grapes themselves is clearly significant, and the need to clarify their motivation even more obvious.

### **2.1.3 Seasonal dietary needs of birds**

Seasonality is an important consideration of bird behaviour. The annual activity of nesting and producing progeny takes place for most species in spring and early summer. Birds need find a suitable nesting site, attract a mate, and, depending on species, males need to defend their site, mate and food resource, and together they must build a nest. The female lays eggs and incubates them. Parents must then feed their young through to the nestlings leaving the nest. For blackbirds and similar passerine species, nestlings remain in the nest for 2-3 weeks, and birds often nest again (Hayman, 1999). During this period birds need to find high-protein food for egg production, and for nestling growth and feather development processes.

Once out of the nest fledglings forage for themselves. Death eliminates up to 60% of blackbird fledglings within the first month of leaving the nest (Magrath, 1990), some of which may be due to inability to forage successfully (Morse, 1990). Arthropods are more difficult to find (due to a life cycle beginning in Spring, where most have by autumn assumed a more mobile form, Herrera, 1982), and juvenile birds have less well-developed feeding skills than adults (Lawrence, 1985). The ground becomes hard and worms descend out of reach (Feare, 1984). Accessible water may also become scarcer in some locations (Herrera, 1982). Autumn is the time of year when many fruits become available, some of which may be poisonous to some birds (Cipollini and Levey, 1997). Juvenile birds are low

in the pecking order, as dominant males and females with nestlings to feed exclude them from easy food sources (Herrera, 1982; Morse, 1990). Consequently many juveniles forage on fruit, which is an easy prey (Stevens, 1985) even though fruit is an inadequate diet to sustain bird life (Herrera, 1982; Wheelwright, 1985; Bairlein, 2002). Those birds that survive into winter have undoubtedly acquired survival skills. Morse (1990) suggests that many passeriform species suffer heavy losses while learning that they have to narrow their foraging repertoires.

#### **2.1.4 Bird populations in autumn**

Autumn is a time of heavy bird pressure on food resources. Each breeding pair of blackbirds, silvereyes and starlings may have raised at least two broods of up to six nestlings (Kinsky and Roberston, 1987). If even only 40% of these have survived the bird population has increased by 250-300%. It is likely then that the growers' perceptions that there are more birds when the grapes turn colour is accurate. That these birds are eating grapes when previously they did not is also likely to be accurate, and not merely a perception due to a heightened awareness.

### **2.2 Economic impacts of birds in vineyards.**

#### **2.2.1 Economic value of grapes**

The value of the grapes taken by birds varies according to the price that may be commanded. The average price for grapes in New Zealand is approximately NZ\$2000 per tonne, though this varies considerably with cultivar and quality. Gisborne may only command \$600 per tonne for Müller –Thurgau while Central Otago Pinot noir may sell for NZ\$4000 per tonne (NZ Winegrowers Statistical Annual 2003). Gisborne may easily produce twelve tonnes per acre while Central Otago will limit yield to 2.5 tonnes per acre



to command the top price. Many wineries work on the principle that lower yield equates to higher quality. Between the two extremes stretches a continuum with all possible combinations of cultivar, quality, yield and price. But New Zealand does not produce the extremely high yields that are known in other parts of the world. Grapes in New Zealand can be therefore individually more valuable than in some other winegrowing regions, and the loss or damage to grapes from birds correspondingly of greater economic significance.

### **2.2.2 Environmental correlates of bird pressure**

Apart from areas of Hawkes Bay, Marlborough and Central Otago, where vine plantings dominate, any vineyards will occupy only part of the landscape. Other activities include all types of farming, crop and livestock, as well as horticultural and residential land use. A landscape of exclusively vineyards with few trees experiences lower bird pressure than vineyards surrounded by trees and buildings, open land and other crops, all of which correlate strongly with bird pressure on grapes (Saxton, unpublished data).

### **2.2.3 Expenditure parameters**

The amount growers are prepared to spend on bird control methods does not strongly correlate with the value of their grapes, nor with the bird pressure on their vineyard (Boyce *et al.*, 1999). That an economic analysis of total bird management costs does not precede bird management decisions is a reflection of the lack of robust data and of tools to collect such, as well as a lack of understanding of the costs and benefits of bird pest control.

## **2.3 Bird control**

Attitudes to pesticides have changed in recent years, with much less tolerance for possible carcinogenic or other long-term residual effects, especially on crops that are destined for

human consumption. At the same time advances in bio-control for insects and fungi have offered alternative control measures for these damaging pathogens. For birds, however, no such advances have been made, with the result that grape growers suffering bird damage are denied chemical controls, and are left with few viable options for bird control.

### **2.3.1 Chemical repellents**

Mesurool® was an effective bird repellent developed in the 1980s (Cummings *et al.*, 1994; York *et al.*, 2000), and was in use in New Zealand until 1992. It has methiocarb as its active ingredient (3,5-dimethyl-4-methylthiophenol, Porter, 1992; York *et al.*, 2000) and works by causing the bird post ingestional distress from which it learns avoidance behaviour (Shah *et al.*, 1992; Conover and Messmer, 1996). However, methiocarb (a precursor to methyl carbamate, which is carcinogenic and not tolerated for human consumption) came to be considered unacceptable at the levels it was detected in wine (Porter, 1992). The cost of meeting the conditions of use imposed by the United States Environmental Protection Agency became uneconomic and use of methiocarb or Mesurool® for bird control on food crops was abandoned. In January 1992 the Wine Institute of New Zealand notified their members that the maximum residue limit of 3ppm Mesurool® had been cancelled, and that they should be sure that grapes they bought were Mesurool®-free (Gregan, 1992).

There is much chemically oriented research seeking an efficient bird repellent (Shah *et al.*, 1992; Clark and Shah, 1994; Cummings *et al.*, 1994; Stevens *et al.*, 1998; York *et al.*, 2000). The main contenders at present are the acetophenones (including methyl anthranilate) that irritate birds' trigeminal chemoreceptors (Shah *et al.*, 1992) and anthraquinones that are accompanied by post-ingestional distress and vomiting (Avery *et*

*al.*, 1998). A third compound is pulegone, found in the mint plant (*Mentha spp.*), which appears to cause both sensory irritation and post-ingestional distress (Avery *et al.*, 1996b) and has volatile cues that may prove aversive to birds (Wager-Page and Mason, 1996a).

Methyl anthranilate (MA) is a naturally occurring compound in some fruits and especially in *Vitis labrusca* grapes, where it expresses a distinctive taste/odour known as 'foxy' (Shah *et al.*, 1992). Research on this compound was intense and ongoing (Cummings *et al.*, 1994; Aronov and Clark, 1996; Avery *et al.*, 1996a, b; Siehl *et al.*, 1997; Avery and Mason, 1997; Stevens *et al.*, 1998). But, although available in the United States as ReJexit® where it is used to deter geese from grass and other similar uses, it is not readily available in New Zealand. It did have formulation difficulties which limited the length of effectiveness (Avery *et al.*, 1996a) and it is not clear whether these have been satisfactorily resolved for grapes. Aronov and Clark (1996) found that it degraded in light, and 100% was lost after 20 days. Sinclair and Campbell (1996) found that silvereyes avoided MA and ate only 7% of food treated with MA even if no other food were available. The retail price of ReJexit® (USD120 per hectare) compares favorably with netting. MA's active ingredient, 2-aminobenzoic acid methyl ester, is approved for human consumption, but is offensive to birds because of its irritant qualities (Avery and Mason 1997). The chemical structure (a phenyl ring with an electron donating or a basic group) is typical of a group of compounds that includes benzoates and acetophenones, which are also under investigation as possible repellents (Shah *et al.*, 1992; Cummings *et al.*, 1994; Sinclair and Campbell, 1996; Wager-Page and Mason, 1996b).

Ortho-aminoacetophenone and other aminoacetophenone isomers have repellent properties to birds similar to those of methyl anthranilate (Wager-Page and Mason, 1996b). Another

chemical with apparent bird repellent properties is anthraquinone (Avery *et al.*, 1998), which induces gastric malaise possibly induced by a rise in body temperature (Coleman, 2001). Benzoates, which are also acetophenones, are used experimentally as a punishing stimulus in conjunction with other repellent stimuli (Miele *et al.*, 1988). They impart a bitter taste that is absorbed by plants, and which lasts for years, and benzoates are not recommended for use with edible plants, which includes grapes destined for wine production. They are used to deter deer ([www.deerrepellents.com](http://www.deerrepellents.com), 2003). There is little evidence that any of these repellents are in widespread use with birds in New Zealand. Commercial adoption of chemical repellents appears to lag behind scientific progress, due to the costs of regulatory requirements and the limited market potential (Avery *et al.*, 1998). Price becomes an issue in the practical use of chemical repellents, as even at low rates of application they are inevitably priced to recoup the cost of research, though York *et al.* (2000) noted that growers are prepared to spend up to USD370 per hectare for a repellent. In Great Britain, evaluation of chemical repellents is limited to registered products, as the cost of developing new compounds and registering them is considered to be too high (Coleman, 2001).

Tannins have bird repellent properties due to their anti-nutritional effects, and high tannin grains are known to be repellent to birds. Tannins can cause digestive disturbance in birds as well as in mammals (Butler, 1982; Dearing *et al.*, 2001). Protein digestibility decreases due to tannins binding to digestive enzymes or to dietary protein. Although tannins are present in ripening grapes, and are sometimes added to wine during vinification to increase colour stability of red wines, the concentrations in grapes are too low to repel birds. There is research that suggests that birds, particularly frugivorous birds that are seed-dispersers, have the ability to tolerate high levels of tannins, either by defecating quickly, by

regurgitating, or by varying their diet by incorporating protein to dilute the tannin effect (Izhaki and Safriel, 1989). Plant secondary metabolites that have toxic properties caused by elevated need for water upsetting avian physiology are also being examined (Dearing *et al.*, 2001). Plant alkaloids such as ergovaline, an endophyte in ryegrass, and endophytic fungi in fescue, are also subject of research projects to repel birds (Conover and Messmer, 1996) Much of this research is aimed at repelling large birds from airports. The grass is not for consumption, and endophytes are known to cause illness such as ryegrass staggers in stock animals. These compounds may not be of any use with crops destined for human consumption.

Birds rarely avoid chemosensory irritants that affect mammals. Starlings are indifferent to ammonia and capsaicin, and insensitive to gingerol and piperine, all of which are avoided by mammals (Wager-Page and Mason, 1996a). Our knowledge of birds' senses is incomplete. There is no known irritant to mammals that is also avoided by birds (Shah *et al.*, 1992). The nature of sensory perception of aromas appears to differ between mammals and birds. Both mammals and birds have a benzene site in trigeminal chemoreceptors, but birds lack the thiol-hydrogen bonding site needed to activate the benzene site. This accounts for the difference in sensitivity to aromatic irritants such as ammonia or capsaicin (Shah *et al.*, 1992; Clark and Shah, 1994).

### **2.3.2 Exclusion - Nets**

Netting places a physical barrier between the birds and the fruit, which is more or less efficient depending on the way the net is applied, the diligence with which it is maintained, and the persistence of the birds. Little is known about the persistence of the birds, but it is possibly linked to other environmental factors, and especially to the availability of

alternative food sources, whether these are another fruit crop, or ‘sacrificial rows’.

Persistence behaviour may possibly embrace other factors such as behaviour of conspecifics, endogenous nutritional needs, and it is likely the birds will be less persistent if they have had less time to develop the habit of eating grapes. If the nets are applied early they may be more effective at keeping birds out. The disadvantages of nets, apart from the original cost which is high, are the cost of labour to install, remove and maintain them. They are expensive, costing approximately NZ\$2 per metre row for all types of netting. Depending on the value of the crop the grapes in that metre row may be worth only a little more. If the netting lasts several years then it is worth the expense. Netting is still the most effective method of exclusion (Boyce *et al.*, 1999), and is in continued widespread use throughout the world for high value crops, especially fruit crops, as protection against many types of predators. In the avian risk-reward model, if a vineyard is relatively predator-free, and the grapes readily available, the energy and time required for a bird to penetrate netting may be well spent.

There are several netting styles in use in New Zealand vineyards. They are made from polymer and are put on at véraison, and taken off at harvest and stored. They include side-netting that encloses both sides of the vine at the fruiting zone only, over-the row netting that covers the whole vine down to the ground, and multi-row nets, which range from double row to eight-row nets, that require mechanised application. Netting the vine can modify the microclimate within the net, increasing humidity, which may increase the occurrence of fungal infections (Boyce *et al.*, 1999), while also reducing the efficiency of any spray application. Netting can also reduce canopy photosynthesis, which reduces the accumulation of photosynthates in the maturing grapes (Trought *et al.*, 1997; Creasy and Trought, 2001). The fact that use of netting is widespread (Boyce *et al.*, 1999) indicates the

economic value that vineyards set on predicted losses to birds, and the assumption that this is the most effective protection.

### **2.3.3 Acoustic bird control**

Birds possess the ability to hear a range of auditory frequencies that is only slightly less than that for humans (King and McLelland, 1984), i.e., humans can hear what birds hear. Contrary to popular belief, they have no ability to detect high frequency sounds such as dogs have (*ibid.*). They do however have a temporal resolution about 10 times faster than humans, and the ability to locate objects by sound is accurate in nocturnal birds (*ibid.*). Several systems have been developed based on repelling by auditory means, the main approaches being to startle the birds (gas guns, noisy vehicles), to interfere with their comfort zone (electronic sounds of an irritating nature), or to simulate alarm or distress calls to elicit a fear response.

Sound fencing is an approach that creates a ‘no-go’ area for birds by making it uncomfortable for them to pass through the sound. There has been some research into the effect of sound on birds (McGregor *et al.*, 1983; Aubin, 1990) and there is no doubt that they, like all vertebrates, do hear and respond to sound. The type and consistency of response varies so that it is not yet a reliable way to control birds by manipulating their behaviour. It is also likely that habituation is involved (Aubin, 1990), since most acoustic methods do not work for long.

Apart from sound-fencing, alarm and distress calls that will elicit a flight response from the birds is the other main area of research in acoustic control (Aubin and Bremond, 1992; Gorenzel *et al.*, 2002). At present, researchers that are looking for a repellent are only

measuring latency of a flight response, with a possible secondary interest in the latency of the birds' return. In view of the importance of species-specific birdsong, there is possibly a whole gamut of avian responses to sound, which is undoubtedly very sophisticated (McGregor *et al.*, 1983). Future research might well develop acoustic tools for bird control, if an understanding of avian response to sound can be gained.

Acoustic bird control is employed to some extent by airports, but there is little scientific documentation or analysis of this success that was locatable through scientific databases. The major portion of literature on birdstrike in aviation consists of analysis of the cost and potential cost of birdstrike to airlines and the military. Airport environs can be stripped of any features attractive to birds by regulating the distance to any crop, water feature, landfill site or other environment that might attract birds. Consequently the reported success of airport scaring methods that include acoustic scarers might well be more a reflection of the environmental manipulation than the effectiveness of these control methods.

#### **2.3.4 Biocontrol**

Biocontrol uses natural systems to manage populations, among which are bio-agents that reduce reproductive success, that induce sterility, or natural predators, to control populations, and is known to be effective for insect populations and for fungi. Predatory birds or other predators (including dogs, cats and also humans) are thought to be effective if sustained pressure is brought on the target species. Falconry, another method enjoying reported success for airports, is often suggested. So far trained falcons and falconers are too expensive, but the hunting characteristics of falcons would possibly deter many smaller birds and have a significant effect (Fox., N. pers. Comm., Nov 2004). In New Zealand it is illegal to keep native or self-introduced birds without a licence (Hayman, 1990). Free-



ranging Australasian harrier hawks are an option that many growers in New Zealand use if they can attract them. Usually the strategy is to set out carrion meat to attract the hawks, and there are many anecdotal reports of partial success. There has to date been no scientific evaluation of the effectiveness of this method, its main disadvantage is the unreliability of hawk presence.

Cats and dogs are sometimes used to good effect. But both tire and are often unable to keep up with the numbers of birds to control. They are an ancillary control, assisting often with chasing birds out of nets once or twice a day.

If man is considered a natural predator, then shooting can be classified as biocontrol.

Shooting is the most used method of bird scaring in New Zealand. Literature on shooting is unanimous in declaring it an ineffective method of population management (O'Connor and Shrubbs, 1986, p224; Coleman, 2001), and there are many instances that show that shooting is not likely to be an effective method to reduce avian populations (due to re-invasion if conditions are favourable to foraging species). Consequently, shooting will not be an effective measure for reducing pressure on the grape crop. Growers clearly feel, however, that this method is worth employing (Boyce *et al.*, 1999), though whether the shooting does more to relieve the grower's stress levels than to effect any reduction in bird pressure remains unevaluated.

## **2.4 Evaluation of present bird management**

### **2.4.1 Adhoc nature of bird control**

Management at present consists of attempting to reduce bird pressure on grapes, preferably by beginning control before the birds begin to eat the grapes. It is not, with our present

state of knowledge, possible to manage bird populations, species, or to control their movements. Consequently methods used to reduce bird pressure are evaluated by growers individually and employed locally where they are seen to be most effective. Reducing bird pressure involves either reducing bird numbers or reducing attacks on grapes, but we do not know much about the hierarchy of cues or the matrix of factors that are implicated in birds eating grapes, or what causes increases in bird numbers when grapes ripen.

#### **2.4.2. Thresholds**

There is only one threshold to consider – the economic threshold where the amount spent is no longer recouped in grapes saved. A very basic difficulty is that it is impossible to know how much damage is prevented (Boyce *et al.*, 1999). However this is the parameter with which all growers must work, and its elusiveness causes some growers to spend much more on bird control than others. It is linked to the value of the grapes and the perceived urgency to harvest as much as possible. Other factors such as a desire not to use nets, or not to shoot, may play a part in some growers' decisions.

#### **2.4.3 Knowledge of birds**

Growers' knowledge of birds is very variable. A recent publication of the "Sustainable Winegrowing New Zealand" is a bird identification booklet (Hamilton, 2003). Some bird species encountered in vineyards do not attack grapes at all, and may be beneficial in occupying perches and nesting sites that might otherwise be occupied by grape foragers. It is important that growers know their bird species and understand the ecology of birds in their vineyards. This will assist in decision-making when considering the economic threshold mentioned above.

#### 2.4.4 Present state of advice to growers for bird control

Historically bird control has been reactive, with growers taking no notice until damage reaches an intolerable level each year, by which time the season's bird foraging patterns are well-established (Porter, 1992; Sinclair, 2000; Lange, 2003). The following summary of advice (Jensen, 1974; Nelson, 1990, 1991; Porter *et al.*, 1994; Sinclair, 2000) applying particularly to resident birds, continues to be valid:

- *Control of resident birds must start before predation on the grapes begins;*
- *Work with neighbours to control effectively;*
- *Have a control strategy in place throughout the year, not just for the 2 month ripening period;*
- *Remove nesting and roosting sites - plant thin shelter belts, remove large shelter trees, prevent nesting on buildings;*
- *Avoid small plantings as small isolated vineyards will suffer more than large expanses;*
- *Establish a shooting regime to alarm birds before using a gas cannon;*
- *Wear bright clothing in the vineyard;*
- *If economically viable, have a decoy crop or a sacrificial row, from which it is important not to scare the birds.*

The principle underlying this advice is that the resident birds are in the locality throughout the year, and will not be easily moved to another location. Consequently the aim is to divert them to alternative food sources. Present popular understanding of birds and their behaviour is anthropocentric, reflecting little knowledge of any of the bird species concerned (P. Mawson, in Tracey and Saunders., 2001). The realisation that an alternative feeding source must be made available for birds to move to is a concept that continues to evade most practitioners of avian pest management systems that are based on trying to move birds (Avery, 2002).

## **2.5 Future management strategies**

### **2.5.1 Environmental management.**

It is clear that modification of the environment is one area where growers can influence birds (Recher, 1990; Gates and Donald, 2000), but research has not yet identified how environmental surrounds of or the vineyard environment itself impacts on birds. This is a field that urgently needs research. Meanwhile our best guesses are based on ecological literature often from other countries and concerned with other bird species. It is interesting to note that the very species that are increasing in New Zealand due to intensification of horticultural methods (Nelson, 1990; Porter *et al.*, 1994) are declining in Britain, the blackbird by 33%, starling by 45% and sparrow by 64% (Crick *et al.*, 1998) and that this decline is attributed to intensification of horticultural and agricultural methods (O'Connor and Shrubbs, 1986; Gates and Donald, 2000; Isenmann, and Debout, 2000). This dichotomy points to a need for more New Zealand specific research to pinpoint environmental factors that favour population increase of species that attack crops. Such factors may include milder winters in New Zealand than Britain, or more extensive grassland.

### **2.5.2 Ecological manipulation**

Bio-control involves using nature to achieve the ends we as humans desire. It is again clear that without the underlying knowledge of natural mechanisms, such as bird behavioural motivational drivers, and the hierarchy of ecological cues, we are unable to design a tool that could use the environment as a bio-control.

### **2.5.3 Long term aims.**

The long term aim of vineyard managers, and of growers of other crops that birds attack, is to minimise loss of crop, of money, energy and time. The fact that pressure on crops from birds is not consistent or equally heavy, and can vary significantly in quite a small area, suggests that local conditions are in part responsible for the problem. There will be underlying principles at work that could be researched, such as nutritional requirements of the individual birds and relative factors that create a favourable or hostile immediate environment. The results could be applied in a relatively short space of time.

### **2.6 Place of this research**

One of the main drivers of this thesis is to isolate significant cues that are perceived by birds, and to attempt to elucidate the underlying reasons why some bird species attack grapes, and why the attacks increase towards harvest. From this point further research into alternative strategies could enable growers to minimise populations of grape-eating birds in vineyards.

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## **Chapter three**

# **Experimental method to investigate and monitor bird behaviour and damage in vineyards. <sup>1</sup>**

Acknowledgement: Funding for the Marlborough Survey from New Zealand Winegrowers.

**Keywords:** artificial grapes, bird damage.

### **3.1 Abstract:**

A method for examining foraging decisions by birds damaging grapes in vineyards was developed using a bird feeder table, time-lapse video, and artificial foods. Behavioural responses of blackbirds and silvereyes to the experimental situation and in a natural vineyard setting were compared. The two species behaved in a similar way in the artificial and natural situations, indicating that the experimental method used is a valid and useful tool to investigate bird decision-making mechanisms with natural grapes.

### **3.2 Introduction**

Damage to grapes from bird predation is a world-wide problem, generally chronic and locally extremely severe. Bird control methods remain primitive and largely ineffective, scientifically untested, often eliciting public disapproval and legal constraints. There is very little understanding of when, why, or even which bird species arrive at vineyards, or why birds take grapes at all, given that grapes are a low-level food resource, providing insufficient nutrients for the birds to survive (Karasov and Levey 1990, Bairlein 2002).

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Since Murphy and King (1982) recommended the use of semi-synthetic diets in laboratory experiments, bird species' dietary characteristics and choices have been frequently investigated using artificially created food sources. Examples include investigations of physiological processes (Afik *et al.* 1997; Totzke *et al.* 1998), color choice experiments (Avery *et al.* 1999; Puckey *et al.*, 1996), experiments in sucrose intolerance (Brugger *et al.*, 1993; Avery *et al.*, 1995), and experiments to develop chemical deterrents to birds (Clark and Shah, 1994; Sinclair and Campbell, 1996). However, the use of artificial foods in a free-range situation is somewhat less frequent, possibly due to the difficulty of controlling the variable of bird population. The main use of field trials is to confirm bird reactions to chemical repellents (Curtis *et al.*, 1994; Cummings *et al.*, 1995). Stephens (1990) suggests that watching foraging behaviour may be the most informative way to study diet choice, and Moermond (1990) suggests that understanding motivation for foraging behavior will be most comprehensive when observations are keyed to how birds respond to resources in a given context. The aim of the study reported here was to create an experimental method where the bird behaviour would be a close reflection of behaviour in the vineyard context, which could serve as a method for further investigations.

Watkins (1999), in a study of bird behaviour with purple and green grapes, found that natural grape parameters, such as sugar concentration, were confounded by the ripening process. In this experiment an artificial grape was developed to separate out different parameters of ripening grapes, and offered on a bird feeder table to free-ranging European blackbirds and smaller silvereyes. Behaviour of the birds was recorded and later analysed. A study of bird populations in the Marlborough region of the South Island of New Zealand was conducted to compare the detailed behaviour of birds in our artificial situation with that of the same species

in vineyards, in order to determine the relevance and value of behavioural observations in our experimental situation.

### 3.3 Materials and methods

**Table experiments:** The experiments were conducted in the Lincoln University Vineyard, Canterbury, New Zealand (blackbirds), and in a pear orchard 4 km from the University (silvereyes). The feeder tables were placed approximately 8 m from shelter trees, which the birds appeared to use as cover. A novel two-tier bird feeder table was used, in which each tier was 50 cm long and 9 cm wide. The top tier was set back approximately 30 cm above the lower tier, allowing birds simultaneous full vision of both tiers. The table was set at a height of 1.5 m, about 1.5 m in front of the video camera. On the table were placed, according the experiment being conducted, artificial grapes (10 on each level) or pots with artificial nectar.

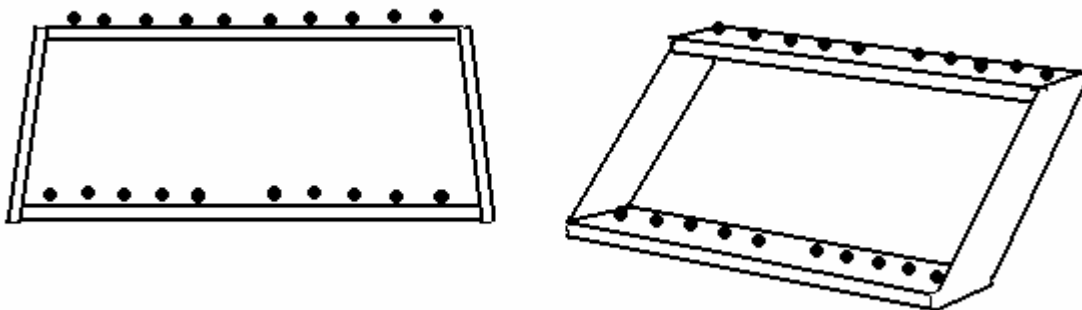


Figure 3.1 Bird feeder table. The table is 50cm wide, each level 15cm across, upper level offset 30cm above lower level. The table is 1.5m above ground. There are 10 grapes on each level.

Bird behaviour was recorded by four colour TPC 5504 EX (Elmo, Nagoya, Japan) cameras with a varifocal autoiris 6 to 12 mm lens (Pentax, Golden, CO). Three cameras were connected to battery-powered Panasonic VHS Time Lapse recorders (model AG-1070DC), and one to a STR960P Samsung time-lapse VCR (with quasi sine wave inverter). The

recorders were set to 24-hr recording on a 3-hr tape, which corresponded to a speed of 7 frames per second. Birds were recorded between 7 am and noon. Tapes were viewed and the behaviour quantified by recording the following: time of arrival on camera and departure of each bird, time of attacks on grapes; how many grapes were pecked, or taken, and the number of birds that appeared together.

The artificial grape mixture used in experiments was 2 g agar (Germantown, NZ) and 2 g



Figure 3.1 Piping the artificial grape mixture into a candlewax mould.



Figure 3.2 Artificial grape after setting.

gelatine (Davis, Christchurch, NZ) in 50 mL water. Additional compounds were added according to the experiment. The mixture, except for the gelatine, was heated to 70°C, the gelatine then added and the mixture piped into 5 mm diameter wax moulds at 55°C and allowed to solidify. Artificial grapes were similar to real grapes in colour, size, and consistency, although they did not have seeds or skins. For some experiments, artificial nectar was made by dissolving 15% glucose/fructose in 100 mL near boiling water.

**Vineyard recordings.** The same recording equipment was used in four vineyards in Marlborough, New Zealand, in March 2002 (see Saxton, 2002). Four cameras recorded in a Chardonnay vineyard, 1.2 m vine, 2 m row spacing, on VSP trellis, planted three years previously. Cameras were placed 1.5 m from a vine focused at the level of the fruiting wire (90 cm from the ground), giving a clear picture of the grapes of one vine, (about 1.2 m length of canopy). Two cameras were positioned at the end of two rows, 10 m apart. The other two were positioned 50 m further into the interior on the same rows. Recording was from 7am to noon for 10 days and from sunrise to sunset for 7 more days. Analysis was similar to that described above. The majority of the data recorded were on cameras at the ends of the rows.

Data from the experimental and the vineyard situations were compared for the two species. The following were recorded: length of feeding bouts (seconds on camera of each individual bird), number of grape attacks at each bird visit (grapes taken by blackbirds, and pecks per individual bird for silvereyes), intervals between visits for each species, number of birds appearing together (occasions when bird visits overlapped proving that more than one bird were present) and type of grape damage (whether the bird had pecked the grape or taken it away). Because of the small datasets and uneven distribution (common in ecological data),



Figure 3.3 Video camera with recorder and battery trained on a vine in Marlborough



Figure 3.4 Experiment set up in a vineyard.

data were analysed by non-parametric tests, Mann-Whitney U test (U) and Kruskal Wallis ANOVA (H), using GenStat 6 software (VSN, Herts, UK), according to Lehner (1996).

### 3.4 Results and Discussion

**Length of feeding bouts, (Table 1).** With artificial grapes, blackbird visits were very short, though occasionally they would perch on the feeder table for some seconds and appeared to be vigilant between grape attacks. Silvereys visits to artificial grapes and to the natural grapes were significantly longer than those of blackbirds ( $U = 709$ ,  $p < 0.001$ ,  $n = 65$ ;  $U = 234.5$ ,  $p < 0.001$ ,  $n = 34$  respectively).

**Number of grape attacks (Table 1).** Blackbirds removed artificial grapes from the feeder table one grape at a time, while silvereys attacked many more artificial grapes per visit ( $U = 483$ ,  $p < 0.001$ ,  $n = 65$ ). In Marlborough blackbirds took grapes singly, only on one visit did a blackbird take more than one grape. Silvereys fed at more locations, and even with grape

**Table 3.1. Feeding data means (median, se) n = bird visits.**

	Seconds at one location	Number of birds together	Number of grapes attacked per visit	Interval between visits (mins)
<b>Blackbirds</b>				
Experiments* (n = 65)	13.4 (6,2.5)	1.1 (1,0.05)	1.8 (1,0.2)	40.9 (28,4.1) min 2, max 165
Marlborough+ (n = 50)	9.7 (5.5,1.76)	1.05 (1,0.05)	1.2 (0,0.3)	92.8(89,18.3) min 20, max 180
<b>Silvereys</b>				
Experiments* (n = 34)	49.4 (35.5,4.7)	2.5 (2,0.3)	16.9 (14,1.8)	22 (4,4.9) min 1, max 191
Marlborough+ (n = 46)	38.4 (27.5,5.6)	1.5 (1,0.1)	4.1 (4,0.4)	52.7(17,19.5) min 1, max 360

\* Data extracted from hexose sugar experiment (Saxton et al., 2004b)

+ Data from 16 days survey in Marlborough vineyards

Experiments: Blackbirds were recorded in October 2000 at the Lincoln University Vineyard and silvereys at a pear orchard 4 km from the vineyard.

Marlborough survey (natural context): both species were recorded in March 2002 at Hunter's Gosling Vineyard near Renwick, Marlborough, New Zealand.

Video recordings were analysed and data collected were: time of arrival and departure of each individual bird, time spent feeding, number of grapes taken (blackbirds) or number of pecks (silvereys).

attacks at each location conservatively scored as one (the video picture was not close enough to see whether they sampled more than one grape at each location), they delivered many more attacks per visit than blackbirds ( $U = 204$ ,  $p < 0.001$ ,  $n = 34$ ).

**Intervals between visits (Table 1).** With artificial grapes a blackbird visit tended to occur at regular intervals, thus the median interval period was close to the mean. Silvereye visits to artificial grapes tended to cluster, with long intervals between the clusters, which causes the median to be much less than the mean. Interval length between visits differs significantly between species ( $U = 1326$ ,  $p < 0.001$ , 77 bird visit intervals). In the recordings with natural grapes, the interval between blackbird visits was more regular (median close to mean) than for silvereyes. The difference between species was again significant ( $U = 29$ ,  $p < 0.001$ , 14 intervals). Comparing experimental and field data for each species, there was no significant intra-specific difference between interval lengths for blackbirds ( $U = 63$ ,  $p = 0.1$ ), nor for silvereyes ( $U = 144$ ,  $p = 1.0$ ). A broad pattern of evenly spaced regular intervals between visits by blackbirds, while silvereye visits were clustered and interspersed with long intervals of no visits, appeared to be similar in both field and experimental situations.

**Number of birds visiting together (Table 1).** Blackbird visits to the Lincoln bird feeder table and to the Marlborough vines were almost exclusively solitary. Silvereyes often fed at the feeder table in groups, with birds flying in and out of the camera view making it difficult to determine how many individual birds were present. In the Marlborough recordings, out of 24 visits recorded, 8 were of two or more birds feeding close together on the vine.

**Type of grape damage.** With artificial grapes, blackbirds arrived on the feeder table and took a grape in their beak, swallowing it either immediately or after a short time or sometimes



flying off with it. Grapes were thus completely removed from the feeder table. In the Marlborough vineyards blackbirds always jumped up into the vines from below, plucked a grape from a cluster and flew off. This resulted in complete removal of grapes, leaving the brush only behind. Silvereyes, on the feeder table, pecked at the grape *in situ*, often sampling many grapes on one visit. In the Marlborough vineyards they sampled several different spots, usually high in the fruiting zone, leaving grapes damaged on the vine. Blackbird damage results in loss of yield while silvereye damage reduces quality of the grapes left.

### **3.5 Summary**

Collection of data from the Marlborough vineyards was not easy – un-netted vines were rare, the location was distant from our base, weather variability and reliability of recording equipment was also problematic. Focusing on one vine only out of a whole vineyard meant that obtaining a reasonable sample size was difficult. It was encouraging that our results showed that behaviour of our two target species was similar in the controlled field experiment to that in the field. The use of time-lapse video recording many hours of behaviour in fine detail enables thorough analysis of bird behavioural characteristics, formation of more specific hypotheses for further experimentation, and robust results. Such experimental tests have examined the effect of grape ripening variables, such as hexose sugars (Chapter 4), grape aromas (Chapter 5), acids (Chapter 6), and tannins and colour (Chapter 7), on behaviour of these two species in the vineyard.

Moermond (1990) suggests that foraging behaviour offers important clues to assessing and interpreting the food exploitation patterns and capabilities of birds. The behaviour of these two species led us to question why there should be such differences in species using the same food resource. Karasov has determined two different mechanisms of glucose absorption in

birds (Karasov, 1990; Afik and Karasov, 1995; Caviedes-Vidal and Karasov, 1996; Afik *et al.*, 1997). Blackbirds exhibited behaviour indicative of protein-eating or generalist-frugivorous birds that have a less efficient glucose absorption system (Karasov and Levey, 1990), while silvereyes exhibited behaviour indicative of honeyeaters or small passerines.

The regular return of blackbirds for more grapes, the short length of time a blackbird needs to digest a grape, their regular taking of many whole grapes and their solitary visits supports the hypothesis that the two species have different glucose absorption systems. Silvereyes spent much longer feeding and pecked grapes many times without removing them. Afik *et al.* (1997) found that honey-eating birds and small passerines have a paracellular absorption mechanism that absorbs glucose very efficiently, which may be a limiting factor to their rate of intake of glucose, since the rate of assimilation will control the rate of ingestion. These birds would need to feed slowly, be selective about their food sources and track them (Morse, 1990), and for protection from predators these birds would need to feed in flocks (Lefebvre, 1986). Although it has yet to be confirmed that silvereyes have this digestive process, it would explain the behaviour of this species in vineyards. They make small punctures in grapes, they spend longer feeding, feed in small flocks and return intermittently as they track their food source (Rooke 1984; Stanley *et al.* 2002).

### **3.6 Conclusion**

European blackbirds and silvereyes cause very different damage to grapes. This is a reflection of two types of behaviour, which leads to the suspicion that endogenous drivers are not the same for these species. It is even possible that each species may be reacting to different exogenous cues of the vineyard environment. Unravelling these physiological and ecological factors will require further investigations, with robust results. That the behaviour of free-

ranging birds recorded in our experiments closely matches that in the natural situation of vineyards gives us an experimental tool for these investigations, where collection of sufficient field data for robust results would be unsustainable.

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## Chapter four<sup>1</sup>

### Comparative behaviour of free-ranging blackbirds (*Turdus merula*) and silvereyes (*Zosterops lateralis*) with artificial grapes.

**Keywords:** feeding behaviour, artificial grapes, hexose sugars, blackbirds, silvereyes, vineyard, timelapse video.

#### 4.1 Abstract

In order to detect bird responses to sugar parameters of ripening grapes, artificial grapes containing controlled concentrations of hexose sugars were offered to free-range blackbirds and silvereyes. Time-lapse video was used to observe the two species of birds feeding on grapes presented on a novel two-tier bird-table. The comparative interest displayed by the birds for grapes of varying concentrations of hexose sugars, and the time spent feeding by each species were analysed statistically, to discover the level of sugar concentration in grapes that is attractive to these birds. Blackbirds exhibited a preference for high sugar concentration, while silvereyes preferred grapes with a lower concentration. Blackbird visits were much shorter than those of silvereyes and they took whole grapes whereas silvereyes pecked. Differences in behaviour of the two species are discussed and the assumption that all frugivorous birds are attracted to fruit for similar reasons is challenged. It may be that differences in digestive glucose absorption processes underlie the observed difference in behavioural responses of the two species.

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## 4.2 Introduction

Watkins (1999) undertook field trials in Canterbury vineyards that suggested that silvereyes prefer purple grapes to green grapes. Whether this was due to colour or differing sugar concentrations remained unclear, as the various parameters of the natural ripening grapes used could not be controlled or compared. A project was conceived to manipulate grape-ripening parameters by creating an artificial grape. Artificial feeding of captive birds is a relatively common approach to identifying avian foraging mechanisms (Avery *et al.*, 1995; Puckey *et al.*, 1996; Afik *et al.*, 1997; Stanley and Lill, 2002). The experimental method reported here used a bird feeder table to offer artificial grapes (see chapter 3) with controlled concentrations of hexose sugars in a free-range situation, thus offering more external validity than laboratory experiments and more internal validity than field surveys (Kamil, 1987; Lehner, 1996).

It was uncertain how free-range birds would respond to artificial grapes, so a pilot study was run to determine whether birds would feed on artificial grapes at all in the field, whether they would show measurable differences in responses to varying hexose sugar concentrations, and to develop a method of measuring their interest. As wild birds are not constrained to feed on what is offered, natural table grapes were offered alongside the artificial ones, with a two-fold aim of attracting birds to the feeder table and ensuring that preferences measured were comparable to those for natural grapes. The pilot study also reduced the possibility that novelty would confound the birds' observed discriminations. Video recordings enabled frame-by-frame playback analysis (Guilford *et al.*, 1987) and after noting the very different behavior from blackbirds and silvereyes, data on time spent feeding by each bird species, and the relative number of grape attacks, were recorded.

Both species showed interest in artificial grapes. Silvereyes pecked, leaving them *in situ* for further pecking. Silvereye preference was assessed on the amount of pecking per visit at each type of grape, which is an accepted method of measuring bird interest (Fantino and Logan, 1979; Jordano, 1987; Puckey *et al.*, 1996; Giles and Lill, 1999). Blackbirds took whole grapes without sampling first, and this posed some difficulty in measurement. A measure of the bird's response after it had sampled the grape was needed. Therefore those grapes that were immediately dropped were considered rejected, while those eaten or taken away were classed as having been accepted.

### **4.3 Materials and methods**

The experiment reported here tested the hypothesis that all birds would prefer the highest sugar concentration in grapes offered. Subsequent to viewing the tapes, the hypothesis that there was no difference between species in time spent foraging was also tested for.

Artificial grapes of 5-6mm diameter were made with 8 drops red (cochineal) and 6 drops blue food colouring (Hansell's) and hexose sugars (50% each D-glucose and L-fructose, see Chapter 3). Initially, grapes with hexose sugar concentrations of 0%, 5%, 20% and 40% were prepared, but a second experiment with silvereyes used grapes of 0%, 5%, 10% and 20% concentrations.

Individual grapes were set out on a bird feeder table (Chapter 3). Grape positioning was randomly assigned in two stages, first each of the five types - four artificial and one natural grape type - was randomly assigned a number, and then the numbers were randomly

assigned to positions on the table. Of the 16 artificial grapes, four each were of 0%, 5%, 20% and 40% sugar concentration, and the natural grapes were 17%.

The grapes were presented to free-ranging blackbirds in the Lincoln University Vineyard, and to silvereyes in a nearby pear orchard, four km away. Bird feeding from a bird feeder table (chapter 3) was recorded on video at a slow speed approximating 7-8 frames per second. Each session ran from approximately one hour after sunrise for three hours, since this is the time of most intense bird feeding activity (Davies *et al.*, 2000), and then the feeder table was removed. There were no confounding visits by another bird species or animal during the recording sessions. Where more than one silvereye visited together, each bird was tracked separately and recorded as a separate visit.

Blackbirds were recorded for eight days in August (47 bird visits), ten days in September (48), and seven days in November 2000 (65), a total of 160 bird visits. With silvereyes fewer sessions were recorded (six sessions in October 2000 (56 bird visits), and eight session in October (124) a total of 180 bird visits.

In order to quantify each species' behaviour, separate ethograms were devised to suit the different behaviours of the two species. Silvereyes' response was quantified by pecks per visit. Since the raw data were not normally distributed, non-parametric tests were used. A Kruskal-Wallis one-way analysis of variance on mean number of pecks per bird visit to each type of grape was used to determine whether the preferences displayed by silvereyes were significant. Blackbirds did not peck but took whole grapes (which agrees with anecdotal evidence from vineyards), thus acceptance of grapes was judged on mean number of grapes of each type that were eaten or taken away per three-hour session.



Grapes that were dropped by the bird were deemed rejected. A  $\chi$ -square goodness of fit determined whether the proportion of each concentration had been dropped by chance only. Grapes not touched, or that fell (or were kicked) accidentally, were removed from the dataset prior to analysis.

The term 'attack' was decided on as a measure of damage sustained by grapes, whether it was complete removal (by a blackbird) or a peck (from a silvereye) as a means of comparing the two species' behaviour when damaging grapes. The length of each bird visit to the nearest second, in the sessions with 0%, 5%, 20%, and 40% hexose concentration grapes (66 bird visits for each species), was compared with the number of attacks on grapes for each visit. A linear regression analysis was performed on the data (Genstat 6 software).

#### **4.4 Results**

Blackbirds showed highest preference for artificial grapes with 40% hexose sugar concentration (Figure 4.1,  $N = 319$ ,  $\chi$ -square 19.44,  $df = 4$ ,  $p < 0.001$ ). Of the  $\chi$ -square individual cell chi-square values, the 40% concentration (4.39 dropped and 1.83 not dropped) and 0% (7.46 dropped and 3.11 not dropped) were the largest, showing that preference was marked at those concentrations.

Silvereyes showed preference for hexose concentration of 10%, followed by 20%. An additional experiment was run presenting 10% concentration instead of 40% with the aim of narrowing their preference. Results from these two experiments were pooled (Figure 4.2,  $N = 319$ ,  $H = 26.82$ ,  $df = 4$ ,  $p < 0.001$ ).

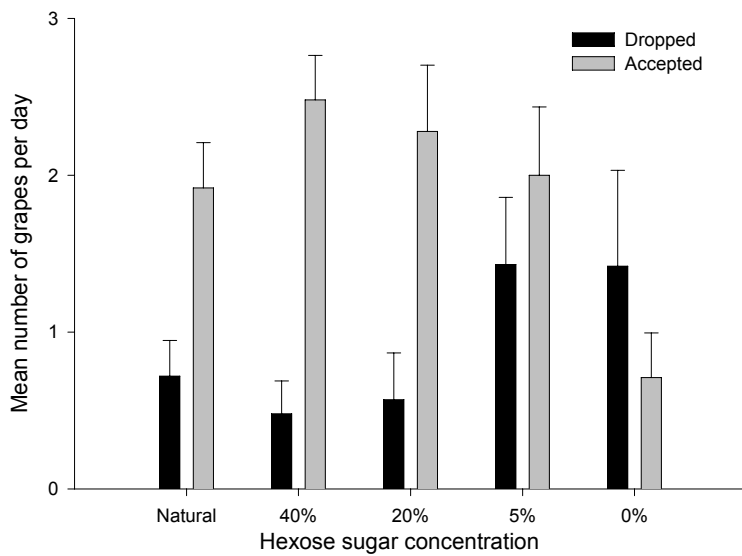


Figure 4.1: Mean number of grapes handled per day by blackbirds, showing relative numbers of grapes dropped immediately compared to numbers accepted (with SE bars).

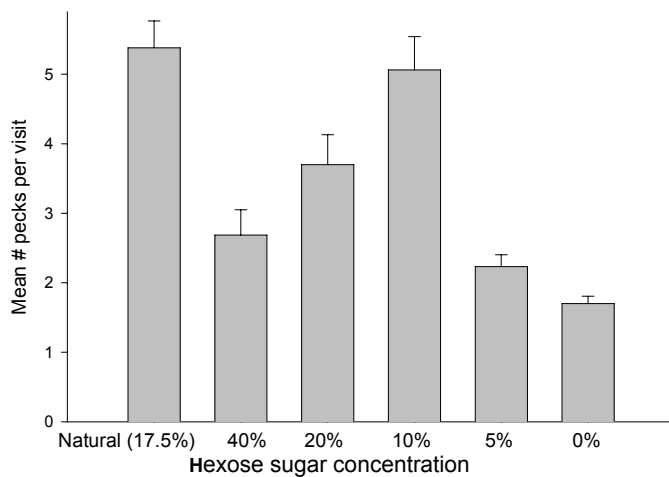


Figure 4.2: Mean number of silvereeye pecks per visit (with SE bars)

Blackbirds (seven 3-hour sessions, 64 visits, Figure 3) stayed fewer seconds on the table and delivered fewer attacks on grapes than silvereeyes (six 3-hour sessions, 66 visits). Figure 4.3 shows a strong correlation between the length of time silvereeyes are present and the number of pecks delivered ( $R= 81.9\%$ ), whereas the correlation between time and

grape attacks is much weaker for blackbirds ( $R = 40.5\%$ ). Two blackbird outlier data points, one visit of 122 seconds with 3 grape attacks, and one of 87 seconds with 3 attacks, were removed.

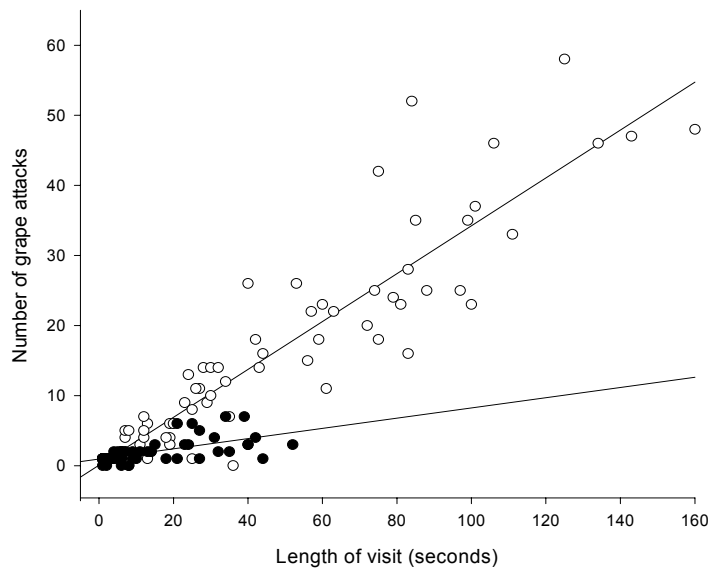


Figure 4.3: Length of stay on the bird feeder table for blackbirds (solid circles,  $n = 64$ , two outliers removed), and silvereyes (open circles,  $n = 66$ ), showing the number of grape attacks on each visit. Blackbirds attack grapes less often per visit (40.5% variance accounted for) than silvereyes (81.9% variance accounted for.)

## 4.5 Discussion

Blackbirds preference for high sugar concentration was as expected, and ranged much higher than is normally found in ripe grapes at harvest (in New Zealand usually about 25%). The silvereyes' preference for a lower concentration however was surprising, since silvereyes anecdotally do not appear in vineyards until grapes have sugar concentrations above 15%. That silvereyes stayed longer and attacked grapes more than blackbirds, leaving them in place, reflects the type of damage sustained by grapes from this species in vineyards.

Many experiments with artificial fruit (Avery *et al.*, 1995; Puckey *et al.*, 1996; Giles and Lill, 1999) have been conducted on captive birds whose food choices were limited to the offered artificial fruit. The experiment reported here monitored a free-choice field situation. Field experiments remove the possibility that the responses are a result of stimulus deprivation and thus not representative of bird behaviour in the 'real world' (Greenberg, 1990). Here, a bird-feeding situation as close as possible to normal behaviour was established by including natural grapes at all times. The two-tier bird table design proved to be a successful design, imitating the two levels of bunches on a trellised grapevine which has two bunches of grapes per upright shoot. The two tiers doubled the number of grapes for the same close view of the birds, and possibly proved more interesting to birds so that they remained longer on camera.

There is minimal literature on the aspects of grape ripening that cue birds to begin to take grapes. The top two contenders are sugar content and colour, with the attendant confounding factor that colour may signal ripeness and therefore sugar content (Giles and Lill, 1999). Giles and Lill (1999) found that for artificial saltbush (*Rhagodia parabolica*) fruit silvereyes pecked eight times more on red fruit than on white fruit when both had equal sugar. They concluded that the birds' initial tendency to select red fruit is reinforced by learning that sugar is present. This suggests that sugar is possibly an important factor in frugivores' ongoing foraging preference, but that initial bird decisions may be influenced by colour cues, specifically the red-purple-black spectrum (Sorensen, 1981; Willson *et al.*, 1990; Stiles and White, 1982). Some authors (Moermond and Denslow, 1983; Gautier-Hion *et al.*, 1985; Puckey *et al.* 1996) suggest that the co-evolution of fruit and frugivorous birds may exert selective pressure on plants that rely primarily on avian seed-dispersers to produce red-purple-black fruit. The experiment reported here attempted to tease apart the

significance of sugar from that of colour by offering artificial grapes where all parameters were controlled and identical, and only sugar concentrations were manipulated.

Our pilot study (unpublished data) found that blackbirds preferred the artificial grapes with 40% hexose sugar even above the natural table grapes offered. This initial preference for very high sugar was thought to be due possibly to seasonal needs of the bird in winter when the thermoregulatory energy needs are high, since seasonal change is one of the many inter-related influences on mechanisms of foraging decisions (O'Connor and Shrubbs, 1986).

Among the artificial grapes in later experiments, blackbirds' preference for 40% sugar was upheld when the choice was expanded in spring to include 10% and 20% with the 5% and 40% concentrations. The conclusion is that blackbirds do indeed prefer a very high concentration of sugar in grapes, even though they are known to take grapes of much lower sugar levels in vineyards. Opportunistic frugivores that survive on a protein-dominated diet for most of the year, which is the case with blackbirds, utilize fruit quite inefficiently, with a nutrient absorption efficiency below that of nectar, seeds, vertebrate prey and arthropods (Karasov, 1990). This means that to obtain the carbohydrate that they need from grapes, the blackbird must eat more of a lower concentration of sugar than of a higher concentration. Decreased efficiency in foraging is a trade-off that can be tolerated when fruit is abundant (Jordano, 1987). Whether blackbirds in vineyards actually take more grapes of lower sugar than they would of higher sugar concentrations, were these available at the same time, remains to be investigated.

In comparison with blackbirds, silvereyes preferred a 10% concentration of hexose sugars. This was an unexpected result, as the initial hypothesis was that all birds would exhibit similar preferences for high sugar concentration. Stiles (1980) suggested that high sugar content is a mechanism by which plants attract both mammals and birds during periods of relatively high food availability such as autumn. It had been anticipated that birds foraging on grapes would conform to the co-evolutionary model that theorises that plants need to attract seed-dispersers, and that sugar is an important reward (Martinez Del Rio *et al.*, 1992; Giles and Lill, 1999).

Study of the co-evolution of frugivorous birds and fruiting plants (Herrera, 1982; Hardie and O'Brien, 1988; Martinez Del Rio *et al.*, 1992) has led to theories of trade-offs in the plants between the energy needed to produce attractive fruit and the need to attract a specific type of seed-dispersing bird. The gut passage rates (GPR) of the dispersing bird species are strongly implicated. A plant whose seed needs to be dispersed over a large distance will produce small seed that will pass through the gut of a bird and be dispersed some time later. Other plants produce a seed that is too large to pass right through the bird, and is regurgitated much sooner and closer to, or even directly beneath, the parent plant (Witmer, 1996). A similar trade-off probably exists if a seed needs to be dispersed at a certain time of year. Seeds that need to be dispersed in winter are generally presented in fruit with high lipid content to supply the needs of birds in winter conditions, while seed requiring summer dispersal comes in fruit with high liquid content, supplying water needs for birds in dry summer conditions (Herrera, 1982). Where or whether grapes fit into this pattern has not been investigated, and in any case would only be relevant with birds that coevolved with wild grapes. Silvereyes that originated in Southern Asia are unlikely to be

implicated in co-evolution with grapes, while blackbirds, originating in Europe, are linked in co-evolution with wild grape species (Hardie and O'Brien, 1988).

Witmer (1996) proposed that sugar levels might well determine the rate of passage of seed through the gut of the bird. The higher the sugar concentration, the more the fruit supplies energy needs of the bird, the slower the passage through the gut, and therefore the less fruit the bird needs. Herrera (1981) records that a blackbird takes about 6 hours to defecate when the fruit consumed is a high-sugar tropical fruit, while Worthington (1989) records the GPR of a frugivore can vary from 0.3 to 3 hours, and surmises that a fast GPR allows the bird to intake more fruit to supply the energy it needs. He notes that larger birds may have slower GPRs. He adds that it is possible that rate of ingestion is limited by rate of evacuation, if rate of assimilation is relatively constant, but if rate of assimilation varies because of fast GPR then more fruit will be needed. Conversely it is probable that a bird that feeds continuously will not require such high levels of sugar since this bird will assimilate adequate, even maximum, quantities of sugar while feeding more slowly, and rate of assimilation may well match rate of intake (Karasov, 1990). Small silvereyes may therefore not need any higher concentration than 10%, in order to match their foraging rate with their digestive assimilation rate.

Gut passage rates are a crucial part of this jigsaw. Martinez Del Rio *et al.* (1992) noted that small frugivorous passerines have poor sucrose assimilation, resulting from two mechanisms – lack of intestinal sucrase activity and fast gut passage rates. Small passerines have developed fast gut passage rates because they need to void large nutrient-poor fruit pulp and seeds since this is ballast they can ill afford to carry in flight (Martinez Del Rio *et al.*, 1992; Stanley and Lill, 2002). But fast passage rates also hinder the

assimilation of sucrose since sucrose requires hydrolysis into glucose and fructose before absorption can occur, a process that cannot happen in a short gut passage time, and which also takes energy the birds can ill afford (Stanley and Lill, 2002). It is likely that passerine pollinated plants secrete hexose-dominated nectars because their pollinators have fast GPRs (Martinez Del Rio *et al.*, 1992).

Further to this point, Karasov has investigated the absorption of glucose by small passerines (Karasov and Levey, 1990; Afik and Karasov, 1995; Caviedes-Vidal and Karasov, 1996; Afik *et al.*, 1997). His research group concluded that in many bird species glucose is absorbed paracellularly, a process that is energetically inexpensive compared to a carrier mediated absorption process. However this high intestinal permeability leaves the bird vulnerable to an indiscriminate absorption of xenobiotics (Caviedes-Vidal and Karasov, 1996), any small hydrolysed compounds among which may be low molecular weight water soluble alkaloids and simple phenolics such as tannins, which are known to have a toxic effect (Bullard and York, 1996). Finally, passive absorption of small water-soluble nutrients and the energy saved from this confers an evolutionary selective advantage, but means that the rate of intake (feeding) must match the rate of absorption. It is impossible for birds with this assimilation process to feed faster than they can absorb. This means they feed at the maximum rate possible, and that their feeding is limited by their assimilation capacity (Lepczyk and Karasov, 2000).

The behaviour of silvereyes observed in this experiment supports this analysis and leads to the thought that they may have a passive absorption digestive pattern. They preferred a lesser concentration of hexose sugars than blackbirds, whose visits were shorter compared to those of silvereyes. Blackbirds consumed whole grapes with high sugar concentrations



and were often present for some time without actually feeding, while silvereyes typically pecked continuously, preferred a lesser sugar concentration, stayed longer and sampled more grapes per visit. This behaviour, when transferred to a vineyard situation, results in extensive damage to grapes, which are often left to rot on the vine. This is in fact the experience in vineyards with silvereyes as pests (Bilney and Fisher, 2002).

## **4.6 Conclusion**

Methods of bird control at present are based at best on observation of some aspects of bird behaviour, but mostly remain broadly targeted, primitive, and most, except for chemical repellents, remain scientifically untested (Tracey and Saunders 2001). If diet drives foraging strategy (Afik and Karasov, 1995), then this knowledge will help in the targeting of individual bird species for control. Models of foraging decisions and population movements need to deduce the function of relevant behavioural traits (Sherry, 1990).

Comparing elements of the same order of complexity is a sound method advocated by Tinbergen (1951). Sherry (1990) divides the study of fruit depredation by birds into two approaches. The first is the ecological (or 'tactical') study of environmental effects on bird choices (encompassing patch models, learning, stochastic variation and fruit characteristics that influence bird selection of fruit), and the evolutionary (or 'strategic') study of internal constraints on choice (the phenotype may have been shaped by ecological circumstances over evolutionary time). He sees the comparative method as a 'strategic' tool where comparisons of the phenotype's characteristics of physiology and behaviour can explain ecological differences of feeding behaviour and diet.

Greenberg (1990) comments that most attempts to model the responses of birds have assumed that bird species all sample and track resources in an equivalent manner, without conceding that there are differences between species. The key finding of this experiment is that these two bird species have different preferences for sugar levels. The results from this experiment point to a possible connection between the preferred sugar concentration, the birds' physiology, the type of nutrition they need, the way each species digests, and the time spent feeding, which ultimately may explain the difference in type of damage inflicted by the two species on grapes. There are potentially other major differences between species, even those using the same food resource.

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## **Chapter five<sup>1</sup>**

### **Response of blackbirds and silvereyes to some grape aromas.**

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**Keywords:** 2-3-isobutylmethoxypyrazine, geraniol, bird damage, grapes

#### **5.1 Abstract**

Grape aromas might be a cue to foraging birds in vineyards, so two typical grape aromas were offered in a controlled experimental situation to free-ranging blackbirds and silvereyes. Bird visits to a bird table with small nectar pots surrounded by a wick, one soaked in geraniol, one in 2-3-isobutylmethoxypyrazine (IBMP), and one an aromaless control, were recorded and analysed. Blackbirds stayed longer and drank more from the geraniol nectar than from the IBMP nectar, and showed least interest in the control. Silvereyes visited IBMP most but drank more and stayed longer at the control. It appears from these results that aroma may cue some bird species to attack grapes.

#### **5.2 Introduction**

Anecdotal evidence from vineyard managers suggest that birds prefer certain grape cultivars to others, but confounding parameters such as stage of ripening, choice of varieties within the vineyard, and proximity to bird cover are difficult to unravel. There is

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still debate on the relative importance of olfaction (the sense of smell) in bird behaviour. Some species do use the sense of smell, for example, kiwis (Guilford *et al.*, 1987), starlings (Avery and Nelms, 1990), and homing pigeons (Wallraff and Andreae 1998). However it appears that, compared to sight and hearing, olfaction may play a relatively small role in a bird's location of food (King and McLelland, 1984; Avery and Nelms, 1990; Wager-Page and Mason, 1996). As grapes ripen their aroma profile changes significantly. Geraniol is a terpene, characteristic of aromatic white cultivars such as Muscat or Riesling, Müller Thurgau and Traminer. Terpenes, which require hydrolysis and warmth to become volatile (Hardie and O'Brien, 1988), are absent in unripe grapes (Marais, 1983) and are therefore an indication of ripeness. 2-3-isobutylmethoxypyrazine is a green pepper/grassy aroma, with a particularly low detection threshold level for humans (2 parts per trillion, Lacey *et al.*, 1991). It is present in Cabernet Sauvignon, Sauvignon blanc, and other cultivars, and is more highly concentrated in grapes that are shaded or that are at an earlier stage of ripening (Allen and Lacey, 1997). In New Zealand's Marlborough Sauvignon blanc wine, it is perceived as distinctive and attractive, but in Cabernet Sauvignon it can contribute to greenness which is perceived as undesirable (Hashizume and Umeda, 1996; Allen and Lacey, 1997). In both cultivars it reduces in concentration as the grapes ripen (Allen and Lacey, 1997). Pyrazines occur in nature as odours associated with aposematic (a warning signal, usually colour or odour, thought to be produced by some insects as a defence against predators) effects, and several authors have tested conditions under which they exert a deterrent effect on birds (Guilford *et al.*, 1987, Avery and Nelms, 1990; Hartley, 1999). In order to discover whether grape aroma alone could be a cue to birds, an experimental method was developed and used to separate and control grape ripening variables (see Chapter 3) and to record the behavioural responses of blackbirds and silvereyes to two grape aromas. Blackbirds are resident species and present in the vineyard

throughout the year, while silvereyes arrive as the grapes ripen. The goals of the experiment reported here were to determine whether aroma would influence bird behaviour, whether each species showed preference or avoidance behaviour with either aroma, and which aroma each bird species preferred. Both number of bird visits and length of stay at each aroma were recorded. Hosler and Smith (2000) suggested that length of reaction is a more sensitive measure of aroma detection than frequency.

### **5.3 Materials and methods**

Artificial nectar was made by dissolving 15% glucose and fructose in equal parts in near boiling water, cooling it and presenting it to birds in identical bright red plastic cups 2 cm in diameter and 1 cm deep. The red pots fitted snugly into slightly larger green plastic caps, sitting inside an internal rim. This rim and the edge of the larger cap formed a shallow channel 2 mm wide, into which was pressed a small 'wick' of soft tissue paper. There four pots and caps on each of three tables, one table for each aroma and the control.

Geraniol was used undiluted, in the concentration supplied by the manufacturer (BRI FCC, #710, molecular formula  $C_{10}H_{18}O$ , 98% pure, Bedoukian Research Inc., Danbury, CT.).

2-3-isobutylmethoxypyrazine (IBMP, molecular formula  $C_9H_{14}N_2O$ , supplied by Rob Sherlock, Lincoln University) was used at a concentration of 1.6% (0.16 mg/L).

Blackbirds were present in the Lincoln University Vineyard (Canterbury, New Zealand) but silvereyes were not, this species arriving much later towards harvest. Blackbird experiments were conducted in the Lincoln University Vineyard and silvereyes in a pear orchard 4km away, where some blackbirds were also present. Silvereyes will readily feed on nectar, but blackbirds normally do not. In the dry summer conditions of this experiment

(3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup>, 7<sup>th</sup>, 16<sup>th</sup> and 17<sup>th</sup> January 2003) they did, thus enabling a direct comparison for experimental purposes.

Pots and caps were placed on three identical bird feeder tables. Each table was two tiered, each tier 50 cm long and 10 cm wide, the upper offset above the lower so the birds had good views of both tiers (see Chapter 3), with two pots on each tier, four pots in total for each table. The table was 1.5 m aboveground. The tables were placed beside each other three metres apart and all the same distance (10 m) from cover. The recording of bird visits was synchronized on three identical video cameras positioned 1.5m in front of the feeder tables.

Video recordings were made as described in Chapter 3. Sessions were run for three hours from 8am to 11am and from 11am to 2pm each day. For each session, pots were filled first with artificial nectar, and then four drops of the aroma compound were placed onto the wick around the pots. Detection thresholds of the aroma compounds being unknown, the relative strength of the two aromas was judged by human nose to be more or less similar at one metre from the tables, but not detectable at two metres. All the data were collected in dry, sunny conditions, with temperatures 19 to 25°C (70 to 80°F), and relatively still, with windrun less than 300 km (160 miles) per day, which is 12kph (7mph) mean wind speed. Position of the tables relative to each other was rotated to control for any edge effect or effect of proximity to possible nest or other favoured cover of the birds.

Data for all three tables were collected simultaneously. As the birds were unmarked the unit of measure was one bird visit. The number and length of each bird visit, and the number of pecks each bird delivered to each pot were recorded. Non-parametric statistical



tests were used. Kruskal-Wallis nonparametric ANOVA (H) was run to discover if response to the aroma tables differed from that to the aromaless control. Then each aroma was analysed against the control, and against the other aroma, with Mann-Whitney U test (U) to discover any significant preferences. The mean length of each bird visit and number of pecks delivered to the nectar pots within the aroma surround were similarly analysed (GenStat 6).

## 5.4 Results and Discussion

For blackbirds there was no significant difference in the number of visits paid to each table (Table 5.1,  $H=2.844$ , d.f. = 2,  $p=0.2$ ). Analysis of geraniol vs. control was also not statistically significant ( $U = 11.5$ ,  $p = 0.09$ ), but there was a tendency for the control to be less visited. There was a significant difference in the length of blackbird visits ( $H = 6.524$ , d.f. = 2,  $p = 0.03$ ) with geraniol and IBMP both significantly preferred to the control ( $U = 55.5$ ,  $p < 0.001$ ;  $U = 89.0$ ,  $p = 0.007$  respectively). There was also a significant difference

**Table 5.1: Blackbird visits:**

	Total # visits	#Visits per day	Length (seconds)	# Pecks
		Mean (median, SE)	Mean (median, SE)	Mean (median, SE)
Geraniol	36	5.1 (4, 0.96)	41.3 (38, 4.97)	9.8 (8, 1.2)
Control	19	2.7 (2, 0.94)	22.8 (22, 3.10)	6.1 (7, 1.1)
IBMP	29	4.1 (4, 1.12)	33.4 (32, 3.34)	8.0 (8, 1.0)

**Table 5.2: Silvereve visits**

	Total # visits	#Visits per day	Length (seconds)	# Pecks
		Mean (median, SE)	Mean (median, SE)	Mean (median, SE)
Geraniol	91	16.4 (15, 3.30)	23.0 (19, 1.89)	5.9 (5, 0.5)
Control	117	22.2 (20, 4.73)	23.7 (20, 1.64)	7.6 (6, 0.6)
IBMP	151	30.2 (30, 5.20)	18.7 (16, 1.22)	4.9 (4, 0.4)

Blackbirds were recorded in January 2003 in the Lincoln University Vineyard, Canterbury, New Zealand. Blackbirds and silvereys were also recorded in January 2003 in a pear orchard 4 km from the vineyard. Video recordings were viewed and data collated were: number of bird visits, time of arrival and departure from the table for each bird visit, number of pecks at each pot.

in the number of pecks ( $H = 9.27$ ,  $d.f. = 2$ ,  $p = 0.01$ ) with both Geraniol and IBMP significantly preferred to the control ( $U = 82.5$ ,  $p = 0.003$ ;  $U = 11$ ,  $p = 0.04$  respectively).

For silvereyes there were no significant differences in number of visits to the three tables (Table 5.2,  $H = 4.02$ ,  $d.f. = 2$ ,  $p = 0.1$ ). However, IBMP was significantly preferred to geraniol for number of visits ( $U = 3.0$ ,  $p = 0.05$ ). There was a significant difference in the length of time spent at each aroma ( $H = 7.09$ ,  $d.f. = 2$ ,  $p = 0.02$ ). For length of time there was no significant difference between geraniol and control, but the control was significantly preferred to IBMP ( $U = 2583.0$ ,  $p = 0.01$ ). The number of pecks differed significantly ( $H = 12.73$ ,  $d.f. = 2$ ,  $p = 0.002$ ). The control was pecked significantly more than either geraniol ( $U = 82.5$ ,  $p = 0.003$ ) or IBMP ( $U = 110.0$ ,  $p = 0.04$ ). There was no significant difference in pecking between the aromas.

Preference for both aromas over the control appeared clear for blackbirds, but whereas silvereyes visited IBMP more often, their visits to IBMP were shorter than to the other two tables; they stayed longer and pecked more at the control than either aroma.

Blackbirds are residential, spending the whole year on their 'patch'. Those that have patches in vineyards are likely to experience grapes at all stages of ripening, and if they can detect aromas, may be able to associate certain aromas with attendant stages of ripening. Our experimental results indicate that blackbirds can detect grape aromas and that it is possible that they have learnt to associate ripeness parameters with aroma. The preference for geraniol observed in this study suggests that the reward of riper fruit and therefore of greater sugar reward (Giles and Lill, 1999; Chapter 4) may be associated with this aroma.

Silvereyes are mobile and are often not present in vineyards until a later stage of ripening (Saxton, unpublished). This may not be due to any factor in the grape ripening process but to extraneous environmental conditions such as cold or drought elsewhere forcing them to move around to find food resources. If they can detect odour, it is likely that they would not have succeeded in relating any specific aroma to stages in grape ripening. Our experiments have shown no consistency in aroma preference, and even suggest that aroma is less preferred than no aroma. However another interpretation of the results is that for silvereyes, as for humans, IBMP is attractive at low concentrations (from a distance) but aversive at the strength encountered on the bird table, which could explain why the high number of visits to IBMP is at variance with length of stay or number of pecks.

## **5.5 Conclusion**

The significance of these results for vineyard managers is that grapes with strong aromas may be under strong pressure of attack from blackbirds. Further evidence on silvereyes (Saxton, unpublished) indicates that grapes alone do not attract silvereyes, but that they form part of a matrix of environmental factors governing silvereye flock movements, details of which remain unclear. However, in New Zealand where the development of vineyards with high value crops is increasing, understanding the cues that draw damaging silvereyes to vineyards is critical, if managers are to maximize control strategies.

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## **Chapter six**

# **Behavioural responses of two species of birds to varying levels of tartaric and malic acids in artificial grapes.**

### **6.1 Abstract**

Diminishing acid concentrations are popularly thought to be one of the effects of ripening grapes that leads to increased bird pressure towards harvest. Blackbirds and silvereyes were offered varying concentrations of tartaric and malic acids in artificial grapes in a field context, where all other ripening grape parameters, such as colour and sugar, were controlled. Results were mixed and inconsistent for both species, which indicated that other factors are more important than diminishing acid concentrations in increasing bird pressure towards harvest.

### **6.2 Introduction**

Anecdotal evidence from vineyards that later ripening grape varieties suffer less damage from birds than earlier ripening varieties nearby, has led to the hypothesis that high acid content in grapes are not palatable to birds and that this is an important reason why birds do not attack high acid grapes (Boudreau, 1972). There is little robust data to support this since most research on frugivory in birds has explored colour and sugar in fruit pulp as avian attractants, or the effects of seed load or ingestion. As part of a larger investigation into cues that attract birds to grapes (see chapter one) the experiments reported here were designed to examine the role of acid as a deterrent to avian foraging.

Organic acids in plants serve various physiological functions in the photosynthetic process known as the Calvin cycle (Hunter *et al.*, 1991; Beriashvili and Beriashvili, 1996; Cheffings *et al.*, 1997). Organic acids contribute to pH, which are implicated in reactions such as enhancing aroma (Hunter *et al.*, 1995) or deepening colour. The sourness of acid deters mammals, but the effect on birds is unclear (Mason and Clark, 2000). The possibility that the sweetness of sugar counteracts or is even enhanced by acid (Boudreau, 1972) must be considered.

Malic and tartaric acids are 90% of total organic acids in grapes (Coombe, 1992; Beriashvili and Beriashvili, 1996), with 80% being tartaric acid (Lavee and Nir, 1986; Hunter *et al.*, 1991), and smaller amounts of malic, p-coumaric, and other acids.

Concentration of tartaric acid reduces due to enlargement of the grape (Coombe, 1992) and some leakage through breakdown of tonoplast (Terrier *et al.*, 2001), while malic acid is rapidly reduced through metabolism in the Calvin and Krebs cycles of the photosynthetic process (Doneche *et al.*, 1985), more so in a warmer climate or a warmer season (Lavee and Nir, 1986). Before colour change (*véraison*) grape acids can register as high as 40g/L, but at harvest are reduced to below 10g/L. In the course of ripening glucose and fructose concentrations in the grape rise while acid concentrations concurrently fall. In previous experiments (chapter four) it was established that sugar was an important cue to frugivorous birds in the vineyard, and that preferred concentrations of sugar varied between species, which agreed with Boudreau (1972).

Although tartaric acid is the main acid in grapes, its lack of implication in metabolic processes (Ruffner, 1982), and, more importantly, its lack of significant reduction during ripening (Robredo *et al.*, 1991) suggest that it might not be significant as a cue to birds of

fruit ripeness. Ruffner (1982) found no function for tartaric acid in the plant, although he suggested it may scavenge calcium. The possibility that reducing malic acid (Doneche *et al.*, 1985; Lavee and Nir, 1986; Coombe, 1992; Robredo *et al.*, 1991; Terrier *et al.*, 2001), which correlates well with falling titratable acidity in ripening grapes (Barbeau *et al.*, 1998) might be an important factor was explored by conducting experiments using both acids.

In order to detect a threshold at which acids no longer deter birds, varying concentrations of tartaric acid, which constitutes the largest fraction of acid in grapes, or malic acid, which reduces in a metabolic process during ripening, were offered to blackbirds and silvereyes in a field situation where birds had free choice. Other ripening parameters such as sugar and colour were controlled for by using an artificial grape. The artificial grape contained controlled concentrations of sugar, in an attempt to discover at what level rising sugar drives the acid 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.

### **6.3 Materials and methods**

The sites used were at Lincoln University vineyard (blackbirds) and a pear orchard some 4km from the vineyard (silvereyes). The artificial grapes were offered on a bird feeder table positioned approximately 8m from trees that the birds appeared to use for cover. The bird feeder table was 1.5m from the ground, with two levels, the top one 30cm above the lower, and offset so that birds on either level could see the other. The table was 50cm wide, each level was a board 15cm wide (see chapter three for methods).

Individual grapes were set out, 10 on the top and 10 on the lower levels in a latin square design so that no two adjacent grapes were the same, and birds videoed for three hours in the morning, starting shortly after sunrise. A Burlie TC395X camera, and a JVC TK-5240 camera were used with Cosmicar/Pentax TS6ZME-5 6.3-38mm lenses, set up approximately 1.5m from and focused on the bird feeder table. A Panasonic Time-lapse VCR recorded bird feeding at a slow speed approximating 7-8 frames per second.

Artificial grapes of 5-6mm diameter and coloured purple (chapter four) were made with hexose sugars (50% each D-glucose and L-fructose). Experiments were run with grapes of sugar concentrations of 10%, 15%, 20% and 25% for blackbirds, and 5%, 10% and 15% for silvereyes, with 0g/L, 10g/L, 15g/L and 20g/L tartaric or malic acid added to the mixture. Sugar was added because the birds would not take artificial grapes with no sugar. In earlier experiments blackbirds had exhibited preference for a higher sugar concentration (>20%) than silvereyes (10-15%, chapter four). The sugar concentrations reflected these preferences to maximise the effect of sugar may have on responses to acid, and to minimise any interspecific confounding effect of sugar preferences in order to gain comparable results between species.

Video footage was viewed and bird behaviour recorded. Data included how long the bird was on the table, and for blackbirds on each bird visit which grapes were eaten or taken away (accepted), or dropped after handling (rejected). Accepted grapes were analysed as a percentage of total grapes handled. Grapes that were not handled are not included in the dataset. 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 and this was recorded as another bird visit. Data



were analysed by non-parametric Kruskal-Wallis Anova (H) and Mann-Whitney U test (U) (Genstat 6 software).

## 6.4 Results

### Blackbirds

At 10% hexose sugar concentration there was a significant preference for no acid in the tartaric acid experiments (Table 6.1,  $H=5.91$ ,  $p=0.08$ ,  $n=109$ ), where 0g/L was significantly preferred to 10g/L and all other tartaric acid levels ( $U=76.5$ ,  $p=0.05$ ,  $n=46$ ), but there were no significant preferences shown with malic acid. At 15% sugar and tartaric acid there was a significant preference shown ( $H=6.1$ ,  $p=0.05$ ), where 20g/L was preferred to 15g/L ( $U=7.5$ ,  $p=0.09$ ,  $n=45$ ) and all other levels. At 15% sugar with malic acid a higher concentration of malic acid was offered (0g/L, 10g/L, 20g/L and 40g/L) to see if the higher concentration would indicate a threshold, but results were not significant. At 20% sugar there was no significant preference for any acid concentration. At 25% sugar a significant preference was shown ( $H=3.92$ ,  $p=0.09$ ,  $n=111$ ), where 20g/L tartaric acid was significantly preferred to 10g/L ( $U=0.0$ ,  $p=0.008$ ,  $n=32$ ). No significant preference was found between concentrations of malic acid.

There was a significant preference for high sugar over all acid treatments (Table 6.1  $H = 12.4$ ,  $p= 0.001$ ,  $n = 56$ ) with both 25% and 20% preferred to 10% ( $U=1153$ ,  $p=0.016$ , and  $U=1140$ ,  $p=0.013$  respectively). 25% and 20% were also significantly preferred to 15% ( $U=1136.5$ ,  $p=0.012$  and  $U=1103$ ,  $p=0.007$  respectively).

**Table 6.1: Blackbird responses to tartaric and malic acid concentration in artificial grapes:(percentage grapes taken, N= session, n=bird visits)**

10% sugar	0g/L Mean (SE)	10g/L Mean (SE)	15g/L Mean (SE)	20g/L Mean (SE)	40g/L	Total	mean percentage grapes taken
Tartaric(N=16, n=63)	90.6(3.6)*	68.7(7.9)	71.9(7.3)	73.9(7.6)		305.1	76.2
Malic (N=9, n=46)	87.0(5.2)	63.9(11.9)	77.9(11.3)	87.9(4.8)		316.7	79.1
<u>15% sugar</u>							
Tartaric(N=7, n=45)	95.8(4.7)	81.9(8.7)	54.1(13.5)	88.9(11.1)*		320.7	80.2
Malic (N=3, n=20)	38.6(19.8)	36.0(21.7)		58.0(12.7)	88.6(11.3)	221.2	55.3
<u>20% sugar</u>							
Tartaric (N=6, n=26)	100.0(0)	81.0(10.3)	83.3(7.45)	80.9(14.3)		345.2	86.3}*}
Malic (N=7, n=61)	92.9(4.6)	100.0(0)	85.7(6.7)	92.8(4.6)		371.4	92.8}
<u>25% sugar</u>							
Tartaric (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 (N=10, n=7)	97.22(2.7)	93.5(4.34)	94.4(4.6)	97.2(2.7)		382.1	95.5
Total	680.4	979.8	537.3	679.6	88.6		
Mean percentage taken	85.0	72.47	67.1	84.95	88.6		

\* indicates a significant preference

**Table 6.2: Silvereye responses to tartaric and malic acid concentration in artificial grapes:(mean pecks per visit, N= sessions, n=bird visits)**

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

\* indicates a significant preference

### **Silvereyes (Table 6.2)**

With silvereyes no significant differences were shown at any level of acid concentration, and no preferences were detectable.

Over all acid concentrations, a significant preference between the sugar levels was shown (Table 6.2,  $H=30.16$ ,  $p=0.001$ ,  $n=34$ ) and this was for 10% sugar over 5% ( $U=174.5$ ,  $p=0.001$ ) and for 15% over 5% ( $U=212.1$ ,  $p=0.001$ ). Between 10% and 15% there was no significant preference.

The means showed a preference for the previously demonstrated sugar levels (see chapter four) of 20% and above for blackbirds and 10-15% for silvereyes. Overall pooled results showed no significant differences in any acid levels for either species.

## **6.5 Discussion**

Results from these experiments were inconclusive with regards to the response of birds to acids, but some results appeared to confirm the results from previous sugar experiments (chapter four). The acid concentrations used were similar to those found in ripening grapes from the period of *véraison* onwards, down to about 10g/L by harvest. (Depending on cultivar and season it may reduce to 9g/L or even 8g/L). This time shortly before harvest is the time of greatest bird depredation on grapes (Boudreau, 1972). Acid concentrations higher than 20g/L are recorded in unripe pre-*véraison* grapes, but bird depredation at this stage is minimal.

To test the possibility that a higher concentration would indicate a threshold between 20g/l and 40g/l, one experiment offered higher concentrations (40g/L. Table 1) of malic acid at 15% sugar concentration to blackbirds, but again without any significant result. It is possible that a threshold of acid as a deterrent may exist and be higher than 40g/L. But this scenario would not apply to grapes at any stage of ripening.

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 even preferring water with acid to tapwater. This tolerance may be due to lack of physiological mechanisms (birds have fewer taste buds than mammals, a pigeon has less than 50, King and McLelland, 1984, while humans have 9000 in total on the tongue and another 2000 inside the mouth, Bartoshuk, L. pers comm. 2004, and see p 43 in Mason and Clark, 2000). Other factors in avian foraging, such as vision (particularly important in birds) or context of the food source may override taste as a cue to nutritional value (Mason and Clark, 2000). High acid in most fruit is repellent to mammals, but there is little evidence for this in 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 since 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). 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, is not upheld, and the results obtained here appear to be either purely random, or possibly indicate a much more complex process than was detectable in this experiment. It is

possible that acid does contribute in an indirect way to avian perception of grapes. There could be an interaction of acid with sugar, or an interaction through pH with other compounds found in grapes, or an enhancement of taste perception generally, none of which were detectable by the experimental procedure used here.

The experiments with higher sugar concentrations resulted in significantly higher levels of interest by the birds. The birds displayed preference for sugar levels that concurred with results from previous experiments that manipulated hexose sugar levels, 20% for blackbirds and 10-15% for silvereyes (chapter four).

Other experiments in this project have shown significant results for other compounds in grapes, such as secondary metabolites (chapter seven), which may be related to avian physiological and seasonal nutritional needs. Cues to these compounds (for instance purple colour maybe cueing blackbirds to secondary metabolites in winter) appeared highly significant. Such cues may well override completely any cues that acid perception may offer. High acid in fruit is often associated with green colour, but the colour experiments in chapter six also indicated that in summer green was not significantly avoided by either avian species when sugar levels were at an acceptable level (15%). It appears possible that neither acid nor green colour are particularly important to birds in summer, or as a cue to nutritional value or otherwise of grapes at early ripening stages.

If acids are neither perceived nor nutritionally valuable to birds then the reason that cultivars such as Riesling are not attacked preferentially by birds must lie elsewhere.

Riesling grapes that are late ripening accumulate sugars more slowly than other cultivars in New Zealand. They are also green at a stage when blackbirds may be looking for purple

grapes (chapter seven). Riesling grapes have thicker skins than other cultivars, which may be a deterrent to silvereyes that feed by pecking through the skin to reach the pulp.

Cabernet Sauvignon is another grape variety that ripens later in New Zealand, and is also not attacked so much. Reasons for this may lie in slower sugar accumulation, but may be more complex. In a very late ripening season in Canterbury, New Zealand, in 2004, Cabernet Sauvignon grapes that were left till late May were heavily attacked by blackbirds (Saxton, pers, observation), even though ripe Gewurztraminer grapes of green-pinkish colour and higher sugar levels were available close by (indicating that choices were not governed by lack of alternative fruit). Secondary metabolites possibly become significant cues to blackbirds in late autumn (see chapter seven).

It is possible that the artificial grapes used in these experiments masked the deterrent in the same way that berry shape was thought to mask quebracho (tannin from bark of the quebracho tree *Aspidosperma quebracho-blanco*) to silvereyes while the same birds detected the quebracho in a mash (Stanley and Lill, 2001). The authors suggested that this may be an ecological strategy of plants to persuade birds to take fruit without detecting high tannin seeds. In the context of a foraging environment the shape of the fruit may override other perceptions.

Many sessions were run in this group of experiments, in an effort to detect thresholds, and some of the datasets were small. The sampling issue of bird numbers was thought about several times. Some of the blackbird datasets may lack robustness due to possible sampling of few birds. Four separate locations were used both at the Lincoln University Vineyard and in the orchard, to maximise the number of individuals recorded, but the birds were not marked, principally because no method could be devised to mark them without catching

them (a process that required training, licensing, approval of an ethics committee and time). Blackbirds were identifiable up to a point from the clear video picture as adult males, females, and young birds. Most datasets had at least one of each of these at each location. Silvereyes were often present several at a time and there were clearly small flocks of about 6-12 birds at a time. Bird visits are used for samples size, one reason for this being that in a vineyard situation grapes can sustain multiple attacks from the same bird and it is the length and severity of the attack on grapes that is the principal point of interest and therefore unit of measure used for this work.

The failure of birds to show significant preferences in these experiments does not preclude the possibility that acids contribute to their nutrition, or that acids play a part in the seasonal interactions of fruit and foraging species. A lack of scientific literature on acid and avian foraging may reflect failure thus far to detect what these effects or interactions might be. However, for practical purposes of understanding the attraction of grapes for birds in vineyards, it appears that acid at levels founding ripening grapes is not an important cue.

## **6.6 Conclusion**

This set of experiments serves at least to cast considerable doubt on the theory that reducing acids in grapes renders them more attractive to birds. This concurs with anecdotal perceptions of blackbirds eating wholly immature pre véraison grapes, with undoubtedly high acidity. In this experiment sugar clearly overrode acid, with higher sugar concentrations being preferred to lower ones for both species. Cues such as sugar, colour and aroma appear to override any effect that acid may have on avian perception of grapes as a food source.



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## **Chapter seven**

### **Comparative behavioural responses of two species of birds to colour and other secondary metabolites in grapes.**

#### **7.1 Abstract**

Secondary metabolites in fruit, which include colour and tannin, are part of a complex matrix of compounds that have dietary significance for birds. To determine whether secondary metabolites in grapes might be important cues to two avian species that feed on wine grapes, tannin concentrations were offered to blackbirds and silvereyes in a controlled field experiment. Behavioural responses differed between the species.

Blackbirds appeared to detect tannin at lower concentrations than silvereyes, and both species showed aversion at concentrations higher than those normally found in ripening grape skins but lower than those in grape seeds. In order to discover any colour preferences of the same two bird species, green and purple artificial grapes were offered in summer and again in winter. Responses to colour in winter differed significantly between the species. In winter blackbirds took only purple berries while silvereyes pecked mostly at green. In contrast, no colour preferences were shown by either species during the summer months.

Colour may cue the two species to different aspects of fruit nutritive value. In the matrix of fruit cues to avian foragers, it appears that colour may be a species specific signal to secondary metabolites, and to the nutritional value or otherwise of that fruit to that species.

#### **7.2 Introduction**

The dietary interactions between plants and those organisms that eat them are complex and even paradoxical (Stanley and Lill, 2001). Plant secondary metabolites are a large group of

varied compounds so called because they do not contribute to the primary metabolism of plants (Cipollini and Levey, 1997), and the energy expended on their production in plants and fruit is at times not self-evident. The many different compounds involved seem to serve different functions, and are perhaps part of an overarching strategy to collectively manipulate, or benefit from, the foraging behaviour of seed dispersers (Mack, 1990, 2000; Hovestadt, 2003). The energy expended by plants, for example to produce colour, is not insignificant (Herrera, 1982; Willson *et al.*, 1990). It seems that secondary metabolites are part of the plant's interaction with foraging species, be it directly through colour to attract seed or pollen dispersers (Izhaki and Safriel, 1989; Cipollini and Levey, 1997; Stanley and Lill 2001), or perhaps a broader function of manipulation of foragers' diet (Izhaki and Safriel, 1989; Murphy, 1994).

Secondary metabolites are involved in plant defence mechanisms against fungal, parasite, insect or vertebrate attack (Butler, 1992; Mack, 2000). It has been suggested that secondary metabolites might contribute to seed viability by controlling light penetration, preventing them from germinating too soon, and also by causing rapid evacuation from the gut of an avian forager thus preserving seed viability (for review see Cipollini and Levey, 1997). In addition to actual function, the role of secondary metabolites in direct foraging cues such as colour and aroma is complex and remains relatively unclear. Flavonoids are one group of secondary metabolites that are sometimes molecularly bound to sugar in the plant (King and Young, 1999), and contribute to colour and to aroma (Winkel-Shirley, 2001). These are primary cues to foraging species (Cipollini and Levey, 1997). Indirect cues to the forager in the form of feedback from the gut, or from inhibition of nutritional gains (Butler, 1992), further complicate the issue. Whereas previous knowledge accepted that tannins generally reduce nutritional value and that high tannin seeds are avoided by frugivores,

recent research showing that some secondary metabolites increase the nutritive value of fruit to birds (Bairlein, 2002) creates a paradox that illustrates how imperfect is the scientific understanding of these compounds in the fruit-frugivore interaction.

Secondary metabolites in plants comprise several groups of complex and relatively unstudied compounds including alkaloids, terpenoids, saponins, and phenolics such as tannins and lignins (Cipollini and Levey, 1997). Flavonoids represent a large group (over 2000, Zoecklein, *et al.*, 1989) of low molecular weight compounds (Butler, 1992; King and Young, 1999). The main groups of flavonoids are flavonols, flavanones, flavones, anthocyanidins, and procyanidins, the latter also called flavan-3-ols or catechins (Gawel, 1998; Kennedy *et al.*, 2000; Harberston *et al.*, 2002), and include chalcones, flavan-diols, anthocyanins and condensed tannins (Winkel-Shirley, 2001). This study focuses on tannins and anthocyanins as two such compounds involved in avian cues.

Catechins (flavan-3-ols or procyanidins) are small molecular weight compounds that are sometimes called monomeric or hydrolysable tannins. These can make longer chains that are called polymeric or condensed tannins. Condensed tannins are also called proanthocyanidins (Butler 1982, 1992; Souquet *et al.*, 1996; Gawel, 1998), or polymeric flavan-3-ols (Harbertson *et al.*, 2002). The two main groups of tannins are the hydrolysable short-chain tannins and polymeric condensed tannins. Both groups have received much interest due to their detrimental effects on nutrition of mammals that ingest them (Zucker, 1983; Butler, 1992; Bullard and York, 1996; Cipollini and Levey, 1997; Gawel, 1998). These effects are attributed to binding properties with proteins (Butler, 1992) leading to enzyme inhibition, and to neutralisation of amino acids in digestive processes.

Tannins are among the most widespread secondary metabolites in ripe fruit (Jordano, 1987; Stanley and Lill, 2001). Short monomeric tannins tend to be bitter, while longer chain condensed tannins tend to be less bitter and more astringent (Brossaud *et al.*, 2001). Birds ingest seeds without suffering noticeable digestive inhibition, which has led to the conclusion that they have, in addition to strategies such as immediate regurgitation and/or rapid gut evacuation, a mechanism for dealing with, or tolerating, the tannins that are present in seeds (Stanley and Lill, 2001). Seed eating birds gain their nutrition from digesting seeds, and for seed-eating specialist birds tannins are not toxic. The liver is responsible for detoxification, and more detoxification needs a larger liver. The seed eating parrot crossbill (*Loxia pytyopsittacus*), for example, has a liver of only 1.4% body mass, because seed tannins are not toxic for this species. The silvereye's liver is 5.4% of body mass (Stanley and Lill, 2001), which suggests that they have to deal with substances toxic to them. Their tolerance of tannin may be limited.

Bairlein has found that birds can benefit significantly from nutritive secondary metabolites of black elder, but not from red elder (Bairlein, F. pers.comm. 04 June 2003). So far the exact difference between the black and red elder that is responsible for the difference in nutritional benefits remains unclear. But a clear preference of birds for black fruit (Willson *et al.*, 1990) may be explained by this difference in nutritional assimilation (Bairlein, 2002). Research into the role of tannins in avian digestion (Butler, 1982; Izhaki and Safriel, 1989; Bullard and York, 1996; Witmer, 1998; Dearing *et al.*, 2001; Stanley and Lill, 2001) has generally not attempted to elucidate how birds detect tannins, whether this is by taste (bitterness) or tactile (astringency) cues. Distinguishing clearly between bitterness and astringency is a process that is at present not possible due to lack of pure standard material (Zywicki, 2003). It remains unclear whether or how birds are cued to

tannins and other secondary metabolites, and how they distinguish between possible toxicity of tannins and nutritional benefits of other secondary metabolites.

Anthocyanins are responsible for the red-purple-black fruit and flower colouring. There is a large body of work exploring the relative attraction of different colours for frugivorous birds, with red-purple-black generally considered the most attractive colours, while green is considered least attractive. Anthocyanins may express colour mainly through glycosylation, acylation or methylation like other secondary metabolites. Increasing pH tends to deepen colours towards blue. It is generally accepted that colour indicates ripeness of fruit and, therefore, its sugar content and energy supply (Sorensen, 1981; Morden-Moore and Willson, 1982; Willson and Thompson, 1982; Feare, 1984; McPherson, 1988; Willson *et al.*, 1990; Giles and Lill., 1999; Stanley *et al.*, 2002). Many authors have investigated the role of colour in the attraction of fruit for avian dispersers of seeds (Sorensen, 1981; Morden-Moore and Willson, 1982; Willson and Thompson, 1982; Willson and Melampy, 1983; McPherson, 1988; Willson *et al.*, 1990; McKean, 1990; Fuentes, 1995a,b; Puckey *et al.*, 1996; Giles and Lill, 1999, Stanley *et al.*, 2002). Conversely, in the scenario of fruit-frugivore evolution, avian foraging behaviour may have exerted a selective pressure on plants regarding fruit size (Sorensen, 1981), fruit accessibility (Fuentes, 1995a), fruit abundance (Giles and Lill, 1999), seed size (Herrera, 1981; Wheelwright, 1985; Stanley *et al.*, 2002). Colour is a very important part of the matrix of cues that are plant strategies for effective seed dispersal.

There are many avian species that forage on fruit, and the meaning of the colour cues are not always consistent between species or even in individuals, since ontogenetic learning, and even simply age (Greenberg and Mettke-Hofmann, 2001) can alter behavioural

responses. Studies have found a preference among frugivorous species for red-purple-black fruits (Sorensen, 1981; Giles and Lill, 1999), and there is evidence that frugivorous bird species dislike green fruit (McPherson, 1988; Willson *et al.*, 1990). However, changes or inconsistencies in colour preferences have been frequently reported. Stanley *et al.* (2002) found that doubling the sugar reward in green artificial fruit resulted in silvereyes preferring green to red or white fruit with less sugar, while Avery and Mason (1997) found that learned responses to colour cues from poisonous baits were not maintained for long.

Grapes are the most economically significant fruit crop worldwide, and are subject to severe predation by birds in some areas. As part of a wider investigation into the attraction of grapes for frugivorous birds avian behavioural responses to various compounds that occur in ripening grapes were tested to see which compounds might influence bird behaviour. Colour and tannins were investigated in the experiment reported here, and experiments were also conducted with hexose sugars (chapter four), aromas (chapter five) and acids (chapter six). Blackbirds and silvereyes are both vineyard pest species in New Zealand and the experiments here reported tested the behavioural responses of these two species Greater knowledge about what is important to birds (and why) might contribute to the evolution of an ecological strategy to deter or inhibit bird pressure on grapes particularly, but also on other fruit and crops generally.

Colour change (*véraison*) begins in grapes when the seed attains maturity. At this stage grape pulp acid levels are high and sugar levels are still relatively low (Coombe, 1992; Gray *et al.*, 1997). From *véraison* on, different cultivars of wine grapes vary in colour from green through grey-pink to dark red and purple. Evidence from vineyards is that both species attack all colours, but is probably confounded by possible learning experiences of

the birds as they sample grapes and learn about higher sugar levels in one colour or another (Watkins, 1999).

Two experiments were conducted. Experiment one attempted to discover whether blackbirds or silvereyes could detect grape tannin, and at what levels they could detect it. In previous experiments artificial grapes or artificial nectar were offered to the birds. Blackbirds were reluctant to take nectar except in very dry conditions (see chapter five). The artificial grape recipe (chapter three) contained gelatine, and difficulties of gelatine combining with tannin were encountered, making it difficult to determine the point at which tannin in the artificial grape would become available, and, further, detectable to birds. Assays showed that to get enough free tannin in the artificial grape so much grape tannin needed to be added that the purple colour changed to dark brown. Blackbirds took all grapes without discrimination anyway. Experiments were then conducted with both species using artificial nectar containing 15% hexose sugar with controlled concentrations of grape tannin.

Experiment two attempted to determine whether green or purple colour alone would prove to be a significant cue when the sugar reward remained constant, and what preferences the birds might show. Artificial grapes coloured green and purple, where sugar remained constant, were offered to the two species in summer and again in winter.

### **7.3 Materials and methods**

Blackbirds were present in the Lincoln University vineyard all year round. Except for clear identification of male, female and juvenile birds, the actual number of individuals could



not be ascertained, but a possibly low number (as few as three and as many as seven or eight) compares with aviary experiments which have been conducted on as few as three individual birds (Schmidt, 2003). The colour experiments were conducted using two sites approximately 60m apart. A further two sites nearby, 100m apart, were used for the tannin experiment. Silvereyes were not present in the university vineyard so these birds were recorded in a pear orchard 4 km away, where they were present throughout the year. The sites used for the colour experiments were approximately 8m from trees, and the same sites were used for the tannin experiments. Silvereyes arrived in flocks and as many as nine birds were counted on the table at one time. The probable number of individual birds was between 12 and 20.

A two-tier bird feeder table (see chapter three) was used. Bird behaviour on the feeder table was recorded on timelapse video, and bird responses quantified recording the length of each bird visit, and an ethogram to quantify the number of pecks delivered (both species with artificial nectar and silvereyes with artificial grapes). For blackbirds that ate or took whole grapes away, this was recorded as grapes taken. Data were analysed by non-parametric tests (Kruskall-Wallis ANOVA (H) and Mann-Whitney U test (U) using Genstat6 software.

### **Tannin experiment**

Tanin SR®, a product mostly sourced from grape seeds, described by the manufacturer (Institut Oenologique de Champagne) as ‘a catechic and ellagic tannin extract, with more than 70% tannic acid content’, was the compound used to simulate secondary metabolites in grapes. HPLC analysis, a process that still cannot be specific with complexes like this, determined the phenolic profile of Tanin SR® as predominantly non-pure, i.e. not simple,

but probably a mixture of oligo- and polymeric phenolics. This reflects the commercial use of Tanin SR®, which is to enhance the soft tannins of wine by adding astringency without bitterness.

Gelatine complexes with tannin, and the artificial grape recipe (chapter three) proved problematic for this experiment. Grapes made in this way became modified in colour, turning from a faintly translucent purple to a dirty brown, depending on the concentration of tannin added. The amount of detectable tannin was uncertain due to the unknown binding capacity of the gelatine. A pilot experimental procedure investigated the binding threshold of the artificial grape, and when the free tannin content in the artificial grapes was ascertained (see results), these were offered to blackbirds in the Lincoln University vineyard in December 2002. Four concentrations of tannin in artificial grapes were offered together with a tanninless control, in a group of five grapes, (10 grapes (two groups) on the top and 10 on the bottom tier) following a latin square design so that no two adjacent grapes were the same concentration, nor was any grape in the same position consecutively.

Due to the difficulty of using artificial grapes, preliminary experiments with silvereyes were then conducted using artificial nectar, which silvereyes take readily. The concentration of tannin was increased above that used in the artificial grapes with blackbirds. As this experiment proved successful, the same procedure was then used with the blackbirds. This species does not normally take nectar, but a previous experiment with grape aromas (chapter five) had successfully attracted blackbirds to artificial nectar in dry summer conditions. In December 2003, which was very dry (1mm rain recorded at Lincoln) blackbirds did visit artificial nectar (though still not as frequently as silvereyes).

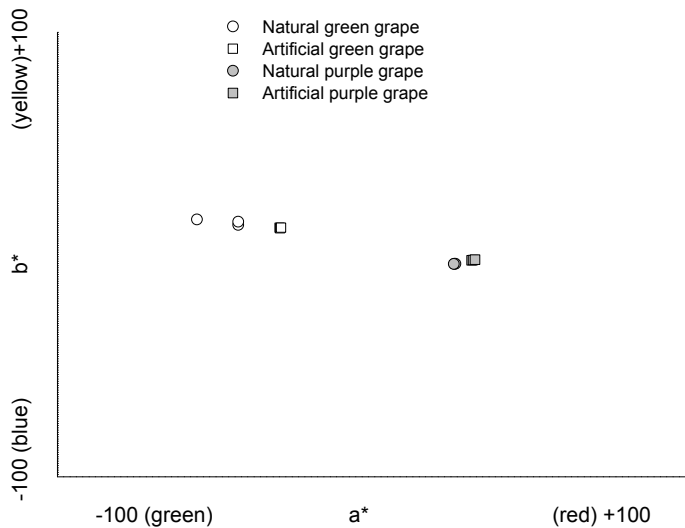
Artificial nectar was made by dissolving 7.5g glucose and 7.5g fructose in 100mL boiling water, which was then allowed to cool. Grape tannin was added at 2.5%, 5%, 7.5% and 10% (w/v) concentration with a control to which no tannin had been added. These concentrations of tannin compare with those normally found in grape skins (2%, Gawel, 1998) or grape seeds (15%, Harbertson *et al.*, 2002). Up to 5% is found in other fruits (Cipollini and Levey, 1997; Stanley and Lill, 2001). This was offered to both silvereyes and blackbirds in small (20mm) pots on a feeder table as was used in chapter five. Five pots with four concentrations of tannin in nectar (2.5%, 5%, 7.5% and 10%) with a tanninless control were set out on each level in a latin square design that progressed each day so that no pot with the same concentration was next to another of the same concentration nor consecutively in the same position.

### **Colour experiment**

For purple grapes 8 drops red (cochineal) and 6 drops blue food colouring (Hansell's Christchurch, New Zealand) were added to the artificial grape mixture before heating, and for green grapes 15 drops yellow and 1 drop of blue food colouring. Visual assessment was used to approximate the colour to that of natural purple and green grapes as closely as possible, though there was a slight difference in hue. Artificial grapes were similar to real grapes in colour, size, and consistency, although they did not have seeds or skins. Three samples of both natural and artificial grapes were measured by Minolta CR-210 colorimeter standardised to a white plate with L at 98.07, a\* at -0.23, b\* at 1.88 to give a measured value for the colours for purposes of repeatability. The artificial grape colour is shown in Figure 7.1, where the CIE a\* (x axis) shows green-red and b\* (y axis) shows blue-yellow parameters. The L parameter (luminosity) values - a third dimension and not

shown - were 77 and 62 for green and purple artificial grapes respectively and 96 and 89 for green and purple natural grapes respectively.

Purple and green grapes were offered together on the feeder table in a simple layout of alternating colours, upper and lower tier beginning with a different colour, and consecutive sessions reversed. No two adjacent grapes were the same colour. A bird had to pass over a differently coloured grape to access a second grape of the same colour.



**Figure 7.1 CIE colorimetric representation of real (N = 3) and artificial (N = 3) grapes.**

## 7.4 Results

### Tannin experiment

The rate at which TaninSR® complexed with the gelatine was calculated by measuring total polyphenols, of which tannin is a large but not exclusive part. The polyphenol concentration was measured for samples with and without gelatine. The average amount of polyphenols of each concentration was calculated afterwards. A tannin concentration of 40 g/L contained a free concentration of tannin of about 12 g/L. Thus about 75% of the tannin formed a complex with the gelatine. The free tannin concentration in grapes with a total tannin concentration of 10, 20 and 30 g/L is about 3, 4 and 5.5 g/L., which is equivalent to 0.3%, 0.4%, 0.55% and 1.2%.

Blackbirds took all the artificial grapes (0%, 0.3%, 0.4%, 0.55% and 1.2%) with no significant discrimination between any concentrations (Table 7.1).

Both species of birds appeared to detect the tannin in artificial nectar. The greatest consumption was of the control tannin-free artificial nectar, followed by 2.5%, 5%, 7.5% and 10% (Table 7.2). The tanninless solution was significantly preferred to all other concentrations for both blackbirds (Table 7.2,  $H = 58.9$ ,  $df = 4$ ,  $p < 0.001$ ,  $n = 22$ ), and silvereyes (Table 7.2,  $H = 21.69$ ,  $df = 4$ ,  $p < 0.001$ ,  $n = 57$ ).

Blackbirds took artificial nectar at 2.5% concentration significantly more than 5% ( $U = 17.5$ ,  $p = 0.003$ ), whereas between 5, 7.5 and 10% there was no significant difference in the number of pecks at each level ( $H = 0$ ,  $df = 2$ ,  $p = 1.0$ ).

**Table 7.1 Artificial grapes with tannin added taken by blackbirds**

Tannin concentration	0%	0.3%	0.4%	0.55%	1.2%
Percentage eaten	93.75	91.6	91.6	95.8	98

**Table 7.2 Mean (median, se) pecks per visit to artificial nectar with tannin added**

Tannin concentration	0%	2.5%	5%	7.5%	10%
<b>Blackbirds (n=22)</b>					
	5.0(3,1.0)	2.1(2,0.28)	1.7(1.5,0.48)*	1.4 (1,0.40)	1.4(1,0.4)
<b>Silvereyes (n=57)</b>					
	4.2(2, 0.56)	3.3(2,0.32)	2.3(2,0.19)	1.7(1,0.13)*	1.6(1,0.1)

\*Significant threshold

Silvereyes showed significant preferences in numbers of pecks per visit (Table 7.2,  $H = 75.7$ ,  $df = 4$ ,  $p < 0.001$ ) between 5% and 7.5% ( $U = 2105$ ,  $p < 0.001$ ). The number of pecks to 2.5% concentration was not significantly different to that of 5% ( $U = 4671$ ,  $p = 0.4$ ), nor was 7.5% significantly different to 10% ( $U = 4946$ ,  $p = 0.89$ ).

### Colour experiment

Blackbirds exhibited no preference for either colour in summer (Figure 7.2a, 7.2b,  $n = 75$  bird visits, total grapes taken 232 over 13 days), either as overall total number of grapes taken or as first grape attacked per visit. The mean number of total grapes per day and of first grape per visit taken was similar for both purple and green grapes ( $U = 70.5$ ,  $p < 0.5$  and  $U = 74.5$ ,  $p < 0.6$  respectively). But in winter blackbirds exhibited a significant preference for purple grapes (Figure 7.3a, 7.3b,  $n = 30$  bird visits, total grapes taken 57 over 8 days), both in the total number of grapes taken and in the first grape taken on each

bird visit. ( $U = 0.0$ ,  $p < 0.001$ ,  $U = 4.5$ ,  $p = 0.007$  respectively). Only one green grape was taken among the total of 59 grapes.

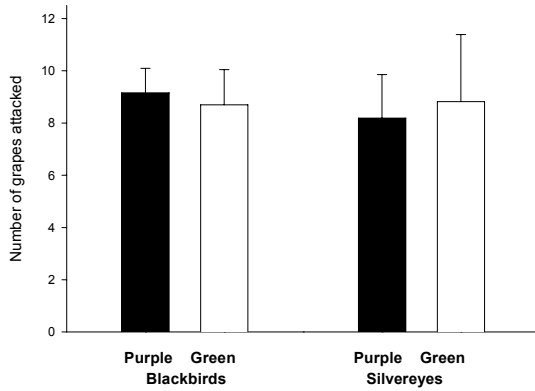


Figure 2a: Summer colour choice - mean number of total grapes attacked per session by blackbirds (N=75 bird visits, 192 grapes) and silvereyes (91 bird visits, 187 grapes) with se bars.

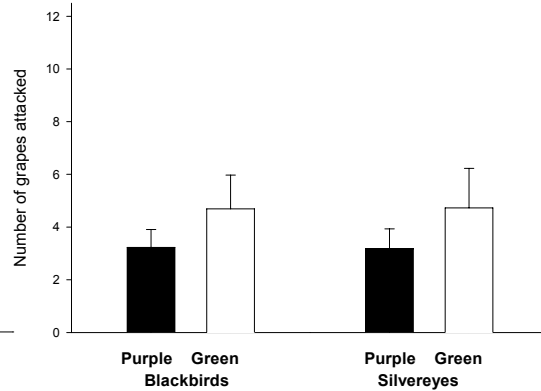


Figure 3a Summer colour choice - mean number of first grapes attacked per session by blackbirds (N=73 bird visits, 66 grapes) and silvereyes (91 bird visits, 87 grapes) with se bars. some bird visits did not take grapes).

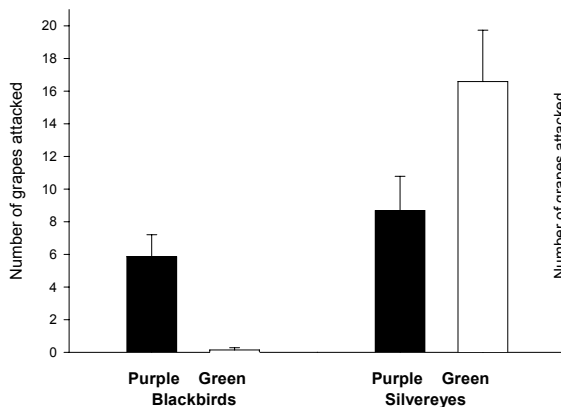


Figure 2b Winter colour choice - mean number of first grapes attacked per session by blackbirds (N=27 bird visits, 42 grapes) and silvereyes (152 bird visits, 303 grapes) with se bars.

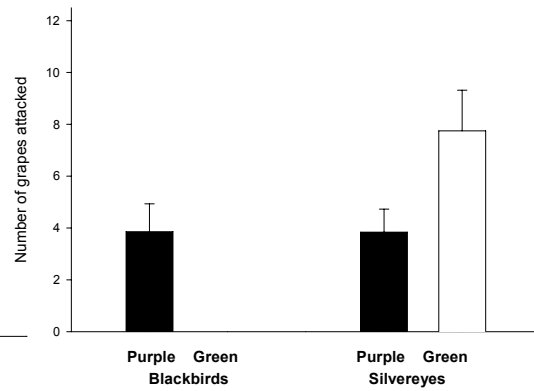


Figure 3b Winter colour choice - mean number of first grapes attacked per session by blackbirds (N=27 bird visits, 27 grapes) and silvereyes (152 bird visits, 139 grapes) with se bars. some bird visits did not take grapes).

Silvereyes also exhibited no clear preference for either colour in summer (Figure 7.2a, 7.2b, n=91 bird visits over 11 days,). The mean of the total number of purple grapes pecked per day and of first grape pecked was similar for both colours ( $U = 54.5$ ,  $p < 0.7$ ,  $U = 55.5$ ,  $p < 0.8$  respectively). In winter (Figure 7.3a, 7.3b, n=152 bird visits over 12 days) the mean number of total grapes attacked and of first grape attacked per visit was

significantly greater for green than for purple ( $U = 39.5$ ,  $p=0.06$ ,  $U = 37.0$ ,  $p<0.05$  respectively), but results were not so pronounced as for blackbirds.

## **7.5 Discussion**

These results indicate that both blackbirds and silvereyes detected tannin and colour differences, and that they may use them as a basis for making foraging decisions with grapes. Both species of birds could detect grape tannin in artificial nectar. Concentrations at which aversion was displayed were different for each species, being lower for blackbirds (between 2.5% and 5%) than for silvereyes (between 5% and 7.5%), suggesting that blackbirds were better able to detect tannin than silvereyes.

In the initial experiment with artificial grapes blackbirds took all the artificial grapes displaying no signs of detecting tannins, but the tannin concentration was lower than that of the threshold of detection shown in the artificial nectar experiments. Blackbirds' foraging behaviour is normally to take a whole grape and swallow this in a second or so (chapter three), so it is possible that the bird was unable to detect tannin when swallowing whole berries in this way. Stanley and Lill (2001) also found that silvereyes showed no aversion to small artificial berries (2-3mm) containing quebracho (a powdered form of tannic acid extract from the bark of the quebracho tree) but did show aversion to a mash with the same tannin concentration. They suggest that berry shape or size may be an ecological parameter of seed dispersal, and that birds may not be able to detect tannin so easily in berries they swallow whole. Low concentrations of tannin in grape skins may mean that a blackbird will not detect tannins in grapes swallowed whole.



From our results it would appear that the concentration of 2% tannin, as is found in grape skins, would not be avoided by these two bird species. However concentrations of 15% such as is found in seeds would be avoided if they were detected. The mechanism by which birds may detect tannin appears to be crucial if they need to avoid ingesting tannins. So far we do not know if birds can differentiate and use the difference between the astringency of long-chain tannins (predominantly in skins) and bitterness of short chain tannins (in seeds) to discriminate between potential benefits and toxicity. Long chain or condensed tannins are known to detrimentally affect digestive efficiency in mammals, while short chain tannins, such as are in seeds, although bitter, appear not to be toxic to birds. Bitterness is a taste while astringency is a sensation. Birds have far fewer taste buds than other vertebrates (as low as 24 (chicken), 46 (bullfinch), 62 (Japanese quail) or 200 (starling) and all of these are on the tongue adjacent to salivary glands (Mason and Clark, 2000). In mammals, tannin binding with salivary protein is often credited with the creation of the sensation of astringency, which is felt on the inside mouth. There is literature that suggests that avian taste detection is primarily by sensation (Clark and Shah, 1994; Wager-Page and Mason, 1996), but Karasov (1996) questions whether birds actually produce the proline-rich saliva that binds with tannins. Low levels could possibly work to enhance the attractiveness of the berry (as astringency enhances taste for humans, Butler, 1992; Adams and Herbertson 1999).

Results from these experiments suggest that lower levels of tannins are not as aversive to birds as concentrations above 5%. Tannin concentrations in seeds are 15%, and the resistance of these two bird species to concentrations above 5% suggests that seeds would not be willingly ingested by these birds if the tannins levels were detected. This is generally accepted as the reason plants offer seeds in an attractive sugared fruit pulp.

Sugar rich fruit pulp, berry size, and colour may be closely linked to manipulate avian perception of tannin. Winkel-Shirley (2001) suggests that while anthocyanins attract seed dispersers, tannins act as feeding deterrents. Seed tannin, whose functions include preserving the immature seed from UV and other degradation (Winkel-Shirley, 2001), must not deter birds from dispersing them. Plant manipulation of seed disperser foraging behaviour in the interests of seed viability is a concept that has received little attention (Murray *et al.*, 1994). Frugivore tolerance of seed tannin is low and their perception is also low. They regurgitate, defecate rapidly, and have large livers, all of which minimise toxifying effects. But why should birds have invested energy in mechanisms for tolerating tannins? What trade-off do the birds receive?

For frugivorous bird species the inclusion of fruit secondary metabolites in their diet has been described as a paradox (Stanley and Lill, 2001). A diet of only fruit leads to weight loss (Bairlein, 2002) and many experiments with captured birds have been halted at this point due to fears for their survival (*ibid.*; Place and Stiles 1992.). This has led to a general acceptance that birds cannot survive on fruit alone. However, Bairlein (1988, 2002; Bairlein and Totzke, 1992) found with his experiments with garden warblers (*Sylvia borin*) that at this critical stage their metabolism slowed, possibly to enable survival on minimal nutrients. At the same time they increased their food intake, possibly triggered by extreme hunger, which led to rapid weight gain, especially when feeding on high lipid fruits that provide fat for deposition (Bairlein, 2002). In addition it appeared that secondary metabolites (as yet undefined) from black elderberries, but not from red elderberries increased nutritive efficiency in the birds (Bairlein, F., pers. comm. 03 June 2003). A process such as this to gain weight may be common not only in birds preparing for migration, but also for birds preparing for over-wintering in cold conditions (Totzke *et al.*,

2000; McWilliams *et al.*, 2002).

The blackbirds in this experiment exhibited no preference for purple over green in summer, but in winter they exhibited an overwhelming preference for purple grapes over green grapes that were otherwise identical. Out of a total of 59 grapes taken in winter only one was green, and this grape was taken second to a purple one in the first visit of a session, possibly an example of the 10% ‘sampling’ that is typical of avian foraging behaviour. This bird was thus able to ascertain that the green grapes were identical in taste and nutritive value to the purple ones, but still green grapes were left. This is even more impressive given that the probability of purple being taken by chance declined significantly as purple grapes were taken and green grapes were left. (It was not feasible to replace grapes to redress an imbalance of probability as purple grapes were taken.) Although the number of individuals in these studies may have been few and results regarded as preliminary only, set beside the summer results where green and purple grapes were both taken equally from the same site, the winter results reported here suggest a marked seasonal preference for purple over green. Two extra blackbirds recorded during the silvereyes winter colour experiments at a different location also took only purple grapes

Many varieties of grape are purple in colour, (wild grapes were all purple, Hardie and O’Brien, 1988) a colour that is a cue for high sugar content (Hrazdina *et al.*, 1984; King and Young, 1999) but which also signals high secondary metabolite content (Winkel-Shirley, 2001). Blackbirds are native to regions with cold winters, while silvereyes (which did not exhibit this colour preference in winter) originated in warmer climates (South-east Asia and Australia). Blackbirds’ winter diet preference for purple-black fruits has been noted by other authors (Sorensen, 1981; Willson, 1986). Recent research supports the

hypothesis that purple in fruit is a seasonal cue to more than just sugar or lipids for those birds that are subject to seasonal metabolic changes (Bairlein, 2002).

Previous experiments (chapter four) comparing the responses of these two species to hexose sugars suggested that they may have different digestive systems, particularly for assimilating glucose (see Afik and Karasov, 1995). Silvereyes, like other small passerines, may have a passive glucose assimilation process that absorbs efficiently. This type of passive digestive system may be vulnerable to assimilation of toxic substances or xenobiotics (Caviedes-Vidal and Karasov, 1996). If tannins and other secondary metabolites are cued by purple colour then this theory would explain a tendency for silvereyes to avoid purple food sources. If they customarily avoid fruit with secondary metabolites by noting the obvious colour cue, they might have less need of a finely-tuned taste detection ability. The experiments with tannins reported here suggest even more strongly that the nutritional strategies of these two species in autumn may be entirely different, governed by different endogenous drivers and may even be genetically programmed. Fruit may well fulfil dietary functions that are totally dissimilar.

Karasov (1996) noted a dietary inhibitive effect of secondary metabolites, though whether this was due to secondary metabolites reducing digestive efficiency per se or decreasing utilization efficiency due to detoxification costs remained in question. Cipollini and Levey (1997), Cipollini (2000), Bairlein and Totzke (1992), Stanley and Lill (2001) Bairlein (2002), all noted a paradox of unpalatable fruit ingested by frugivorous birds in spite of assumed dietary costs. That the costs actually confer benefits to some species to some extent might explain this paradox. It is possible that species preparing for migration, or for winter, such as blackbirds, needing to accumulate seasonal fat rapidly, and being unable to

detect tannin in a berry swallowed whole, use colour as a cue to fruit that will provide the secondary metabolites and lipids that will be beneficial to them. Silvereyes may also use colour to cue them to secondary metabolites, but in this case purple may cue to toxicity that they need to avoid. The assumption that colour cues ripeness and therefore sugar, while justified, may underplay the significance of colour to frugivorous species in the matrix of seasonal avian ecology.

## **7.6 Conclusion**

It is possible that in attempting to protect grape crops from frugivorous birds growers are combating a problem that is more complex than simple accessibility and abundance of the fruit. Differences between the two species investigated in this project that were already apparent from the sugar experiments appear to extend beyond the digestive assimilation of glucose and behavioural characteristics that could be related to those physiological differences (see chapter four for discussion). Some frugivorous birds, in this case blackbirds, may need grapes for their winter survival, and use purple as a cue to the secondary metabolites that are part of a seasonal metabolic process. Silvereyes, however, may use green as a cue to less toxicity in the fruit. Grapes are available in New Zealand late into autumn and the significance of the colour cue may change during the ripening season in a different way for each of these species. Avery (2002) noted that an ecological solution to bird damage rests in providing an alternative food source. With greater understanding of what each avian species needs from grapes, an ecological solution to the problem may come closer.

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## **Chapter eight**

### **Conclusions**

#### **8.1 Summary of findings**

The underlying theme of this research was to increase an understanding of bird depredation on ripening grapes in a vineyard situation, with a view to managing birds on a sound ecological basis. The experimental academic research reported here was conducted in tandem with field surveys (Davies et al., 2000; Saxton et al., 2002), a grapegrower damage survey card (See Appendix II) with data collection from grapegrowers, and a survey on the bird species populating native tree remnants in Marlborough (McEntee *et al.*, 2004). The experimental research is reported in this thesis, but the ecological and environmental knowledge gained through the management of the research and from academic papers has led to an appreciation of the very wide scope of this problem. Successful bird management will almost certainly be effected at the environmental level, and understanding of bird motivations has been increased through this work. The experiments were conducted with European blackbirds and Australasian silvereyes, both of which do substantial damage to grapes in New Zealand. There are three other major species that damage grapes – starlings, mynas and thrushes - that deserve attention but could not be included in this project.

From the start, behavioural differences between the two species was striking. The survey in Marlborough (chapter three) was undertaken to confirm the similarity of bird behaviour on the bird feeder table to that of the same species in a vineyard. With that confirmation in place the project was conducted as a comparative behavioural study. Tinbergen (1963) suggested that a comparison of elements of the same order throws up differences in

behaviour that pinpoint genetic differences. He also saw a dual approach, that ethology (animal behavioural psychology) breaks down complex phenomena into smaller scenarios, while neurophysiology builds up a picture from tiny physiological details. In this project, the bird damage picture was broken into small blocks for study. Focus was put on the major compounds of ripening grapes one by one, using an artificial grape to control for simultaneous changes in other compounds during ripening. The major compounds investigated were sugar, acid, colour, aroma, and tannins. Aspects such as abundance, conspicuousness, size, position, tactile and visual characteristics such as softness, translucency and ultra-violet reflectance were not addressed directly in this study, but undoubtedly play a role. Environmental factors were also not addressed directly, but are probably the major tool available immediately for influencing bird populations and behaviour in vineyards. During ripening, grape compounds change simultaneously, and these changes are interlinked both to each other within the grape and in the matrix of cues that may be perceived by birds. Reporting the results of the experiments has become a complex task of cross-referencing between chapters. In order to make sense of the cross-referencing, and in order to elucidate the meaning of the cues to the birds, each bird species is reported separately.

Blackbirds are relatively large birds (18-30cm) and are generalist foragers (Snow, 1958; O'Connor and Shrubbs, 1986). They are resident and territorial at nesting time, but probably do not defend a food source as abundant as ripening grapes (Carpenter, 1987). They are ground-hopping and typically fly up into the vines to pluck one whole grape and take it to cover to consume (Watkins, 1999; Saxton *et al.*, 2002). They spend a lot of time not eating, are solitary and watch for predators during this time, and they may also be using this time

to digest the large amount of food consumed at one time (Karasov, 1993). They return at fairly regular intervals for another grape (chapter three). At nesting time (November-December) they were observed to take many grapes in their beak and fly off (chapter four), but this did not occur at other times of year.

Blackbirds displayed a preference for very high hexose sugar concentrations (chapter four) and the geraniol aroma of ripe grapes (chapter five). They were impervious to high acids (chapter six), preferred purple coloured grapes in winter but took both green and purple in summer (chapter seven), and could detect tannins at levels above those found in grape skins but below those of seeds (chapter seven). Karasov (1990) suggested that blackbirds have an enzyme or carrier-mediated digestive system for absorbing glucose that is similar to other protein-eating vertebrates. Such a system has a relatively low rate of glucose absorption efficiency (between 35% and 70%, Karasov and Levey, 1990), and correlates inversely with gut passage rate (GPR) to void the food to make room for more. A fast GPR is common for birds eating fruit, where the mean gut retention time is only 46 minutes, compared to 62 minutes for insects (Afik and Karasov, 1995). Blackbirds typically take whole grapes including the seeds, suggesting that co-evolution with wild grapes as a seed-disperser (Hardie and O'Brien, 1988) may explain some characteristics of their preferences. Sugar is a source of calories only, and is not an essential nutrient (Karasov and Levey, 1990), and grapes are relatively low in nutritive value (Herrera, 1981).

However, it is possible that, similar to the migratory birds reported by Bairlein (2002), in preparation for winter, blackbirds eat a fruit only diet to modify their metabolic rate. When this has slowed down to match the minimal nutrition (some eight days later), extreme hunger precipitates over-eating and leads to fat accumulation. Secondary metabolites,

possibly signalled by purple colour, may be instrumental in fat accumulation for winter, as they are for some migratory species and species preparing for winter (Bairlein, 2002).

Abiding in the vineyard year-round, blackbirds may well become conversant with stages of ripening grapes on their patch, and be able to interpret aromas as a cue to ripeness.

Blackbirds take unripe grapes, and high levels of acid appear to be no deterrent (chapter six), while the aroma of unripe grapes was preferred to no aroma at all (chapter five). In summer (January in these experiments) or pre-véraison, blackbirds did not show aversion to green coloured grapes – the preference for purple was a winter preference.

Grapes may support a metabolic strategy that blackbirds need in preparation for winter, thus the presence of grapes may contribute to their selection of nesting site and territory.

Blackbirds are often seen in scrub and small shrubs beside vineyards, which do not provide dense or particularly safe cover. Their need for grapes may be great enough to encourage adaptation to extreme environmental modifications. Their depredation of grapes may well increase as the season approaches winter, and thus contribute to the extreme pressure experienced by vineyards close to harvest. Shooting blackbirds is a common strategy – they are ground hopping and visible. However, they may not be the worst offenders in the vineyard.

Silvereyes are a small (8-12cm) bird that is self-introduced from Australia, and is predominantly insect-eating at nesting season and nectariferous in autumn (Moed, 1981).

They fly in small groups and typically arrive in vineyards during the last six weeks of ripening. They fly into the vines from surrounding cover, preferring willows particularly,

and feed continuously, pecking at many grapes and leaving them on the vine (chapter three). This results in further damage from insects or acetobacter, which causes sourness and off-odours in the grapes and deterioration of quality. Such grapes are often refused by the winery (Harvey, D. pers. comm.. 29 April 2004) .

In these experiments silvereyes exhibited a preference for 10-15% hexose sugar (chapter four, chapter six), preferred the aroma-less control to grape ripening aromas. They were attracted by the IBMP (2-3 isobutyl-methoxypyrazine) aroma of unripe grapes but only from a distance, and showed an aversion close up (chapter five). They did not show a dislike of acid (chapter six) and preferred green to purple grapes in winter, though this preference was not shown in summer (chapter seven). They displayed less ability to detect tannins than blackbirds (chapter seven).

Many small passerines, of which silvereyes are one species, have a passive glucose absorption system that assimilates glucose directly through the intestine wall without the mediation or protection of a carrier medium (Caviedes-Vidal and Karasov, 1996).

Assimilation can be as much as 95%, or almost total, which means that rate of assimilation controls rate of feeding. The need to continually feed at low levels of glucose intake would explain silvereyes' behaviour. Because they need to ingest slowly, they must feed at length, which is expressed in continual pecking rather than whole grape ingestion, and need the protection of a flock where many eyes look out for predators, instead of the solitary habit of blackbirds that take much less time to feed. This type of digestive system may also be vulnerable to absorption of toxic substances (small hydrolysable xeno-biotics, Caviedes-Vidal and Karasov, 1996), so silvereyes, if they have this digestive system,

would need to be selective in choice of food and sensitive to cues to possible toxicity. This would explain their dislike of malic acid, which metabolises during ripening and therefore is indicative of unripe grapes. Malic acid is sharper than tartaric and may be less masked by sugar concentrations. It would also explain their aversion to purple, since purple could be a cue to tannins that are possibly toxic to these small birds.

Silvereyes have no history of evolution with grapes, and generally do not inhabit vineyards throughout the year, so would not be familiar with stages of grape ripening. This could explain their lack of response to grape aromas as a cue to ripening grapes. It is possible that silvereyes are seeking purely water and sugar from grapes, or may feed on grapes because other food sources have disappeared. Rooke (1984) found that given a choice of white and red grapes, figs, nightshade berries, nectarines, and sugar water, they liked grapes and nectarines least of all. Their relative insensitivity to tannins compared to blackbirds is interesting, but may be because they use the initial colour cue to avoid tannins before tasting or ingesting them.

This leads to the conclusion that grapes do not complement silvereyes needs in the same way as they may do blackbirds, and that silvereyes may arrive in vineyards for environmental reasons of cold, lack of food elsewhere, or other unknown reasons, rather than to seek out grapes particularly as a food source. This concurs with anecdotal evidence from grape growers, and with the observations from Margaret River, Western Australia that when the Marri bush flowers well (as in 2004) grape damage from silvereyes is considerably lessened (Rooke, 1984; M. Gray, viticulturist, pers. comm, 26 July 2004). The provision of an alternative food source does appear to influence silvereye movements.

This thesis work has shown clearly that the species involved are very different in their vineyard behaviour, and will possibly require targeted management. Silvereyes may be easily attracted by a more satisfactory food source, blackbirds may be much more difficult to move because they need grapes in Autumn. The role played by the grapes themselves is clearly not as obvious as has been thought. For blackbirds, grapes or similar fruit may be essential to a bird's winter survival. For itinerant flocks of silvereyes grapes may simply be a source of water and energy. It is clear that birds in vineyards represents a complex ecological system, details of which remain uninvestigated, and while this is so our ability to manage with foresight will be minimal. A need exists to define areas for immediate research for effective management of bird populations.

## **8.2 Where to next?**

In 1972 Boudreau recommended that bird behaviour and neurophysiology be studied to determine the correlates of bird depredation of grapes. Research studies on applied ecological parameters of bird management are few. Research supporting the findings of this work was found mainly in ecological studies of bird frugivory generally. Work by William Karasov in Wisconsin has contributed greatly to our understanding of avian glucose assimilation, and that of Franz Bairlein in Europe to that of the possible role of secondary metabolites in avian metabolism, both studies have been drawn on in the discussion of the results presented here (chapter four; chapter seven). Avery (2002) stated that for an ecological solution to bird damage to fruit and crops to be effective an alternative food source for the birds would be essential. This would require knowledge of what birds need nutritionally and the physiological correlates of their foraging. The two species investigated in this work displayed contrasting behavioural patterns that reflect a

likely raft of basic cultural and physiological differences. This leads to question of how these two species interpret cues from ripening grapes, which cues are significant, and why. Further physiological and neurological work on the specific avian species would be needed to ultimately confirm the explanations advanced above. Such work is time-consuming and expensive. Meanwhile how can bird behaviour in vineyards be better managed?

Management of problem bird populations is an immediate need of the viticultural industry in New Zealand. Operations research or management science (OR/MS) often utilises a scoping diagram as a preliminary formulation of a system with a view to modelling it to solve problems or to improve outputs (Daellenbach, 2001). Systems are human conceptualisations, often depicted as artificially closed systems, and hardly ever represent the total picture, but even so are useful tools for representing problem situations.

Using this technique bird damage to grapes can be formulated as a system, albeit a complex ecological one where most of the inputs are not controllable. Figure 8.1 represents a scoping diagram, or formulation of the problem. A scoping diagram can be a first step towards system modelling so that outcomes may be predicted when inputs change.

Application of this to the problem of bird damage is not possible yet, since many of the inputs cannot yet be quantified, making it difficult to predict outcomes. However, with further work in other areas of this scoping diagram, modelling may be feasible.

The white boxes represent inputs elucidated by the research reported here. Diagramming the problem situation at least shows the where management of some of the blue inputs, located mainly on the periphery of the system, and subject to complex ecological



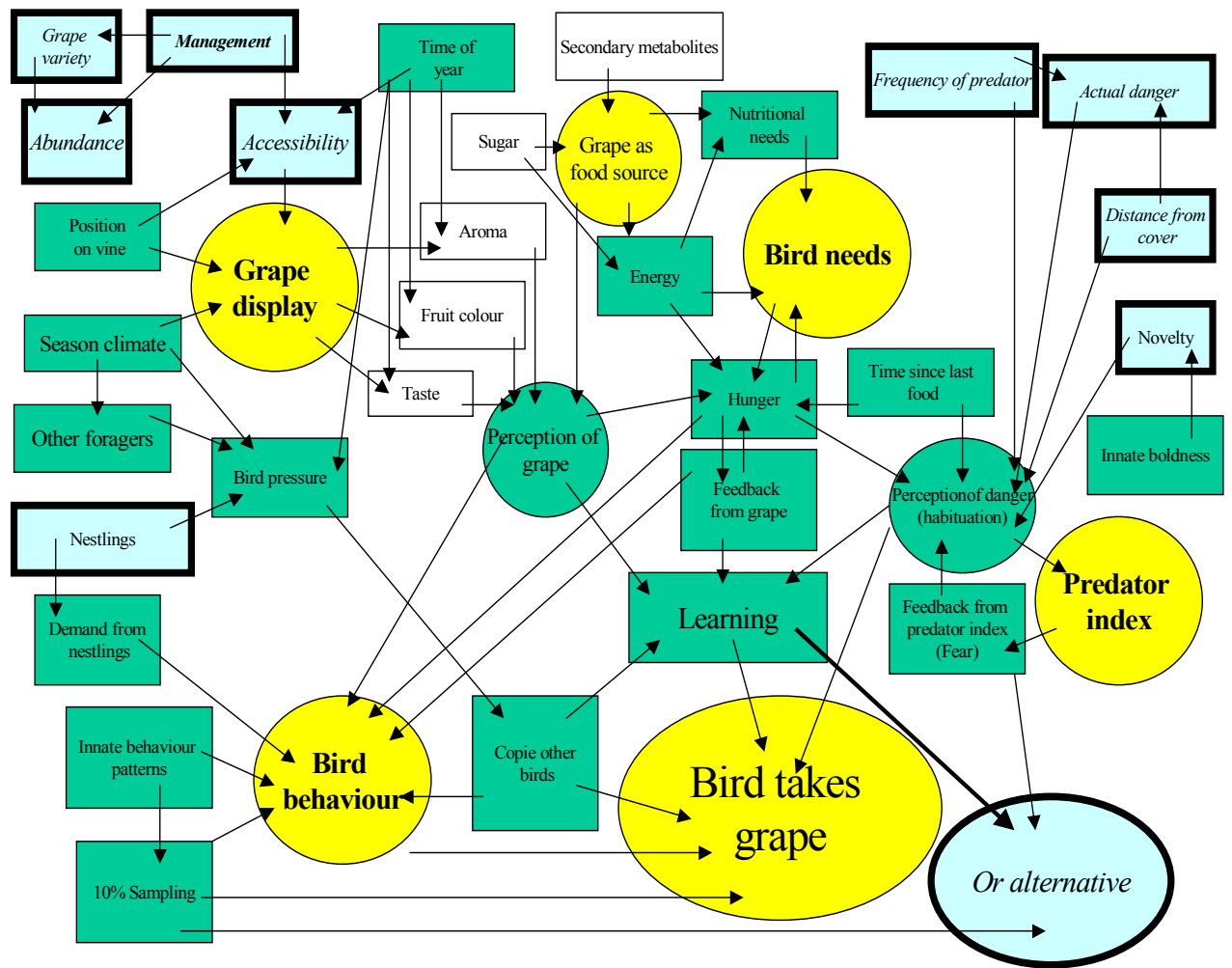


Figure 8.1 Scoping diagram of the system of foraging decisions by birds. Inputs are in square boxes and outputs are oval. Green signifies an ecological input or process. Yellow circles are clusters of inputs and processes, or subsystems. Not surprisingly for a complex ecological system, many inputs contribute to more than one subsystem or process. White indicates the scope of this experimental research. Blue indicates points at which human inputs may contribute to the system

processes before contributing to outputs. The main areas that could be manipulated with our present knowledge are in vineyard planning (cultivar choice, trellis height, canopy management and other management), and in maintaining a high predator index (or keeping the birds scared). At present this process appears to be undervalued for its contribution to

bird decision making. This diagram shows clearly areas where managers could influence bird pressure in the vineyard. It also underlines the crucial role of bird learning to the process of foraging. The remaining blue box, that of nestlings, could be interpreted in several ways – reducing nesting sites, harvesting eggs or nestlings, and disturbing birds at nesting time.

The predator index, or how to keep birds scared, involves inputs of novelty, distance from cover, actual presence of a predator, all building on innate bird timidity. A natural predator that could reliably be expected to harass birds continuously during the ripening season would be the natural answer to this aspect of the diagram.

### **8.3 Final conclusions**

The research reported in this thesis has contributed to knowledge of bird behaviour with grapes. The context was as natural as possible, the subject birds were undisturbed in their natural environment. This lends credibility to the authenticity of their behaviour. Increased internal validity will decrease external validity and vice versa (Kamil, 1987). There is a compromise involved in field experiments in that they are less controlled than laboratory experiments, notably in the reliability of sample sizes. The possible danger of pseudoreplication, in that bird visits might not have represented independent data, was the subject of much thought throughout the work, particularly with the blackbirds, which, being territorial birds and having the characteristic pattern of repeat visits (chapter three), are likely to have been the same birds on repeated visits. At each of four sites there were at least three individual birds that could be identified from plumage and beak colour differences, and data were pooled to give larger samples. Results obtained were consistent,

and corroborated previous experiments, as in the acid levels (chapter six) confirming the sugar preferences (chapter four) and colour preferences were confirmed by blackbirds that were recorded at the silvereve sites in winter. Reports from the literature, as in blackbird colour preference for purple/black which has been reported by other authors (McCann, 1953; Sorensen, 1981,1984; Watkins, 1999). Birds were not marked principally because no method could be devised to mark them without catching them, though much thought was given to this. Banding birds is a process that requires training, licensing, approval of an ethics committee and time. Silvereys were often present several at a time and there were clearly small flocks of about 6-12 birds. Silvereve banding exercises have historically returned little information, as the birds move around so much. It was not unlikely that tagged birds would not be seen again.

In a vineyard situation, grapes can sustain multiple attacks from the same bird and it is the length and severity of the attack on grapes that was the principal point of interest and, therefore, an appropriate unit of measure used for this work. For both species many locations were tried without birds visiting, so the possibility of driving away those birds that did visit was a factor that encouraged minimal intervention in the natural field situation where birds were free not to visit. For further experiments an aviary context where the bird population sample can be controlled might be appropriate, though aviary populations are often only a few individuals, as few as three (Schmidt, 2003), which is fewer than in most of these experiments. The size of the bird visit samples (often above one hundred) reduces to some extent the risk of unreliable results through pseudo-replication. A final reason was that if time had been taken to mark or catch the birds in some way fewer experiments could have been conducted. As the methodology and scope

of the research was untried, it was judged a better use of time to proceed with the experiments on the understanding that significant results could, if necessary, be pursued more rigorously in future work. Field experiments are most successful when they obtain large effects with simple designs (Haven-Wiley, 2003), and, finally, the call for laboratory work to be confirmed by field experiments is a common one (Stanley, 2001). The trade-off of internal validity for external validity has clearly been worthwhile.

Robust, statistically sound data from vineyards are lacking, anecdotal evidence is sporadic and may or may not be a true reflection of the majority of cases. The need for further field research is clear, and the research presented here provides a sound foundation for further work.

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# Epilogue

## Future directions

This research sits at the beginning of an ecological approach to developing an avian management strategy. Several immediate questions beg to be investigated:

### **1. Seasonal colour choice change**

The colour choice change from summer to winter – is this an individual bird learning process or is it genetically programmed? Is it an individual learning for silvereyes and genetically programmed for blackbirds? Comparative aviary experiments with identifiable birds would elucidate this.

### **2. Confirming sugar and secondary metabolite conclusions.**

With sugar and secondary metabolites further work in the fields of neurophysiology and endocrinology on these particular species to determine the actual physiological processes involved would remove the conjecture of these conclusions. Work has been done in the United States on avian glucose absorption of some species, and in Europe on the role of secondary metabolites in avian nutrition. In view of the costs involved in research of the type just mentioned, it may be preferable to base bird management decisions for these species on conclusions reported here even though they are to some extent conjecture, until they prove false.

### **3. Study of starling behaviour**

Boudreau already in 1972 advocated the study of bird behaviour and neurophysiology in the interests of solving the problems of bird depredations of vineyard grapes. This work

has addressed blackbirds and silvereyes and shown that the two species are very different. Similar work to this with starlings, a real problem species in many parts of the world, might advance knowledge about cues that grapes give to them.

#### **4. Development of a systems model.**

Development of a management model such as is outlined in the conclusion is an obvious next step. This work would also pinpoint areas in which knowledge is incomplete and identify experiments that would help understanding. The eventual outcome would be a model that could be applied to any individual vineyard situation.

#### **5. Evaluation of present environmental management**

To date management solutions to the problem have been adhoc, consisting mainly of modifications of the environment to create monocultures that are inhospitable to birds. Bird diversity results from landscape diversity, and whilst the development of a monoculture has reduced bird pressure in the immediate timespan, it is possible that species specificity will arise. The pest birds concerned with grape depredation are resilient, intelligent and adaptable. A monoculture that supports few species will harbour only these very birds.

In Marlborough this landscape is perceived by non-industry perspectives as sterile and unattractive, and where industries such as tourism co-exist with viticulture, this monocultural ecology is not considered satisfactory. In addition there are some perceptions that birds are becoming as numerous as ever despite considerable removal of trees. Work has already begun to evaluate possible alternative planting strategies to increase landscape diversity without increasing bird pressure on vineyards (McEntee *et al.*, 2004).

## **6. Alternative food source.**

Any ecological solution will need an alternative food source (Avery, 2002). Research is needed to define the constituents needed to

- provide an inexpensive alternative food source that costs the bird less to obtain (obeying energy budget models). This involves developing the perfect food at little expense.
- introduce so much difficulty for the bird to obtain the original food source that it becomes more energy efficient to move to another location at least temporarily (obeying predator prey model). This involves interfering in the habituation process so that birds remain scared.

Research is needed to define underlying drives and mechanisms that encourage birds to eat grapes, and to make grapes less desirable by counteracting some of these (by increasing predator pressure or decreasing the attraction of a vineyard for birds). Birds learn ontogenetically, and almost certainly cultural transmission means that such learning can be passed on to flockmates and to future generations of birds. This means that if the problem can be solved in an ecological way the birds themselves would continue the behavioural pattern.

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## **Appendix I**

### **Comparison of a vineyard silvereye population with that in a nearby orchard.**

#### **Introduction:**

The experiments reported in this research were conducted in an orchard where silvereyes were present year-round. Silvereyes are known to be mostly itinerant (Kikkawa, 1962; Stanley and Lill, 2002; Reese, pers. comm., 2004) and anecdotally appear in vineyards some weeks prior to harvest, which indicates that these vineyard visitors re-locate at least twice annually, and maybe more often. Birds that adapt rapidly to a new situation are generally considered neophilic (curious and bold) comparative to similar closely related species, and this is characteristic of species that colonise comparatively widely and successfully (Greenberg, 1990; Greenberg and Mettke-Hofmann, 2001). The opposite tendency is that of neophobia, expressed as stereotypicity, or a narrowing of behaviour, that often accompanies successful graduation to adulthood, where learned choices have led to survival and are therefore safe (Bateson, 1971).

Silvereyes have clearly adapted to changes in landscape, and profited from the establishment of vineyards, which in New Zealand has occurred in large numbers in the last twenty years. It is possible that the present scenario of silvereye flocks arriving in certain vineyards shortly before harvest will prove not to be an established pattern. But at present it appears that silvereyes are colonising widely into vineyards and that they are adapting to the food source that grapes offer, which is often after other food sources that

they regularly exploit are exhausted. The implication is that more silvereyes will survive and populations that colonise vineyards will increase.

In order to test the hypothesis that vineyard birds would be more neophilic, a population that arrived regularly in a Canterbury vineyard was compared to the population in the orchard that was used for further experiments afterwards. A novel food source was presented to each population and the responses of the birds measured to discover any significant differences between the populations.

## **Materials and methods**

Two populations of silvereyes were observed and compared. The first was a population that arrived annually at a vineyard in early April, the second permanently resident in a pear orchard approximately 9km from the vineyard. The two sites were similar in size (2Ha), and in elevation (100m asl). The vineyard was at the foot of the Port Hills, near Christchurch, New Zealand, the orchard approximately 4km from the foot of the hills. The vineyard had willow trees *Salix matsudana*, some *Eucalyptus nitens* and some New Zealand native flax *Phormium tenax*. The pear orchard also had willow trees and gums of the same species. Silvereyes are often seen to use willow trees for cover. The part of the vineyard that annually received most damage from silvereyes was planted in Pinot blanc grapes (silvereyes preferred green to purple grapes in chapter seven).

A bird feeder table, similar to that used in other comparative experiments (chapter three) was used. Bird behaviour on each table was recorded on timelapse video, behaviour quantified and resulting data analysed. Bird visits were recorded more frequently in the orchard than in the vineyard, so tables were set at three locations in the vineyard, more

than 50m apart, and one table in the orchard. Recording began in the vineyard on 7<sup>th</sup> April 2002, but no birds were recorded till 12<sup>th</sup>. From 12<sup>th</sup> April recordings were made in the vineyard and the orchard simultaneously until 23<sup>rd</sup> April 2002.

Nectar pots coloured bright red (birds have shown aversion to bright red, Greenberg and Mettke-Hofmann, 2002) 2cm wide, 1cm deep, filled to the rim with artificial nectar were set on each of the two feeder table levels (four pots per table). Between the two pots on each level was set a cluster (approximately 30 grapes, 8-10cm across) of Pinot blanc grapes from the vineyard. The artificial nectar was made to 15% concentration by dissolving equal amounts of fructose and glucose in boiling water, which was then allowed to cool. The sugar concentration of the Pinot blanc grapes was measured on 18<sup>th</sup> April 2002 at 18.2%.

For each visit by a bird to the table time of arrival and departure were recorded. Whether the bird ate grapes, whether it then moved to the nectar, and how many seconds lapsed between arriving at the table and moving to the nectar were also recorded. Data were then analysed by non-parametric Mann-Whitney U test (U) and  $\chi$  square using Genstat6 and Minitab software.

## **Results and discussion**

From the three locations in the Bentwood Vineyard, 11 sessions successfully recorded 165 visits by silvereyes. Two sessions recorded starlings only, several sessions recorded no birds at all. In the orchard five sessions recorded 134 silvereye visits successfully.

The latency (length of time in seconds) for a bird to move to the novel pots, either from grapes on the table or from first alighting, was significantly shorter for the orchard (resident) population than for the vineyard one (Figure 1,  $U=4814.5$ ,  $p=0.016$ ). The percentage of birds that fed from novel pots was greater in the orchard population than in the vineyard population (Figure 1,  $\chi^2=1.089$ ,  $df=1$ ,  $p=0.29$ ).

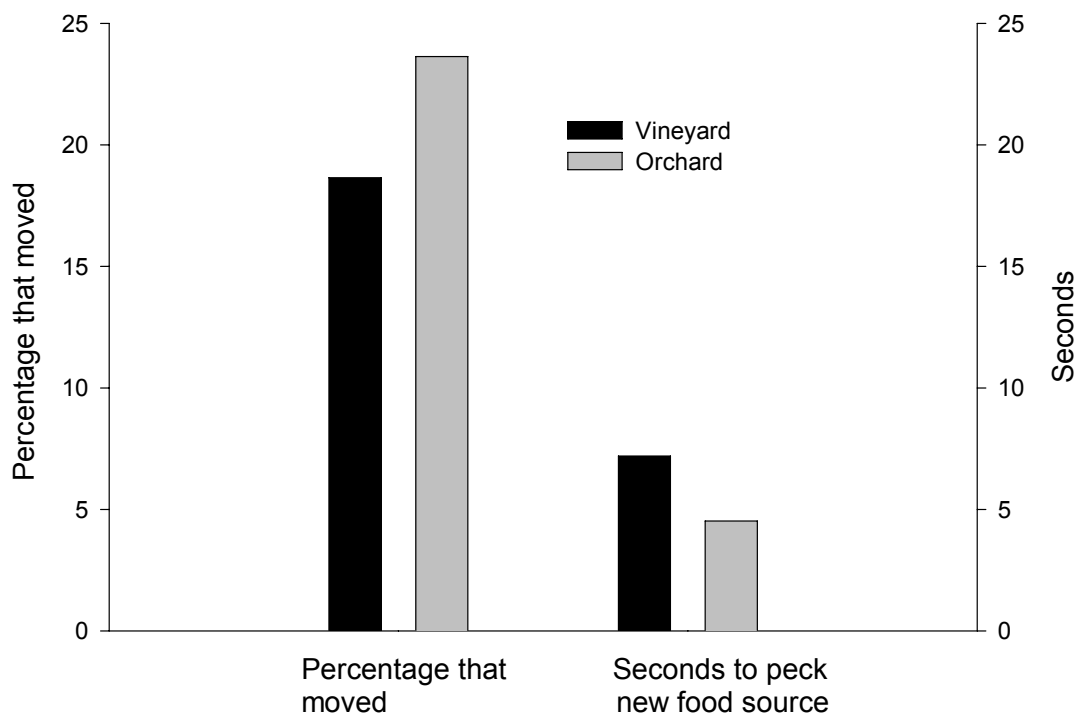


Figure 1. Fewer silvereyes from a vineyard population approach a new food source. Those that do take significantly ( $p=0.016$ ) longer than an orchard population.

The hypothesis that vineyard silvereyes are more neophilic than orchard silvereyes is rejected. In fact they proved significantly more neophobic. The two populations exhibited different latency in behaviour, and it is clear that the two populations were not identical. Suitability of site as a habitat, which includes factors such as food resources for the young,

can act as feedback that causes minor genetic changes, which can lead to major behavioural change (Klopfer and Ganzhorn, 1985). The implications for control of vineyard silvereyes are that behavioural patterns are becoming stereotypical, evidenced by more neophobic responses to a novel food source than those exhibited by a different population of the same species.

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## **Appendix II**

QuikCARD survey card for bird damage in vineyards.

<http://www.lincoln.ac.nz/v&o/pubs/quikcard.pdf>



## VINEYARD BIRD-DAMAGE 'QuickCARD'

### INTRODUCTION

The Winegrowers Association, in collaboration with Lincoln University, is implementing a 'QuickCARD' procedure to enable growers to assess bird damage in their vineyards\*.

Filling out the QuickCARDS is a way to help you keep a record of bird problems as they develop in your vineyard each year.

Standardising the way this information is collected will also, over time, help us to determine the broad extent of bird damage in the nation's vineyards. This information will help fund and guide future research efforts to develop better bird management techniques.

### WHAT WE WOULD LIKE YOU TO DO

- 1) **Select one or more blocks of your vineyard that you would like to monitor for bird damage.**
- 2) **Approximately one week before harvest, fill out a QuickCARD damage assessment on each of those blocks.**
- 3) **At the end of each season, keep the original QuickCARDS for your own records and post a copy of each to the Winegrowers' Association in Auckland, or fax to 03 325 3843.**

### WHAT WE WILL DO FOR YOU

- 1) The QuickCARDS received by Winegrowers will be analysed each year to determine broad regional trends in bird damage. (At no stage will any information that identifies your individual vineyard be made public)
- 2) You and the other growers who have sent in QuickCARDS will receive back a small annual newsletter containing a summary of these regional analyses, progress reports on current bird research, and any information on any useful methods for damage mitigation that has become available during the year.

**QuickCARDS and instructions for their use are attached.**

If you have any queries please contact:

Valerie Saxton, Centre for Viticulture and Oenology, PO Box 84, Lincoln University.  
[saxtonv@lincoln.ac.nz](mailto:saxtonv@lincoln.ac.nz)

You can also check out the 'Frequently Asked Questions' page on our website:  
[www.lincoln.ac.nz/v&o/birddamage.htm](http://www.lincoln.ac.nz/v&o/birddamage.htm)

\*After Saunders and Tracey, Orange Research Institute, NSW, Australia

## QuickCARD INSTRUCTIONS

- 1) **One week prior to harvest, select one or several blocks within your vineyard that you would like to score.** For the purposes of scoring a ‘block’ means all the adjacent rows of vines within some kind of well-defined physical boundary (i.e, fences, tracks or shelterbelts). There may be several varieties or vine ages within a block. For *scoring*, ignore any perceived differences in damage levels within the block.
- 2) **Fill out a separate QuickCARD for each block.** Please be sure to fill out ALL the information asked for on the card, and briefly sketch a map of the block’s layout including its edges, sides and interior (see below) and any trees, water, buildings, neighbouring activities or other features that may affect bird movements. *On the map* indicate areas where damage seems heavier, or identified as due to a particular bird species.
- 3) **Record four separate estimates (see below) of % grape damage in the block – one for each ‘side’ and one for each ‘end’.**

**This is the first stage of assessment. If any of these four damage estimates is higher than 5%, then proceed to a second stage, which is a fifth estimate from the ‘interior’ of the block.**

The sides of the block are the two outermost rows on each side of the block.

The ends of the block are the first two, and last two, vines in each of the intermediate rows between the sides.

The remaining vines are all considered to be within the interior of the block.

Do not worry about any changes in grape variety within the block.

- 4) **Each of these estimates of % damage is obtained by scoring 1 bunch from each of 10 vines.**

On each side of the block, select a vine at random along each of the two outer rows. Inspect one bunch selected at random from it and each of the next four vines (i.e., giving a total of 10 bunches on each side).

At each end of the block, inspect one bunch on the end vine of a random row and one bunch on the second-to-last vine on that row. Repeat this on the four adjacent rows (i.e., giving a total of 10 bunches at each end).

The damage score for each bunch is *your estimate* of what proportion of the berries on that bunch have been either removed or pecked by birds.

- 5) **Add up the bunch scores separately for each side, divide this by 10, and enter these four damage estimates on the QuickCARD. This completes the first stage.**

*e.g. At the north end of the vineyard the bunches you inspect are scored as follows:*

*10%, 5%, 0%, 20%, 15%, 30%, 10%, 0%, 0%, 20%*

*Summing these gives 110. Dividing this sum by 10 gives a damage estimate of **11%**.*

- 6) **If the damage to any side or edge of the block exceeds 5% proceed to the second stage: which is to estimate damage from the interior of the block. Select a total number of samples from the table below according to the highest damage level from the first stage (interior damage is expected to be lower.)**

Table 1. Total number of vines to be inspected in the interior during Stage 2

%Damage (Stage1)		5-10	20	30	40	50	60	70	80	90+	
No. of vines		10	30	50	65	70	65	50	30	10	

- 7) **In the interior of the block, choose adjacent rows more-or-less randomly and adjacent vines randomly along each row – inspect one bunch from each of these vines. The total number of samples (rows x vines) should equal the sample number selected from the table. Sum all the bunch scores and divide by the number of samples. Enter this %figure, plus the number of bunches inspected, on the QuickCARD. This completes the second stage.**

- 8) **Mail the completed duplicate QuickCARDS to IWP, Winegrowers of New Zealand, PO Box 90276, Auckland Mail Centre, Auckland, or fax as indicated on page 1.**



**QuickCARD**

Vineyard Name.....block name.....size(Ha).....  
 Address.....Phone.....  
 Assessment date..... Cultivar 1..... °Brix at assessment date. ....  
 Cultivar 2.....°Brix ..... Cultivar 3.....°Brix .....Projected date of harvest.....

<b>Edge:</b>  five vines from two rows  %  <b>Damage</b>  .....	<b>Side: two vines from ends of five rows % Damage</b>  .....	<b>Edge:</b>  Five vines from two rows  %  <b>Damage</b>  .....
	<b>Interior rows (if sampled) % damage from ..... vines:</b>  .....	
	<b>Side: two vines from ends of five rows % Damage</b>  .....	

**Block map: Please indicate cultivars, trees, power lines, water, buildings, adjacent activities, and areas of greater damage. Also indicate areas where type of damage suggests a particular bird species.**

**Please return you completed QuickCARDS.... As soon as they are complete to :**

IWP, QuickCARD, Winegrowers of New Zealand, PO Box 90276, Auckland Mail Centre, Auckland or fax to 03 325-3843