Simazine Treated Mulch an Integrated Management Tool for Vinifera Grape

(Vitis vinifera L.) Production

Dissertation

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By

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Abstract

European type grapes (*Vitis vinifera* L.) account for over 90 % of all grapes grown in the world. However, an extra so-called "winter-hilling" practice is required for winter protection of grafted vinifera grapes in northern states including Ohio. Winter hilling consists of two tillage activities. The first is to mound soil up to cover the graft union in fall for protection from lethal cold temperatures during winter, and the second is to remove the mounded soil in the next spring for prevention of rooting from the vinifera scion. Due to the intensive soil disturbance associated with winter hilling, any vineyard along a hillside is facing elevated risk for severe soil erosion. Along with soil erosion, any chemicals applied into the vineyard, such as fungicides, insecticides, herbicides and fertilizers, are also more likely to runoff during heavy rainfall. Loss of the fertile surface soil harms grapevines, and offsite movement of chemicals pollutes the environment. Therefore, there is a great research need to find a potential substitute to winter hilling.

In this study, we used simazine treated mulches (STM) to replace winter hilling and explored the following aspects: 1) weed population shifts as affected by winter hilling, 2) the efficiency of STM on weed control and herbicide resistant weed management, 3) grape production variables as affected by STM, including winter protection, nitrogen nutrition, fruit yield and quality, and 4) the potential of mulches as a mitigation tool to reduce chemical offsite movement.

The results indicated that winter hilling increased the number of weed species and density in vinifera vineyards relative to vineyards that had not been hilled. STM overall provided outstanding weed control, largely due to the season-long weed suppression by mulches. Pre-winter application of simazine herbicide resulted in little residual activity by the next spring, and simazine reapplication made in June controlled weeds but was not sufficient for the balance of the growing season. Triazine resistant (TR) common lambsquarters became more prevalent in response to simazine treatment, contributing to the low efficiency of simazine on weed control. However, simazine treated bark suppressed common lambsquarters and did not result in accumulation of a more TR population. These results suggested that STM could be used as a tool for management of TR weeds in vineyards. STM protected the graft union through winter and conserved soil moisture. The effect of STM on grape yield varied on different varieties from year to year, but overall STM either had no effect or increased grape yield. The fruit quality of grape was not affected by STM with the exception that sugar content in juice of Auxerrois was lower than un-mulched treatment one of two years. Lastly, the in-lab simulation trial indicated that straw reduced simazine leaching and runoff by 40 % and 68 %, respectively, after intensive simulated rainfalls. These results indicated that mulches could be an effective tool to mitigate chemical offsite movement from the vineyard. In conclusion, STM is an integrated management tool with multiple beneficial impacts including winter protection, soil conservation, weed control and chemical runoff mitigation.

Dedication

Dedicated to the students at The Ohio State University

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Fields of Study

Major Field: Horticulture and Crop Science

Table of Contents

Abstractii
Dedicationiv
Acknowledgmentsv
Vitavi
List of Tablesix
List of Figuresxii
Chapter 1: Introduction1
Chapter 2: The Effect of Herbicides and Cultural Practices on Weed Communities in
Vineyards, an Ohio Survey12
Chapter 3: The Effect of Simazine Treated Mulches on Weed Control in an Auxerrois
Vineyard
Chapter 4: Dynamics of Common Lambsquarters (Chenopodium album L.) Biotypes in
Two Vineyards Determined with a Modified Molecular Method Targeting the
<i>psb</i> A Gene65
Chapter 5: Soil Moisture, Winter Protection, and Grape Yield and Quality as Affected by
Simazine Treated Mulches
Chapter 6: The Effect of Straw Mulch on Simulated Simazine Leaching and
Runoff119

Bibliography	
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List of Tables

Table 2.1. Relative abundance (RA), frequency (F), field uniform (FU), mean field	
density (MFD), and mean occurrence field density (MOFD) of weeds in Ohi	0
vineyards. The number in parentheses is the ranking number of this weed	
species based on RA value	.31
Table 2.2. The effect of herbicide management program on density of 20 dominant wee	ed
species based on the Relative Abundance statistic	34
Table 2.3. The effect of hilling practice on weed species density	.35
Table 3.1. Relative abundance of weed species in response to two-years of simazine	
treated mulches at Kingsville, Ohio	.57
Table 3.2. Weed species observed at the Auxerrois vineyard at Kingsville, Ohio	.58
Table 3.3. The effect of simazine treated mulches on total weed density in the Auxerroi	is
vineyard in 2008 and 2009 at Kingsville, Ohio	.59
Table 3.4. The effect of simazine treated mulches on annual broadleaf weed density in	the
Auxerrois vineyard in 2008 and 2009 at Kingsville, Ohio	60
Table 3.5. The effect of simazine treated mulches on grass weed density in the Auxerro	ois
vineyard in 2008 and 2009 at Kingsville, Ohio	61

Table 3.6. The effect of simazine treated mulches on perennial broadleaf weed density in
the Auxerrois vineyard in 2008 and 2009 at Kingsville, Ohio62
Table 3.7. Interactions of mulch and simazine on total, annual broadleaf, perennial
broadleaf, and grass weed control in 2008 and 200963
Table 4.1. The effect of simazine treated mulches on total common lambsquarters
density87
Table 4.2. The effect of simazine treated mulches on percentage of triazine resistant (TR)
common lambsquarters
Table 5.1. The effect of simazine treated mulch on soil moisture in 2008
Table 5.2. The minimum temperature (°C) recorded during winter months (November to
June)110
Table 5.3. Winter injury assessment of Pinot gris at Wooster in 2009111
Table 5.4. The effect of simazine treated mulches on scion rooting at Kingsville,
Ohio112
Table 5.5. The effect of simazine treated mulch on total nitrogen content in leaf petioles
of Seyval at Wooster, Ohio and Auxerrois and Pinot gris at Kingsville,
Ohio113
Table 5.6. The effect of simazine treated mulch on yield and quality of Auxerrois grapes
harvested in 2008 and 2009 at Kingsville, Ohio114
Table 5.7. The effect of simazine treated mulch on yield and quality of Seyval harvested
in 2008 and 2009 at Wooster, Ohio115

Table 5.8. The effect of simazine treated mulch on yield and quality of Pinot gris at
Kingsville, harvested on 10-19-2009116
Table 5.9. The effect of simazine treated mulches on cluster number, cluster size and
pruning weight of Seyval Blanc in 2009 at Wooster117
Table 6.1. The effect of straw mulch and simazine application rate on simazine recovered
from leached water, soil and straw after 12 simulated, 20 mm, rainfalls146
Table 6.2. The effect of straw mulch on daily simazine runoff in response to a simulated
10 mm rainfall147
Table 6.3. The effect of straw mulch on simazine recovered from runoff water, soil and
straw after 6 simulated, 10 mm, rainfalls148
Table 6.4. The effect of simazine application rate on daily simazine runoff in response to
a simulated 10 mm rainfall149

List of Figures

Figure 1.1. Soil erosion occurred in the research vineyard at the Ohio Agricultural
Research and Development Center (OARDC) in Wooster11
Figure 2.1. Geographical distribution of surveyed vineyards across the State of Ohio30
Figure 3.1. The interaction of simazine rate and mulch on total weed control in July (a)
and August 2008(b), and on broadleaf weed control in July of 2008 (c) and
2009 (d)
Figure 4.1. The effect of simazine treated-corn stubble, -wheat straw, simazine alone, and
simazine treated-shredded wood bark on common lambsquarters on July 07,
2007 from left to right, respectively
Figure 4.2. Polymorphism in PCR products targeting the <i>psbA</i> gene in TR and TS
common lambsquarters using primers psbF, psbR, psbSF, and psbRR84
Figure 4.3. Alignment of the <i>psbA</i> gene sequences in control TS common lambsquarters,
the putative TR common lambsquarters (samples 1 to 3), and that of the
known TS and TR biotypes86
Figure 5.1. Monthly maximum, minimum and average temperature recorded from
November 2007 to July 2008 at Kingsville. A similar pattern was observed at
Wooster

Figure 6.1.	The effect of straw mulch and simazine application rate on weekly simazine
	leaching (a) and cumulative simazine leaching (b)143
Figure 6.2.	Daily suspended, dissolved and total simazine runoff, averaged across straw
	mulched and bare soil treatments and herbicide rate, following a simulated 10
	mm rainfall144
Figure 6.3.	The effect of straw mulch on daily soil erosion in response to a simulated 10

mm rainfall145

Chapter 1: Introduction

Grape Production in Ohio. The Ohio grape industry has been growing rapidly during the past decade. The number of licensed wineries almost doubled in Ohio from 1997 to 2004 (Dami et al. 2005). There were 125 wineries by 2009, making Ohio one of the top 10 wine-producing states in the US. According to Ohio Grape Industries Committee (OGIC 2010), the total wine produced by Ohio wineries was 850,000 gallons in 2007.

In contrast to the rapid increase in the number of wineries, the supply of wine grapes has not kept up with the demand due to the low percentage of wine grape acreage in Ohio. Currently, three types of grapes are grown in the Midwest: American grapes (e.g. *Vitis labrusca*), European grapes (*V. vinifera*) and the interspecies hybrid between American and European grapes. Although Ohio wineries are making wines primarily from vinifera and hybrid grapes, the majority of grape acres (1280) in Ohio in 1999 were American grapes for making grape juice, and only 275 acres of grapes were European varieties or hybrids (USDA, 2010). As a result, Ohio wine grape growers cannot fulfill the needs of the increasing tonnage required by Ohio wineries. A vineyard expansion assistance program funded by USDA was launched by OGIC in 2009, proposing to increase the

vineyard acreage by 10 % (220 acres) in the following five years. Altogether, driven by this stimulus program as well as the increasing demand for Ohio wine grapes, vinifera grape acreage will rapidly grow in the near future.

Winter hilling in vinifera grape production. Due to its sensitivity to phylloxera, a soil insect that attacks the root system, grafting a vinifera scion onto a phylloxera-resistant rootstock has been extensively adopted. Moreover, most vinifera grapes are cold-tender and appropriate winter protection is needed in the Midwest, including Ohio. The so-called "winter hilling" method is routinely used to address the cold-tender problem (Dami et al. 2005). Winter hilling comprises two tillage activities in the vineyard: the first to mound soil to cover the graft union in late fall for protection from cold temperatures, and the second to remove the mounded soil from the graft union in spring in order to prevent vinifera scion rooting and subsequent phylloxera infection.

However, winter hilling can render soils susceptible to severe erosion and thereby threaten the long-term productivity and profitability of a vineyard. We observed that the top 8 to 12 cm layer of soil in the soil-hilling zone under the trellis of an OARDC research vineyard established in 1994 had eroded by 2009 (Figure 1.1). This vinifera vineyard had a slope of less than 5 degrees. Considering that many vineyards in Ohio are located in hilly areas, the threat of long-term soil erosion is a significant challenge to a sustainable production of Ohio vinifera grapes.

According to USDA (2009), a national average of 180 kg of pesticide active ingredients were applied to every hectare of vineyard, making grape a very chemical-

2

intensive crop. It is probable that extensive runoff of applied pesticides would occur concomitant with soil erosion from the under-the-trellis zone. Winter hilling is also likely to contribute to increased pesticide leaching (Gish et al. 1995), since most vineyards are established on well drained soils.

Weed control in the vinifera vineyard. Besides diseases and insects, weeds are another problem that grape growers have to manage in order to achieve a profitable yield. Weeds can directly compete with grape for nutrients, sunlight and water, leading to loss in yield. Moreover, weeds can function as shelter and food for various pests and pathogens. A dense weed population can inhibit air circulation in the vineyard, facilitating grape disease development (Dami et al. 2005).

Because grape is sensitive to many herbicides, weed control relies on a limited set of available herbicides. As a result most vineyards have been treated repeatedly, sometimes for decades, with only a few herbicides or a single product. This practice has imposed a heavy selection pressure for herbicide resistant weeds. Triazine resistant common groundsel (*Senecio vulgaris*) was first identified in a vineyard in Switzerland in 1982, following several years of extensive use of simazine (Heap, 2010). Polos et al. (1985) found triazine resistant horseweed (*Conyza canadensis*) in Hungarian vineyards. In 2003, buckhorn plantain (*Plantago lanceolata*) was identified as glyphosate resistant in a South Africa vineyard (Heap 2010). Moreover, multi-herbicide resistant weeds also have shown up in vineyards. Polos et al. (1988) reported horseweed that was resistant to both simazine and paraquat, and in a vineyard from California, hairy fleabane (*Conyza*)

bonariensis) was found to resist both glyphosate and paraquat (Heap 2010). Grapes are also very sensitive to herbicides registered in vineyard; within the short list of recommended herbicides there are many restrictions on grape age or crop dormancy (Bordelon et al. 2007). Growers often violate these restrictions, resulting in crop injury.

Winter hilling may create a favorable condition for certain weeds to establish (Jiang et al. 2008). Tuesca et al. (2001) reported that broadleaf species had higher populations under conventional tillage than non-tillage. Similarly, our survey of weeds growing under the trellis in 31 Ohio vineyards also indicated that winter hilling contributed to a more severe weed problem than occurred in non-hilled vineyards of American or French hybrid grapes (Jiang et al. 2008). Winter hilling further complicates weed control by negatively interacting with soil-applied herbicides. Since many vineyards have a soil with good water drainage, leaching of preemergence herbicides applied to soil is likely increased by tillage (Gish et al. 1995). Along with soil erosion due to winter hilling, herbicide runoff is also enhanced (Montgomery 2007). Therefore, the off-target movement of herbicide is expected to reduce herbicide weed control efficiency in vinifera vineyards.

Why herbicide treated mulches? Using organic mulches for winter protection has been widely adopted in strawberry production (Askew and Smith 2008). Zabadal (2003) compared a mulch of wheat straw to winter hilling for winter protection in a vinifera vineyard in Michigan and observed that straw protected grapes as effectively as winter hilling. Mulching also has a great potential to address soil erosion associated with winter hilling. Maass et al. (1988) and Döring et al. (2005) indicated that mulch could reduce soil erosion by more than 90 % under field conditions.

Mulch is also an effective way to control weeds. Since many weeds need light to trigger germination (Wesson and Wareing, 1967), the shading provided by mulch can significantly reduce seed germination. Mulches also prevent many germinated seedlings from reaching the surface because their energy reservoir is insufficient. Besides physical shading and suppression, allopathic compounds associated with organic mulches can further improve weed control. Some allelopathic compounds include benzoxazolinones released from rye (*Secale cereale*) straw (Barnes and Putnam 1987), and sorgoleone released from residues of *Sorghum* species (Netzly and Butler 1986; Nimbal et al. 1996).

Synergistic effects on weed control between soil active herbicides and organic mulches have been reported to improve upon the physical weed control provided by the mulch. Atrazine treated wheat straw provided a better weed control than straw and herbicide alone (Prihar et al. 1975). Case and Mathers (2006) reported that flumioxazin-treated hardwood or pine nugget mulch provided better weed control than the herbicide or mulch alone, and they attributed this to a slow release of herbicide from the mulch. Teasdale et al. (2005) observed a synergistic effect between metolachlor and hairy vetch residue on the emergence of various weeds, including smooth pigweed (*Amaranthus hybridus*), common lambsquarters (*Chenopodium album*), giant foxtail (*Setaria faberi*) and velvetleaf (*Abutilon theophrasti*).

Simazine is the only triazine herbicide registered in grapes. As a photosynthesis inhibitor, simazine controls a broad range of geminating annual broadleaf and grass

5

species. Because of its weed control efficiency and low cost, simazine has been used extensively in vineyards since the early 1960s (Elmore and Lange 2008). Simazine was the second most frequently used preemergence herbicide in California wine grape vineyards: 37.6 % were treated with simazine from 2002 to 2005 (Elmore and Lange 2008). However, the extensive use has resulted in triazine resistant weeds in vineyards (Heap, 2010) as described previously and also contaminated groundwater. Spurlock et al. (2000) examined simazine contamination of groundwater in two counties in California where grapes were a major crop. They observed that 97 % of the wells (n=33) were contaminated by simazine. Therefore, a method for use of simazine is needed for grape production that does not contribute to contamination of surface and ground water.

The central hypothesis of my Ph.D. research was that using simazine treated mulches (STM) would provide outstanding weed control, winter protection, soil conservation and reduction in simazine offsite movement in vinifera vineyards. Our long-term goal is that STM will enable Ohio vinifera vineyards to be more sustainable, improving competiveness and profitability through these integrated benefits. The specific objectives of this study were to 1) characterize the weed problem in Ohio vineyards through a survey; 2) examine the effect of STM on weed control in vineyards; 3) determine the effect of STM on grape growth including winter protection, yield and quality; and 4) explore the potential of straw as a tool to reduce simazine offsite movement.

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Figure 1.1. Soil erosion occurred in the research vineyard at the Ohio Agricultural Research and Development Center (OARDC) in Wooster. The grape cultivar was Pinot gris, planted in 1994. Picture was taken on July 13^{th} , 2009. The unit for the left side of the ruler was inches (1 inch = 2.54 cm).

Chapter 2: The Effect of Herbicides and Cultural Practices on Weed Communities in Vineyards: An Ohio Survey

Linjian Jiang, Tim Koch, Imed Dami, and Douglas Doohan*

Thirty one Ohio vineyards were surveyed in 2004 to document weeds that persisted following weed control practices. Weeds were identified and density was determined during visits to each vineyard. Herbicide use history, grape varieties, and grape age were recorded during interviews with the growers. Data were analyzed by SAS 9.1 using the GLM model, and means were compared according to Student-Newman-Keuls (SNK) at the 0.05 level. Crabgrass, dandelion, pigweed, foxtail, fall panicum, clover, chickweed, common ragweed, smartweed, and oxalis were the most prevalent 10 weeds in Ohio vineyards based on relative abundance values. The frequency and density of crabgrass, dandelion, fall pancium, oxalis and common purslane were significantly higher in vineyards in which glyphosate was the only herbicide used than in vineyards where other herbicides were applied. The number of species and density were higher in vinifera

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vineyards that were had been hilled for winter protection than in vineyards that had not been hilled.

Nomenclature: glyphosate; chickweed, *Stellaria media* (L.) Vill. or *Cerastium fontanum* ssp. *vulgare* (Hartman) Greuter & Burdet; clover, *Trifolium repens* L. or *Trifolium pratense* L.; common ragweed, *Ambrosia artemisiifolia* L.; crabgrass, *Digitaria sanguinalis* (L.) Scop.; dandelion, *Taraxacum officinale* G.H. Weber ex Wiggers; fall panicum, *Panicum dichotomiflorum* Michx.; foxtail, *Setaria faberi* Herrm. or *Setaria pumila* (Poir.) Roemer & J.A. Schultes or *Setaria viridis* (L.) Beauv.; oxalis, *Oxalis corniculata* L.; pigweed, *Amaranthus hybridus* L. or *Amaranthus retroflexus* L.; smartweed, *Polygonum pensylvanicum* L.; vinifera grape, *Vitis vinifera* L.

Key words: Glyphosate resistant, vineyard, vinifera, weed survey.

Growing wine-grapes is a rapidly expanding industry in the United States. The number of licensed wineries in Ohio, Indiana, Michigan, Illinois, and Missouri, almost doubled from 1997 to 2004 (Dami et al. 2005). Ohio is one of the top 10 wine-producing states with more than 1.9 million liters produced every year (Ohio Grape Industries Committee, 2007). In surveys conducted during the previous ten years, Ohio grape growers have identified weeds as a major factor limiting vineyard productivity and expansion (The Ohio Grape Team unpublished data 1997). Similar rankings have been made by growers in other states as reflected by the research priorities published by the Viticulture Consortium East (2007). A recent survey of Ohio viticulturists showed that weeds were even more difficult to control than insects and diseases (Dami et al. 2006). Weeds can compete with grape for nutrients, sunlight, and water resulting in losses in yield. Weeds also serve as habitat for other pests; thereby contributing to damage by insects and diseases (Dami et al. 2005).

The nonselective herbicide glyphosate was licensed for use in orchard crops during the product's early commercial development and its use has been widely adopted by growers. In Ohio, glyphosate applications are a preferred weed management method of many grape growers because of the herbicide effectiveness and lack of soil activity. However, the emergence of glyphosate-resistant biotypes is a concern that has not escaped the viticulture industry. Such concerns have escalated since Roundup Ready[®] soybean, corn and cotton crops have attained dominance in the US market (Duke 2005). New glyphosate-resistant biotypes continue to be reported. The current list of glyphosate resistant species is common waterhemp (*Amaranthus rudis* Sauer), common ragweed (*Ambrosia artemisiifolia* L.), giant ragweed (*Ambrosia trifida* L.), hairy fleabane (*Conyza bonariensis* (L.) Cronq.), horseweed (*Conyza canadensis* (L.) Cronq.), goosegrass (*Eleusine indica* (L.) Gaertn.), wild poinsettia (*Euphorbia heterophylla* L.), Italian ryegrass (*Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot), rigid ryegrass (*Lolium rigidum* Gaudin), buckhorn plantain (*Plantago lanceolata* L.), Johnsongrass (*Sorghum halepense* (L.) Pers.) (Heap 2006) and Palmer Amaranth (*Amaranthus palmeri* S. Wats.) (Culpepper et al. 2006). Of these, hairy fleabane, horseweed, goosegrass, Italian ryegrass, rigid ryegrass, and buckhorn plantain have been found in glyphosate treated orchards or vineyards (Heap 2006). Thus fear among Ohio grape growers that glyphosate resistance might develop or spread into local vineyards is strong. Determining the prevalence of potentially resistant biotypes was an impetus to conduct the survey reported in this work.

As previously described weed management is one of the most serious problems encountered by Ohio grape growers. Currently, three types of grape are grown in the Midwest region: American, French hybrid and European (*Vitis vinifera* L.) also referred to as vinifera grapes. Vinifera grapes are required for production of high-value wines and most new vineyards in the state are planted to vinifera varieties. The vinifera grape is more difficult to grow because of its cold- and grape phylloxera- sensitivity. American and French hybrid grape varieties are more tolerant of both colder temperatures and phylloxera than are vinifera. To protect vinifera grapes from the aphid-like phylloxera it must be grafted on American grape rootstocks, which are resistant to this pest. However, the graft union must be protected from low winter temperatures so that it can be used to generate a new vine if the grape trunk is damaged. Currently, winter protection is achieved by covering the graft union with several inches of soil in autumn. The mounded soil must be removed each spring to prevent scions from forming roots (Dami et al. 2005), or the plant will lose phylloxera resistance. The annual process of building an insulating hill of soil in autumn and removing the hill in spring may complicate weed management for growers and affect weed community structure; however, the impact has not been measured. Excessive soil tillage is known to change soil structure (Shepherd et al. 2001), and may dilute soil-active herbicides, bring weed seeds close to the soil surface and stimulate weed seed germination.

In 2004, we surveyed 31 Ohio vineyards with 3 objectives in mind: (1) determine which weeds persisted after control practices were completed; (2) compare weed communities between hilled and non-hilled vineyards; and (3) detect weed species potentially resistant to glyphosate.

Materials and Methods

Questionnaires on vineyard weed problems and weed management methods were mailed to 90 Ohio grape growers in 2004, and 36 responses were received. We visited these 36 vineyards and selected 31 for data collection (Figure 2.1). Survey vineyards were located throughout the state, but somewhat clustered in 3 geographic regions: Lake Erie area that is part of the Lake Erie appellation, Southwest Ohio that is part of Ohio River Valley appellation and Central Ohio that is between the two appellations. Vineyards were surveyed from July to September; about 2 to 3 weeks after the last herbicide application. Each grower provided us with a block of grapevines ranging from 0.33 to several acres that contained vines planted at the same time, had received homogeneous management for several years and was representative of the general weed problems in their vineyard.

Weeds under the grape trellis were identified and counted in 20 quadrats (25×25cm) dropped at random along two diagonal line transects in each field. Weeds showing severe injury likely to cause death in response to herbicide treatment were not counted. It was not always possible to clearly differentiate recently emerged seedlings of certain species. In such cases a general common name was used. For example data tabulated as foxtail may include giant foxtail, yellow foxtail and green foxtail. Information on herbicide applications, vineyard age, grape cultivar, and cultural practices was gathered by interviewing growers.

Weed frequency, field uniformity, mean field density, mean occurrence field density, relative frequency, relative field uniformity, relative mean field density, and relative abundance were calculated according to the method of Thomas (1985). Frequency of a species was the number of the fields where this species occurred expressed as a percentage of the total number of surveyed fields (31 vineyards). Field uniformity (FU) of a species was the number of quadrats where this species occurred expressed as a percentage of all the surveyed quadrats (31×20). Mean field density (MFD) refers to the number of individuals of a species per square meter and was calculated by totaling seedling number of a species in each field and dividing by the total number of fields (31

17

vineyards). Mean occurrence field density (MOFD) refers to the density when only occurrence fields are included in the area determination. Throughout this paper relative abundance (RA) is frequently reported. Relative abundance summarizes frequency, field uniformity and mean field density into one value to facilitate comparisons across species. Relative abundance for a species is the sum of relative frequency, relative field uniformity and relative mean field density for that species. The relative values for frequency, field uniformity, and mean field density for a species express those statistics for the species as a percentage of each variable summed across all species. For example, relative frequency of a species was the frequency of this species over the sum of the frequency of all species in this survey. Weed management strategies were summarized according to the prevailing herbicides (or lack of) used in each vineyard: no herbicide, glyphosate only, glyphosate + preemergence herbicide, and paraquat + preemergence herbicide. Preemergence herbicide was either simazine (6-chloro-N,N'-diethyl-1,3,5triazine-2,4-diamine), diuron (N'-(3,4-dichlorophenyl)-N,N-dimethylurea) or dichlobenil (2,6-dichlorobenzonitrile). Data were analyzed by SAS 9.1 using the GLM model. Means were compared by Student-Newman-Keuls (SNK) at the 0.05 level. The main factors were weed management strategies, tillage intensity (hilled or non-hilled), and geographic regions of Ohio.

Results and Discussion

Weeds Surviving Control Practice In Ohio Vineyards. Fifty-three weed species were identified in the 31 vineyards included in this survey (Table 2.1). The top 10 weeds in relative abundance were crabgrass (*Digitaria sanguinalis* (L.) Scop.), dandelion (Taraxacum officinale G.H. Weber ex Wiggers), pigweed (Amaranthus hybridus L. or Amaranthus retroflexus L.), foxtail (Setaria faberi Herrm. or Setaria pumila (Poir.) Roemer & J.A. Schultes or Setaria viridis (L.) Beauv.), fall panicum (Panicum dichotomiflorum Michx.), clover (Trifolium repens L. or Trifolium pratense L.), chickweed (Stellaria media (L.) Vill.), common ragweed (Ambrosia artemisiifolia L.), smartweed (*Polygonum pensylvanicum* L.) and oxalis (*Oxalis stricta* L.) were the most prevalent. Respective relative abundance (RA) values for these weeds were 44.2, 25.4, 17.7, 17.1, 14.3, 11.6, 11.3, 10.6, 10.3, and 9.3 (Table 2.1). Although the ranking of weed species differed in the lists based on field uniformity (FU), mean field density (MFD), or mean occurrence field density (MOFD), crabgrass consistently was at the top. This result indicates that crabgrass is clearly the most important grass weed in Ohio vineyards in early summer.

Certain species were unevenly distributed across the state (data not reported). Annual bluegrass, common chickweed, dandelion, common groundsel, and quackgrass were more prevalent in the Lake Erie appellation than in central Ohio or in the Ohio River Appellation. Common purslane and clover were more prevalent in central Ohio; whereas, crabgrass and prickly sida were more common near the Ohio River in the south west corner of the state.

Examining the entire data set indicates that broadleaf weeds are more prevalent than grasses in Ohio vineyards; for example, there are 13 broadleaf species with a RA value of 116, 6 grass species with a summed RA value of 101, and 1 nutsedge species among the top 20 weeds (Table 2.1). Dandelion and pigweed species were the two most abundant broadleaf weeds. Dandelion occurred in 87.1 % of the total surveyed vineyards and 28.2 % of the total surveyed quadrats, and had similar MFD and MOFD values of 16.7 and 19.1 plants/m², respectively (Table 2.1). The characteristic of dandelion to flower in spring and in autumn (Stewart-Wade et al. 2002) may contribute to the species ability to colonize bare strips under the grape trellis in the fall when preemergence herbicide residues in the soil are low or non-existent. Also, at that time of year growers may be less vigilant. Dandelions' regular occurrence in turf, coupled with tight restrictions on the use of 2,4-D in vineyards (Dami et al. 2005; Stewart-Wade et al. 2002), probably further contributes to ready growth in the grass-covered alleys between rows and dispersion to bare ground maintained under the grape plants. In contrast to dandelion, pigweed had a much higher MOFD (42.2 plants/m²) than MFD (19.1 plants/m²), suggesting that pigweed flourished in the vineyard where it occurred (Table 2.1). Significantly higher pigweed density in the herbicide free (no-herbicide) vineyards (Table 2.2) no doubt contributed to the difference between MFD and MOFD, as well as the relatively low frequency and FU. These results indicate that herbicide-based weed management programs for vineyards are controlling pigweed effectively (Table 2.2),

even though resistance to different herbicides including ALS inhibitors, photosystem II inhibitors and glyphosate has been reported (Heap 2006).

Annual weeds dominated in Ohio vineyards, with the significant exemption of dandelion. Other than crabgrass the density of annual weeds in occurrence fields (MOFD) was much higher than mean field density (MFD), suggesting that site-specific and/or management specific factors were contributing to survival of most annual weed species. Annual bluegrass, prickly sida (Sida spinosa L.), common purslane, groundsel (Senecio vulgaris L.) and eastern black nightshade (Solanum ptychanthum Dunal) had MOFD values that were more than 10 plants/ m^2 and at least three times higher than MFD. For example annual bluegrass had a MOFD of 30 plants/ m^2 , and a MFD of 8.7 plants/ m^2 (Table 2.1). The low frequency (29 %) and FU (5.3 %) contribute to the difference between MOFD and MFD. Unlike pigweed, no significant difference was shown for annual bluegrass control using different herbicide management programs (Table 2.2), suggesting that herbicide application did not contribute to the difference between MOFD and MFD. In contrast, herbicide application could contribute to the difference between MOFD and MFD for fall pancium, which had a significantly higher density when glyphosate was applied alone (Table 2.2). Some perennial weeds, such as quackgrass, nimblewill (Muhlenbergia schreberi J.F. Gmel.) and wirestem muhly (Muhlenbergia frondosa (Poir.) Fern.) also had a much higher MOFD than MFD (Table 2.1). Quackgrass had a significantly higher density in herbicide-free vineyards than in other vineyards (Table 2.2); which contributed to the difference between MOFD and MFDD. Wirestem muhly was observed to occur in only one vineyard, where paraquat combined with a

preemergence herbicide was applied twice per year. As pointed out by Czapar and Fawcett (1997), wirestem muhly has become problematic in some regions of the North Central states (Ohio included). This result suggests that wirestem muhly should be watched closely in vineyards with similar herbicide management program.

Effect Of Weed Management Programs On The 20 Most Abundant Weed Species.

Analysis of the effect of management on the weed community was restricted to the 20 most abundant species for which the effect was relatively clear (Table 2.2). Since an important objective of this survey was to detect prevalence of potentially glyphosate resistant biotypes, the glyphosate alone program was the primary focus.

Crabgrass, dandelion, clover, fall pancium, oxalis and common purslane (*Portulaca oleracea*) had a significantly higher density when glyphosate was used alone than with other herbicide-based management programs. Resistance might explain the higher density of these weeds when glyphosate was used alone; however, confirming resistance was beyond the scope of this survey, and it has not been reported for these species elsewhere (Heap 2006). It is likely that other factors contribute to survival of these weeds in vineyards where glyphosate is used exclusively. For instance seedling establishment and reproduction during intervals between glyphosate applications may enable summer annuals to perpetuate. Similarly, dandelions may establish in autumn after the final glyphosate treatment is applied and flower in spring before weed control activities commence. However, the observed relationship between these weeds and glyphosate-only weed control suggests that glyphosate resistance may be developing in these weed
species and justifies close monitoring in the future. One in three weed scientists surveyed by Culpepper (2006) thought grasses would increase in response to a glyphosate-alone program in glyphosate resistant field crops. This speculation is supported by the results of this survey, which showed that crabgrass flourished in vineyards managed with glyphosate alone (Table 2.2).

Management of crabgrass, dandelion, clover, fall pancium, oxalis and common purslane, was greatly improved when glyphosate was used in conjunction with a preemegence herbicide (glyphosate + residual) (Table 2.2). This observation indicates that preemergence herbicides efficiently prevented new seedlings from developing between herbicide applications. This also supports our suggestion that the higher density observed under the glyphosate alone program was due to the germination of weed seeds after glyphosate application (Tharp and Kells 2002). However, this may not be true in every case because multiple glyphosate applications were used by some farmers under the glyphosate-alone program. Glyphosate was used 2 or more times per season in 7 of the 11 vineyards in which the herbicide was used alone. In contrast when glyphosate was used in combination with a preemergence herbicide (glyphosate + residual), glyphosate was applied only once per season in 8 out of 11 vineyards. Multiple applications of glyphosate per season is a known factor that increases the probability of resistance development (Heap 1997) and is likely to have decreased the ability for susceptible populations to establish and reproduce between glyphosate applications.

Other factors in addition to possible glyphosate resistance may contribute to the higher density of crabgrass, dandelion, clover, fall pancium, oxalis and common purslane. Ohio grape growers regularly mow the grass-covered alley-ways between rows of grapes. Mowing may benefit species such as dandelion and clover which flourish in the absence of a heavy turf canopy. Growers often neglect to control broadleaf weeds growing in the grass alleyways between the rows; thereby, providing a ready nearby source of seeds for reinfestation. Perennial root stocks of uncontrolled perennials such as morningglory and Canada thistle are likely to invade the trellis area where weed control has been maintained. This may also be a factor contributing to the invasion of the under-trellis area by crabgrass due to its creeping stems. This speculation is supported by Kim et al.'s (2002) survey, which showed crabgrass was a common weed that flourishes in turf in the northern region.

Weed Density In Hilled And Non-Hilled Vineyards. The data analysis revealed that 18 species had significantly different populations in hilled versus non-hilled vineyards (Table 2.3). Crabgrass, foxtail, and common purslane were more prevalent in non-hilled vineyards; 15 other species were more prevalent in hilled vineyards (Table 2.3). Invariably, hilling vineyards resulted in more severe weed problems.

Changes in weed communities and population density in response to different tillage practices have been observed by other researchers (Ball and Miller 1993; Tuesca et al. 2001). Tuesca et al. (2001) reported that broadleaf species had higher populations under conventional tillage than non-tillage. A similar result was also observed in this survey considering that the hilling practice constitutes a more intense tillage regime. Of the 15 species that had higher population densities in hilled vinifera vineyards; 13 species were

broadleaf weeds (Table 2.2). Tuesca et al. (2001) also found wind-dispersed species increased in no-till wheat/soybean rotation fields; however, in our survey wind-dispersed species such as dandelion had a higher population in hilled vineyards. Hilled soil may capture more windborne seeds during early spring when dandelion is dispersing. Increased tillage aerates soil and may provide a more suitable habitat for seedling establishment. During establishment of the hill deeply buried weed seeds are likely to be brought close to the soil surface where germination is most likely to occur. Simultaneously, concentration of residual herbicides in the soil is likely to be diluted. It is also possible that residual herbicides may leach more readily from the tilled soil in the hill (Gish et al. 1995).

Currently there are no widely accepted alternatives to hilling for winter protection of vinifera grapes. However, the practice may not be sustainable in some vineyards because it contributes to loss of soil structure and creates conditions conducive to soil erosion (Bhatt and Khera 2006; Kurtural 2005). The heavier weed problems observed among hilled vineyards in this study suggest further incentive to look for alternative methods to protect vinifera vineyards from winter injury.

This survey demonstrated that weed communities present in Ohio vineyards were affected by herbicide programs and by the hilling practice used in vinifera vineyards. This survey also indicated that crabgrass, foxtail, fall panicum, annual bluegrass, barnyard grass, and quackgrass were dominant grass species, and that dandelion, pigweed, clover, common ragweed and smartweed were the most prevalent broadleaf species in Ohio vineyards. Considering that improving weed control is a priority of viticulturist throughout the US, these results indicate that a focus is needed on these species. This survey also indicated that glyphosate resistance might play a role in the significantly higher populations of crabgrass, dandelion, fall pancium, oxalis and common purslane in those vineyards where glyphosate was applied alone. However, growers are likely to minimize both the competitive impact of these weeds and the probability of resistance by including a preemergence herbicide with glyphosate application. Several weed species had higher populations in hilled vineyards (vinifera), indicating that this practice along with the potential to increase likelihood of soil erosion, is incentive to develop alternate methods of winter protection.

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Figure 2.1. Geographical distribution of surveyed vineyards across the State of Ohio. Asterisk marks the location of survey vineyards.

RA Rank	Common Name	Scientific Name	RA	F	FU	MFD	MOFD
				Q	% ——	— plan	mts/m^2 —
(1)	Crabgrass	Digitaria sanguinalis (L.) Scop.	44.2	83.9	34.0	51.4	61.3
(2)	Dandelion	<i>Taraxacum officinale</i> G.H. Weber ex Wiggers	25.4	87.1	28.2	16.7	19.1
(3)	Pigweed	Amaranthus hybridus L. or Amaranthus retroflexus L.	17.7	45.2	13.2	19.1	42.2
(4)	Foxtail	<i>Setaria faberi</i> Herrm. or <i>Setaria pumila</i> (Poir.) Roemer & J.A. Schultes or <i>Setaria viridis</i> (L.) Beauv.	17.1	64.5	16.3	12.5	19.4
(5)	Fall panicum	Panicum dichotomiflorum Michx.	14.3	58.1	10.8	12.0	20.7
(6)	Clover	<i>Trifolium repens</i> L. or <i>Trifolium pratense</i> L.	11.6	51.6	12.6	6.0	11.7
(7)	Chickweed	<i>Stellaria media</i> (L.) Vill. or <i>Cerastium</i> <i>fontanum</i> ssp. <i>vulgare</i> (Hartman) Greuter & Burdet	11.3	41.9	9.8	9.0	21.5
(8)	Common ragweed	Ambrosia artemisiifolia L.	10.6	51.6	12.7	3.8	7.4
(9)	Smartweed	Polygonum pensylvanicum L.	10.3	58.1	11.2	3.6	6.1
(10)	Oxalis	Oxalis corniculata L.	9.2	48.4	10.5	3.3	6.9
(11)	Barnyard grass.	Echinochloa crus-galli (L.) Beauv	9.0	45.2	7.4	5.7	12.6
(12)	Plantain	Plantago lanceolata L. or Plantago major L.	8.5	38.7	9.4	4.3	11.1
(13)	Annual bluegrass	Poa annua L.	8.5	29.0	5.3	8.7	30.0
(14)	Common lambsquarter	Chenopodium album L.	7.8	45.2	6.9	3.6	7.9

Table 2.1. Relative abundance (RA), frequency (F), field uniform (FU), mean field density (MFD), and mean occurrence field density

(MOFD) of weeds in Ohio viney	yards. The number in parentheses	is the ranking number of this w	veed species based on RA value.
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Continued

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1 auto	2.1 continued.						
RA Rank	Common Name	Scientific Name	RA	F	FU	MFD	MOFD
(15)	Quackgrass	Elymus repens (L.) Gould	7.7	25.8	7.7	5.8	22.6
(16)	Prickly sida	Sida spinosa L.	7.4	22.6	4.5	8.2	36.1
(17)	Dock	<i>Rumex crispus</i> L. or <i>Rumex obtusifolius</i> L.	6.0	38.7	4.0	3.2	8.3
(18)	Common purslane	Portulaca oleracea L.	5.8	22.6	7.1	3.0	13.1
(19)	Yellow nutsedge	Cyperus esculentus L.	5.6	35.5	4.4	2.6	7.3
(20)	Virginia copperleaf	Acalypha virginica L.	4.7	29.0	4.4	1.7	5.8
(21)	Canada thistle	Cirsium arvense (L.) Scop.	4.5	25.8	4.4	1.8	6.8
(22)	Ground ivy	Glechoma hederacea L.	4.3	25.8	4.0	1.7	6.4
(23)	Groundsel	Senecio vulgaris L.	4.0	16.1	4.5	2.3	14.1
(24)	Indian tobacco	Lobelia inflata L.	3.9	19.4	3.6	2.2	11.3
(25)	Horsenettle	Solanum carolinense L.	3.6	29.0	2.7	0.7	2.5
(26)	Nimblewill	Muhlenbergia schreberi J.F.Gmel.	3.5	16.1	1.9	3.1	19.4
(27)	Red sorrel	Rumex acetosella L.	3.1	19.4	1.9	1.8	9.1
(28)	Carpetweed	Mollugo verticillata L.	2.4	16.1	2.3	0.7	4.5
(29)	Eastern black nightshade	Solanum ptychanthum Dunal	2.2	12.9	1.5	1.4	10.8
(30)	Knotweed	Polygonum arenastrum Boreau	2.2	19.4	1.6	0.3	1.3
(31)	Sowthistle	Sonchus oleraceus L.	2.0	12.9	1.9	0.6	4.6
(32)	Pokeweed	Phytolacca americana L.	1.5	12.9	1.0	0.3	2.0
(33)	Wild carrot	Daucus carota L.	1.4	12.9	1.0	0.2	1.4
(34)	Marestail	Hippuris vulgaris L.	1.3	12.9	0.7	0.2	1.6
(35)	Morningglory	Ipomoea pandurata (L.) G.F.W. Meyer	1.3	6.5	1.8	0.3	5.2
(36)	White campion	Silene latifolia Poir.	1.3	9.7	1.1	0.3	3.2
(37)	Galinsoga	Galinsoga quadriradiata Cav.	1.2	9.7	0.7	0.4	4.5
(38)	Bramble	Rubus spp.	1.1	9.7	0.8	0.2	2.1

Table 2.1 continued

32

Continued

Table	2.1 continued.						
RA Rank	Common Name	Scientific Name	RA	F	FU	MFD	MOFD
(39)	Honeyvine milkweed	Funastrum cynanchoides (Dcne.) Schlechter	1.1	9.7	0.8	0.2	2.1
(40)	Shepherd's purse	Capsella bursa-pastoris (L.) Medik.	1.1	6.5	1.0	0.5	7.2
(41)	Hemp dogbane	Apocynum cannabinum L.	1.0	9.7	0.7	0.1	1.1
(42)	Spurge	Chamaesyce maculata (L.) Small	1.0	9.7	0.7	0.1	1.1
(43)	Wild mustard	Sinapis arvensis L.	1.0	9.7	0.5	0.2	1.9
(44)	Devil's beggarticks	Bidens frondosa L.	1.0	9.7	0.5	0.1	1.3
(45)	Velvetleaf	Abutilon theophrasti Medik.	0.8	6.5	0.3	0.4	6.4
(46)	Speedwell	Veronica persica Poir.	0.8	6.5	0.7	0.2	2.4
(47)	Bindweed	Calystegia sepium (L.) R. Br. Or Convolvulus arvensis L.	0.8	6.5	0.7	0.1	1.6
(48)	Buttercup	Ranunculus parviflorus L.	0.8	3.2	1.0	0.3	9.6
(49)	White heath aster	Symphyotrichum pilosum (Willd.) Nesom	0.7	6.5	0.5	0.1	1.6
(50)	Wirestem muhly	Muhlenbergia frondosa (Poir.) Fern.	0.7	3.2	0.3	0.7	22.4
	-	Physalis heterophylla Nees or Physalis					
(51)	Groundcherry	longifolia (Nutt.) var. subglabrata	0.7	6.5	0.3	0.1	1.2
		(Mackenzie & Bush) Cronq.					
(52)	Cinquefoil	Potentilla recta L.	0.7	6.5	0.3	0.1	0.8
(53)	Wild buckwheat	Polygonum convolvulus L.	0.3	3.2	0.2	0.0	0.8

Table 2.2. The effect of herbicide management program on density of 20 dominant weed species based on the Relative Abundance statistic.

		Den	sity ^a	
Weed Species	Non-chemical	Glyphosate	Paraquat + Residual ^b	Glyphosate + Residual ^b
		plant	s/m ²	
Annual bluegrass	0.2 a	0.1 a	0.4 a	1.0 a
Barnyard grass	0.9 a	0.5 ab	0.0 b	0.1 ab
Crabgrass	3.9 b	25.7 a	1.7 b	8.2 b
Chickweed ^c	0.1 b	1.8 a	0.0 b	0.8 ab
Clover ^c	0.7 ab	1.9 a	0.0 c	0.1 bc
Common purslane	0.0 b	0.7 a	0.0 b	0.1 b
Common ragweed	1.2 a	0.5 a	0.0 b	0.3 a
Dock ^c	0.1 a	0.3 a	0.0 a	0.0 a
Dandelion	3.0 b	14.7 a	0.0 c	0.3 c
Fall panicum	0.0 b	2.7 a	0.1 b	0.0 b
Foxtail ^c	1.4 b	0.6 b	16.9 a	1.8 b
Lambsquarter	0.2 a	0.2 a	0.0 a	0.2 a
Oxalis	0.5 ab	0.9 a	0.0 c	0.1 bc
Pigweed ^c	5.2 a	0.4 bc	0.0 c	2.6 ab
Plantain ^c	0.9 b	0.3 bc	7.7 a	0.0 c
Prickly sida	0.0 a	0.0 a	0.0 a	1.0 a
Quackgrass	8.5 a	0.0 b	0.3 b	0.0 b
Smartweed	1.4 a	0.1 b	0.0 b	0.6 a
Virginia copperleaf	0.0 ab	0.3 a	0.0 b	0.0 ab
Yellow nutsedge	0.0 b	0.0 b	1.2 a	0.1 b

^a Means within species followed by different letters are significantly different

according to SNK test (P<0.05) Square root transformation was applied to density (plants/m²) before statistical analysis. Square root data were back transformed for presentation.

^b Residual herbicide = simazine or diuron or dichlobenil.

^c pigweed = redroot and smooth; foxtail = giant, yellow and green; clover = red and white; chickweed = common and mouseear; plantain = broadleaf and buckhorn; dock = broadleaf and curly.

Wood Spacios	Ľ	Density ^a
weed species	Vinifera (Hilled)	Non-vinifera (Non-hilled) ^c
_	pl	ants/m ²
Barnyard grass	0.96 a	0.15 b
Clover ^b	1.06 a	0.39 b
Common ragweed	0.81 a	0.33 b
Crabgrass	3.47 b	15.90 a
Dandelion	5.98 a	2.83 b
Foxtail ^b	0.78 b	2.17 a
Groundsel	0.54 a	0.01 b
Hemp dogbane	0.01 a	0.00 b
Horsenettle	0.06 a	0.01 b
Knotweed	0.02 a	0.00 b
Lambsquarter	0.61 a	0.07 b
Oxalis	1.26 a	0.12 b
Plantain ^b	0.73 a	0.21 b
Common purslane	0.01 b	0.31 a
Quackgrass	2.05 a	0.09 b
Smartweed	0.74 a	0.25 b
Sowthistle	0.06 a	0.00 b
White campion	0.03 a	0.00 b

Table 2.3. The effect of hilling practice on weed species density.

^a Means within species followed by different letters are significantly different

according to SNK test (P<0.05) Square root transformation was applied to density

(plants/m²) before statistical analysis. Square root data were back transformed for

presentation.

^b foxtail = giant, yellow and green; clover = red and white; plantain = broadleaf and

buckhorn.

^c Non-vinifera = American or French hybrid grape varieties.

Chapter 3: The Effect of Simazine Treated Mulches on Weed Control in an Auxerrois Vineyard

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Weed control in a vineyard with simazine treated mulches (STM) was studied over two years. Simazine applied at 2.7 and 5.4 kg ai/ha in November to wood bark or straw mulch, which had been applied primarily for winter-protection of the vine graft union, was largely ineffective in controlling weeds the following spring. A second application of simazine in June, after the mulch was pulled away from the vine graft union, controlled weeds for a month but the efficiency of weed control decreased as the season progressed. In contrast, wood bark (125 tons/ha) and wheat straw (20 tons/ha) controlled most weeds effectively for the entire growing season. A significant antagonistic interaction between mulch and simazine was detected with simazine controlling weeds in bare-soil plots, while not contributing to weed control in wood bark and straw mulched plots. Because wood bark and straw mulch reduced total weed density to an extremely low level (5 to 10

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plants/m²), we were unable to detect a significant effect of simazine on weed density in their presence. Despite evidence for impressive weed control with wood bark and straw we predict continued use without herbicide intervention will result in a shift to poorly controlled species and reduced weed control. During the time-course of the study perennial broadleaf species and grasses became more prevalent in response to STM. In particular grasses were more prevalent in the second year, especially in straw mulched plots.

Nomenclature: Simazine.

Key words: Mulch, simazine, vineyard.

Winter hilling is an additional practice extensively adopted for growing vinifera type grapes in northern states including Ohio (Dami et al. 2005). Winter hilling consists of two tillage activities. The first operation, called "hilling", is to mound soil to cover the graft joint, which connects the vinifera scion and the root stock. The thermal insulation property of soil protects the graft union from deep freezes ensuring that sufficient scion material survives to regenerate a new vine in case of a freezing injury. The second tillage operation, called "dehilling", is to remove the hilled soil from the graft joint in the spring in order to prevent scion rooting. This is necessary because roots generated from vinifera scion are sensitive to a soil insect, namely phylloxera.

The annual double tillage required by winter hilling has a number of undesirable side effects that threaten the sustainability of the practice. Soil erosion is one such effect that is a huge challenge to growers. In one research vineyard at the Ohio State University, we observed a loss of 8 to 13 cm of surface soil after 15 years of winter hilling. Considering that the slope of the vineyard was less than five degree it is clear that vineyards planted on hillsides would face severe soil erosion. Winter hilling may also negatively affect weed control. A considerable portion of preemergence herbicides applied to soil under the trellis are subject to runoff during a soil erosion event (Jiang 2010) resulting in reduced weed control efficiency and non-target environmental impacts. A weed survey completed by our research team supported this notion, showing that weeds occurred at higher density in vineyards that were winter-hilled compared to those that were not (Jiang et al. 2008).

Organic mulch is an alternative to winter-hilling that has been used extensively for winter protection in strawberry production (Askew and Smith 2008). Zabadal (2003) compared winter protection effectiveness of winter hilling and straw mulch. He found that straw protected vinifera grapes as good as soil hilling. Besides winter protection, mulches can also contribute to weed control. Physical shading by mulch reduces germination of weed seeds that require light as a stimulus (Wesson and Wareing 1967). Species that do not require light for germination may germinate but not emerge because they do not have sufficient autotrophic energy to enable them to reach the surface. Mulches may also control weeds by releasing allelopathic compounds. Examples include benzoxazolinones released from the straw of winter rye (*Secale cereale* L.) (Barnes and Putnam 1987), and sorgoleone released from residues of *Sorghum* species (Netzly and Butler 1986; Nimbal et al. 1996).

Mulches have been shown to intercept a large portion of applied herbicide (Crutchfield et al. 1986; Banks and Robinson 1986). Reichenberger et al. (2007) proposed that mulch would be an effective mitigation tool to reduce pesticide offsite movement. Moreover, in many studies soil active herbicides augmented the physical control provided by mulch in a manner analogous to that provided by naturally occurring allelochemicals. Prihar et al. (1975) reported that combining atrazine with wheat straw provided a better control than straw or the herbicide alone. Case and Mathers (2006) reported that flumioxazin-treated hardwood or pine nugget mulch controlled weeds more efficiently than did either input alone. Teasdale et al. (2005) also observed a synergistic effect between metolachlor and hairy vetch residue on the emergence of various weeds, including smooth pigweed (*Amaranthus hybridus* L.), common lambsquarters (*Chenopodium album* L.), giant foxtail (*Setaria faberi* Herrm.) and velvetleaf (*Abutilon theophrasti*). However, survival of some species appears to have been enhanced when treated with herbicide treated mulch. Control of fall panicum (*Panicum dichotomiflorum* Michx.) was antagonized when metolachlor was combined with a residue-mulch of hairy vetch (Teasdale et al. 2003). Liebl et al. (1992) pointed out that annual grasses and perennial broadleaf weeds were becoming more and more abundant under no-till cropping systems in which surface-mulch of crop residues accumulated.

Beattie (1955) reported long term benefits, including increased yield and plant vigor, of annual straw mulch application in Concord grapes at the conclusion of a nine-year study. These benefits could be attributed to the positive effects of mulch on the soil environment, including conservation of soil moisture, alleviation of sharp fluctuations in soil temperature, and enhancement of soil microbial activity. Enhancement in weed control may have been another important factor. When mulches were combined with postemergence herbicides, weeds were effectively controlled in vineyards (Elmore et al. 1997). However, the use of preemergence herbicides in combination with mulch in vineyards has not been investigated. In this study, simazine was used because it is an important vineyard herbicide in Ohio. Nationally, simazine was applied to over 20 % of vineyards in 2005 (USDA 2006). However, simazine use is constrained by problems associated with resistant weeds and environmental contamination. Innovations that would mitigate these constraints and provide more efficient and environmentally benign use of simazine should be of great interest. Specifically, the objective of this research was to

examine the vineyard weed control potential of combining simazine applications with mulches of wheat straw or wood bark.

Materials and Methods

Field trials. Experiments were conducted in research vineyards of the Ohio Agricultural Research and Development Center at Kingsville (41°53'5"N, 80°41'52"W). The cultivar Auxerrois was planted in 2000 and trained to a vertical shoot position. A conventional management program was used including winter hilling, fertilizing and pesticide applications as needed. Vines were pruned to 5 buds per 30 cm in the spring before bud break. The soil was a Bogart loam with pH of 6.1, organic matter content of 1.98 %, and a CEC of 6.6 meq/100g. Each plot (1 by 3.6 m) consisted of two vines spaced 1.8 m apart in the row. From the onset of the experiment in November 2007, vines were either hilled with soil or treated with a layer of mulch that extended about 50 cm from the center. Prior to applying mulch or mounding soil in November 2007, seeds of common lambsquarters (*Chenopodium album* L.), redroot pigweed (*Amaranthus retroflexus* L.), velvetleaf (*Abutilon theophrasti* Medik.) and giant foxtail (*Setaria faberi* Herrm.) were evenly scattered by hand over the surface of each plot at 18.9, 3.0, 17 and 5.5 g, respectively, in an attempt to increase emerged weed density. Mulch (mixed hardwood-species bark or

wheat straw) was applied by volume to each plot in late autumn 2007. The graft union was covered by 10 cm of mulch forming a cone with a diameter of about 1 m. The rest of the plot area was covered by mulch to a depth of 5 cm. This application rate required 20 tons/ha of straw and 125 tons/ha of bark mulch. Soil hilling covered the graft union approximately 20 cm high. As the standard industry treatment for managing winter injury, soil hilling was the control for the experiment. In November 2008, additional bark or straw was added to the original treatments to restore the depths of mulch around and between the vines as described above. This required 20 additional L of straw or bark around each vine to cover the graft union, and soil was also hilled for the soil hilling treatment. The replenishment rate was equivalent to 4.5 and 28 tons/ha for straw and bark, respectively.

Simazine¹ was applied as an aqueous emulsion to the top of the mulch or soil at 0, 2.7 or 5.4 kg/ha in November 2008 and 2009. The following June soil and mulch were pulled away from the graft union. Simazine was reapplied to the original plots at the same rate as in November after all surviving weeds were removed by hand. The CO_2 backpack sprayer used to apply simazine included a single 8003EVS nozzle² and delivered 230 L/ha at 240 kPa pressure. Total density of each identified weed species was counted monthly throughout the growing season until the end of August. Relative abundance (RA) of each species was calculated according to the method of Thomas (1985). A complete randomized block design with four replications was adopted in the vineyard.

Data analysis. Weed species density data were synthesized into the categories of total weeds, annual broadleaf weeds, perennial broadleaf weeds, and grasses. Density data were square-root transformed and subject to an ANOVA model by SAS³. Means were separated by the Student-Newman-Keuls test at the 0.05 level.

Results and Discussion

Principal weed species in the vineyard. The vineyard had a diverse weed community, and the major weed species, based on their relevant abundance (Table 3.1), consisted of annual broadleaf weeds [common lambsquarters, common chickweed (*Stellaria media* (L.) Vill.), common purslane (*Portulaca oleracea* L.), knotweed (*Polygonum aviculare* L.) and smartweed (*Polygonum pensylvanicum* L.)], perennial broadleaf weeds [Canada thistle (*Cirsium arvense* (L.) Scop.), horsenettle (*Solanum carolinense* L.) and red sorrel (*Rumex acetosella* L.)], annual grass species [giant foxtail (*Setaria faberi* Herrm.), crabgrass (*Digitaria sanguinalis* (L.) Scop.) and fall panicum (*Panicum dichotomiflorum* Michx.)] and a perennial grass species [quackgrass (*Elymus repens* (L.) Gould)] (Table 3.2).

The effect of STM on total weed density. Simazine did not effectively control the total weed community in the vineyard (Table 3.3). For the most part applications made in November did not reduce weed density, with the single exception of the rating that

occurred on June 3, 2008. Simazine application in June significantly reduced weed density compared to that in untreated plots for about two months. However, the level of control achieved was one that most growers would find unacceptable, ranging from 20 to 34 plants/m² after simazine was applied in June at 5.4 kg/ha in 2009. The efficiency of June application declined as the season progressed. For example, simazine at 2.7 kg/ha controlled weeds in July, but had lost its effectiveness by August, 2009.

The effect on total weed control could be attributed to lack of effective control on annual broadleaf weeds (Table 3.4) and grasses (Table 3.5). Previously simazine had been reported to poorly control grasses (Flanagan 1959; van Goor and Jager 1962), and another companion trial indicated that triazine resistant (TR) common lambsquarters accumulated in response to simazine treatment (Jiang 2010). Common lambsquarters was the most important annual weed species in the plot and although not confirmed as TR it is likely they were; moreover, the relative abundance of annual grasses increased over the time course of the study (Table 3.1). Dissipation or offsite movement of the herbicide from the weed seed germination zone though not documented likely contributed to reduced efficacy during the months following June application.

In contrast to simazine, mulches controlled weeds very effectively throughout the growing season (Table 3.3). Bark reduced weed density by a factor of 26 on June 3rd, and of 22, 23 and 28 on June 19th, July 22nd and August 26th 2008, respectively. However, bark controlled weeds to a larger extent in 2008 than in 2009, when a reduction of 11, 6, 9 and 6 times was observed in May, June, July and August, respectively. Similar to bark, straw also provided a season-long weed control and was less effective in 2009 than 2008.

To compensate for the degradation of straw mulches, Beattie (1955) applied straw at a rate of about 10 tons/ha annually to the original mulch in a nine-year study conducted in Ohio. In our study a replenishment rate of 4.5 tons/ha was used in the fall of 2008 for the purpose of winter protection and a significant degradation of the mulch was observed in spring of 2009. Degradation of the organic material was likely a contributing factor to the higher weed density in straw-mulched plots in 2009. It was also likely to be the same case for bark that the replenishment could not compensate for degradation and resulted in reduction in weed suppression by bark in 2009.

The effect of STM on annual broadleaf weed density. Simazine applied before winter had little residual activity by the beginning of the next growing season (Table 3.4). When simazine was reapplied in June, effective suppression of weeds was observed in July but not in August. This pattern was very similar to the effect on the total weed density as affected by simazine (Table 3.3), largely due to the fact that annual broadleaf weeds were the major component of the whole weed community in both years (Table 3.1).

Because these annual broadleaf weeds, such as chickweed, common lambsquarters, common purslane, knotweed and smartweed, are among those reported as sensitive to simazine, dissipation or off target movement was very likely an important contributor to the ineffective weed control by simazine. During the interval from November 1st 2007, when simazine was applied, to the first rating date (June 3rd 2008), 620 mm of precipitation occurred (OARDC Weather System 2010). Because the vineyard had a

sandy soil, this large amount of precipitation may have resulted in a significant simazine offsite movement.

Occurrence of triazine resistant common lambsquarters was another important factor in the less than satisfactory weed control observed. In 2008, we analyzed the dynamics of common lambsquarters in response to simazine, and found that simazine resulted in a predominantly TR population (Jiang 2010). Because common lambsquarters was the most important annual broadleaf weeds in the vineyard (Table 3.1), a TR population would largely reduce the overall efficiency of simazine on weed control.

Overall, mulches suppressed annual broadleaf weeds with high efficiency throughout both growing seasons. Annual broadleaf density in bare-soil plots ranged from 42 to 59 plants/m² in 2008; in contrast, the weed density was reduced down to 0 to 1 plants/m² in bark mulched plots and 1 to 5 plants/m² in straw mulched plots. A similar pattern was observed in 2009. In addition to physical suppression (Teasdale et al. 2005) and possible allelochemicals (Steinsiek et al. 1982) mulch affected common lambsquarters population dynamics. Mulch prevented selection of an emerged TR common lambsquarters population (Jiang 2010), thereby contributing to the higher weed control efficacy of mulches compared to simazine.

The effect of STM on perennial broadleaf weed density. Simazine did not affect perennial broadleaf weed density in the spring of 2008 and 2009 (Table 3.6). However, simazine reapplied at 5.4 kg/ha in June reduced perennial weed density in July and August both years. According to simazine labels (Simazine 90DF; CDMS 2010), the

herbicide can control many perennial broadleaf weeds when applied at higher rates. Skroch et al. (1975) reported that simazine at 3.4 kg/ha suppressed horsenettle. Therefore, it is reasonable to assume that simazine applied at 5.4 kg/ha suppressed perennial weeds in this study. Whereas, over-winter leaching of simazine may have contributed to poor control of annuals, such movement of simazine down through the soil profile would have increased the opportunity for uptake of simazine by deep-rooted perennials. Further research should be conducted to confirm these observations and explanations because of the low density of perennial broadleaf weeds in this study.

Mulches controlled perennial broadleaf weeds in the spring but became less effective as the growing season progressed. In addition to the physical smothering of weeds, we observed that mulch reduced soil temperature and the rate of increase in temperature starting from the beginning of growing season (Jiang 2010; Van Wijk et al. 1959). Soil temperature is a primary cue that stimulates breaking bud dormancy and emergence of perennial weeds, such as Canada thistle (Hamdoun 1972); therefore, it is likely that mulches delayed the occurrence of perennials in the spring. Over the long term mulch may be beneficial to perennials by alleviating drought stresses. Elmore et al. (1997) and Crutchfield et al. (1986) pointed out that mulches do not effectively control certain perennial weeds. In this study, mulches did not control perennial weeds as the season progressed into summer in both years.

Lack of effective control of perennial broadleaf species by both mulch and simazine resulted in a shift in the community to one more abundant in perennials (Table 3.1).

47

Postemergence herbicide applications may be required to control perennials in a vineyard that employs mulch for winter protection and control of annual weeds.

The effect of STM on density of grasses. Simazine applied before winter did not affect grass density in spring (Table 3.5). Grass control with simazine reapplied in June, while generally significant at the 5.4 kg/ha rate, was at a level unlikely to be acceptable to vineyard managers, particularly in 2009. These results suggested that the suppression of grasses by simazine was very poor, corroborating results published by Flanagan (1959) and van Goor and Jager (1962).

Bark controlled grasses more effectively than straw in 2009, although no statistical differences were found between bark and straw on grass control in 2008 (Table 3.5). Less grass in bark-mulched plots than in straw-mulched plots was observed on three of four rating dates in 2009.

Mulch and simazine interactions. When simazine was applied before winter interactions between mulch and simazine were not observed the following spring (Table 3.7) except for annual broadleaf weed density on June 11th, 2009. In contrast, simazine application made in June interacted with mulch to reduce annual broadleaf weeds in July of both years; however, these effects were short-lived and could not be detected by August. Annual broadleaf weed density was reduced as simazine application rate increased in bare-soil plots; whereas, weed density was not sensitive to increasing simazine application rate in the straw- or bark-mulched plots (Figure 3.1a to d).

The interaction observed was antagonistic. Banks and Robinson (1983) observed that mulch intercepted a large portion of applied herbicide, reducing the amount reaching the soil and thereby, the weed control efficiency. In this research mulches alone controlled weeds so effectively that they caused an extremely low weed density (Figure 3.1). The total weed density was nearly 700 plants in a bare-soil simazine-free plot in July 2008; in contrast, the total weed population was only 23 to 32 plants in a straw- or bark-mulched simazine-free plot (Figure 3.1a). With so few weeds surviving mulch application, the likelihood of detecting a significant simazine effect was very low. Therefore, this antagonistic interaction should not be interpreted as suggesting that mulch attenuated the effectiveness of simazine.

Weed community change over two years. Dynamics of the weed community were examined by comparing the relative abundance (RA) of species over time (Table 3.1). We observed a significant shift in the community of the entire vineyard from season to season and from year to year. The season to season changes reflected different weed lifecycles. For example, common chickweed (a winter annual) became less abundant when the season progressed from spring to summer in both years, while common purslane (a summer annual) became more abundant. That grasses and perennials became more abundant in 2009 than 2008 was likely affected by a variety of factors, such as differences in rainfall and temperature from year to year. However, the STM treatments were the dominant factors as previously described. Regardless of mulch or simazine treatment, more grasses were observed in 2009 when compared with the corresponding date in 2008 (Table 3.5). A constant increase in grass RA was also observed from 2008 to 2009 (Table 3.1); grass RA values were 38, 58, 61 and 84 in June and August 2008 and May and August 2009, respectively. Like grasses, perennial broadleaf weeds were becoming more and more prevalent from year to year. The RA values for Canada thistle and horsenettle were 21 and 20, respectively, in August 2008, and were 32 and 27, respectively, in August 2009.

The nature of selectivity is such that a weed management tool should not be expected to control all existing species with an equally good efficiency. This characteristic inevitably leads to increasing abundance of poorly controlled weeds. Dramatic increases in abundance of herbicide resistant biotypes to repeated herbicide application are the most extreme example of this phenomenon (Triplett and Lytle 1972). In this study, since perennials were not effectively controlled by mulches or simazine, it was not surprising to see their increasing abundance after STM treatments were repeated for two years. Reduced tillage associated with STM was an additional factor; Buhler (1995) reported that perennial species became more common in reduced tillage systems.

Here we have reported outstanding weed control with STM that is largely due to the season-long weed suppression by mulches. Wood bark and straw had a fundamental role in the weed control achieved suggesting great potential to reduce herbicide input through their use in vineyards. Pre-winter application of simazine to mulch or bare-soil had little residual activity the next spring. Reapplication of the herbicide in June controlled weeds temporarily but was not sufficient for the balance of growing season. These results

suggested that simazine applied in early spring to fall-applied mulch instead of before winter would increase the herbicide contribution to weed control. The occurrence of more grasses and perennial weeds over the course of the study was an overall defining effect of STM on the weed community. Together with a likely high incidence of TR common lambsquarters, weed community composition and its trajectory were such that the efficacy of simazine was minimal even at high rates. Application rates of wood bark and straw used in this research would require large additional expenditures unlikely to be recouped through improved weed control alone. However, in related research the use of these organic mulches in the vineyard brought about a series of integrated benefits, including soil conservation, winter protection, vineyard nutrient management as well as a potential to increase yield but not necessarily reduce fruit quality (Jiang et al. 2010). Therefore, the long-term efficiency of mulch use in the vineyard with respect to a host of potential benefits including interaction with herbicides and other pesticides needs to be investigated in order to develop a more economically and environmentally sound weed control program.

Source of Materials

- ¹ Princep® 4L, Syngenta Crop Protection, P.O. Box 18300, Greensboro, NC 27419.
- ² Spraying System Co., P.O. Box 7900. Wheaton, IL 60189.
- ³ SAS 9.1, SAS Institute Inc. 100 SAS Campus Drive, Cary, NC 27513.

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	June 2008	8	August 20)08	May 2009	9	August 200)9
RA Rank	Weeds	RA^{\P}	Weeds	RA	Weeds	RA	Weeds	RA
1	POLAV ^{**}	44	GRASS	58	GRASS	61	GRASS	84
2	STEME	44	CHEAL	33	CHEAL	42	CHEAL	43
3	GRASS	38	POROL	29	STEME	38	CIRAR	32
4	CHEAL	37	STEME	22	POLAV	33	SOLCA	27
5	POLPY	25	CIRAR	21	CIRAR	31	RUMAA	24
6	CIRAR	21	SOLCA	20	POLPY	31	POROL	19
7	OXACO	21	AMARE	20	LEPVI	20	STEME	18
8	LEPVI	20	ABUTH	18	SENVU	14	AMARE	17
9	TAROF	12	LEPVI	16	RUMAA	14	POLPY	10
10	ABUTH	12	POLPY	14	CONAR	11	CONAR	10
11	CONAR	11	POLAV	14	PLAMA	4	ABUTH	8
12	SENVU	8	OXACO	12			SENVU	7
13	AMARE	5	SENVU	11			POLAV	2
14	PLAMA	4	CONAR	10				
15			PLAMA	1				

Table 3.1. Relative abundance of weed species in response to two-years of simazine treated mulches^{*} at Kingsville, Ohio.

^{*} Mulches were initially applied in November 2007, followed by simazine application on the same day. Simazine application was repeated in June 2008 after all weeds were removed. Two buckets of straw or bark (total volume of 40 L) were added to the mulched plot to supplement the loss of mulches during the past year, and the same simazine application was repeated in November 2008. In

June 2009, the same application was again made to the same plots after all weeds were removed.

** Full scientific names were listed in Table 3.2. [¶]Relative abundance, calculated according to the method of Thomas (1985).

57

Common name	Scientific name	Code
Bindweed	Convolvulus arvensis L.	CONAR
Canada Thistle	Cirsium arvense (L.) Scop.	CIRAR
Common Chickweed	Stellaria media (L.) Vill.	STEME
Common Lambsquarters	Chenopodium album L.	CHEAL
Common Purslane	Portulaca oleracea L.	POROL
Dandelion	Taraxacum officinale G.H. Weber ex Wiggers	TAROF
Grasses (large crabgrass, giant foxtail, fall panicum and quackgrass)	Digitaria sanguinalis (L.) Scop. or Setaria faberi Herrm. or Panicum dichotomiflorum Michx. or Elymus repens (L.) Gould	GRASS
Common Groundsel	Senecio vulgaris L.	SENVU
Horsenettle	Solanum carolinense L.	SOLCA
Prostrate Knotweed	Polygonum aviculare L.	POLAV
Oxalis	Oxalis corniculata L.	OXACO
Pepperweed	Lepidium virginicum L.	LEPVI
Plantain	Plantago major L.	PLAMA
Red Sorrel	Rumex acetosella L.	RUMAA
Redroot Pigweed	Amaranthus retroflexus L.	AMARE
Smartweed	Polygonum pensylvanicum L.	POLPY
Velvetleaf	Abutilon theophrasti Medik.	ABUTH

Table 3.2. Weed species observed at the Auxerrois vineyard at Kingsville, Ohio.

85
Table 3.3. The effect of simazine treated mulches on total weed density in the Auxerrois vineyard in 2008 and 2009 at Kingsville,

Ohio.

Tuestasent					2008^{*}					2009**								
Treatment	June 03		Jun	June 19		July 22		ıst 26		May 15		Jun	June 11		July 15		ıst 12	
									plants/m ²									
Simazine									1									
-kg/ha-																		
0	40	a^{\P}	33	а	69	а	47	а		28	а	48	а	49	а	66	а	
2.7	28	a	39	а	19	b	24	b		32	a	53	а	28	b	57	a	
5.4	18	b	30	a	16	b	24	b		31	а	48	а	20	b	34	b	
Mulch																		
Bare soil	77	a	86	a	93	a	83	a		70	a	100	a	66	а	95	а	
Bark	3	b	4	b	4	b	3	b		6	b	17	b	7	c	17	с	
Straw	6	b	12	b	8	b	9	b		15	b	33	b	24	b	44	b	

59

* Simazine was applied on November 1st, 2007 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 19th, 2008 and the same simazine treatment was repeated in the same plot after all weeds were removed.

** Simazine was applied on November 16th, 2008 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 23rd, 2009 and the same simazine treatment was repeated in the same plot after all weeds were removed.

[¶] Means followed by the same letter were not significantly different from each other in the same column within mulch or simazine treatment at P<0.05.

Table 3.4. The effect of simazine treated mulches on annual broadleaf weed density in the Auxerrois vineyard in 2008 and 2009 at Kingsville, Ohio.

	Ture the suit				2	2008^{*}					2009**								
	Treatment	June 03		Jun	e 19	Jul	y 22	2 Augus		<u> </u>	Ma	y 15	June	e 11	Jul	y 15	Augi	ust 12	
										plants/m ²		-							
	Simazine																		
	-kg/ha-																		
	0	30	a^{\P}	22	a	44	а	22	a		15	а	12	a	18	а	13	a	
	2.7	19	ab	21	a	8	b	9	a		17	а	15	a	8	b	11	a	
	5.4	13	b	18	a	9	b	12	a		22	a	22	a	9	b	13	a	
6	Mulch																		
\circ	Bare soil	58	а	54	a	59	а	42	a		53	а	49	a	35	а	36	a	
	Bark	1	с	1	c	1	b	0	b		0	b	0	b	0	b	0	b	
	Straw	3	b	5	b	1	b	1	b		0	b	0	b	0	b	1	b	

* Simazine was applied on November 1st, 2007 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 19th, 2008 and the same simazine treatment was repeated in the same plot after all weeds were removed.

** Simazine was applied on November 16th, 2008 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 23rd, 2009 and the same simazine treatment was repeated in the same plot after all weeds were removed.

[¶] Means followed by the same letter were not significantly different from each other in the same column within mulch or simazine treatment at P<0.05.

Table 3.5	5. The effect of	simazine treated	mulches on gras	s weed density in th	ne Auxerrois	vineyard in 20	008 and 2009 a	at Kingsville,
Ohio								

Table 3.5. The effect of simazine treated mulches on grass weed density in the Auxerrois vineyard in 2008 and 2009 a	t Kingsville,
Ohio.	

	The stars and				2	008^*					2009**								
	I reatment	Jun	e 03	Jun	e 19	Jul	y 22	Augı	ist 2	6	Ma	y 15	June	e 11	July	/ 15	Augi	ust 12	
	Simazine -kg/ha- 0 2.7	65	a¶ a	7 9	a a	4	a a	4	a a	plants/ m ²	99	a a	22 17	a a	27 14	a b	23 22	a a	
	5.4	I	а	3	a	1	b	1	b		8	а	15	а	9	b	12	a	
	Mulch																		
	Bare soil	7	а	10	а	4	a	5	а		11	a	24	a	24	а	16	b	
ע	Bark	1	b	3	b	2	a	2	а		4	b	18	а	4	b	8	b	
	Straw	3	b	6	b	3	a	4	а		11	a	11	а	20	a	33	a	

^{*} Simazine was applied on November 1st, 2007 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 19th, 2008 and the same simazine treatment was repeated in the same plot after all weeds were removed.

** Simazine was applied on November 16th, 2008 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 23rd, 2009 and the same simazine treatment was repeated in the same plot after all weeds were removed.

[¶] Means followed by the same letter were not significantly different from each other in the same column within mulch or simazine treatment at P<0.05.

Traatmont				2	008^{*}					2009**									
Treatment	June 03 Ju		June	: 19	19 July 22		Augu	August 26		May	y 15	June 11		Jul	/ 15	Augı	ıst 12		
								_	plants/ m ²										
Simazine									-										
-kg/ha-																			
0	6 8	a¶	7	a	4	a	4	а		4	a	15	a	4	a	11	а		
2.7	5 e	ı	9	a	5	а	6	a		7	a	21	a	6	a	13	а		
5.4	1 8	ı	3	a	1	b	1	b		1	a	12	a	2	a	3	b		
Mulch																			
Bare soil	7 a	ı	10	a	4	a	5	a		6	a	27	a	7	a	12	а		
Bark	1 ł)	3	b	2	a	2	a		3	a	6	b	3	a	6	а		
Straw	3 t)	6	b	3	a	4	a		4	a	14	b	3	a	8	а		

Table 3.6. The effect of simazine treated mulches on perennial broadleaf weed density in the Auxerrois vineyard in 2008 and 2009 at Kingsville, Ohio.

* Simazine was applied on November 1st, 2007 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 19th, 2008 and the same simazine treatment was repeated in the same plot after all weeds were removed.

^{**} Simazine was applied on November 16th, 2008 after mulching and soil hilling. The graft union was unburied from soil or mulch on June 23rd, 2009 and the same simazine treatment was repeated in the same plot after all weeds were removed.

[¶] Means followed by the same letter were not significantly different from each other in the same column within mulch or simazine treatment at P<0.05.

Table 3.7. Interactions of mulch and simazine on total, annual broadleaf, perennial broadleaf, and grass weed control in 2008 and 2009.

Mulah bu simazina		20	08 ^a		2009 ^b							
Whiteh by simazine	June 03	June 19	July 22	August 26	May 15	June 11	July 15	August 12				
Total Weeds	NS ^c	NS	**	*	NS	NS	NS	NS				
Annual Broadleaf weeds	NS	NS	**	NS	NS	*	**	NS				
Perennial Broadleaf weeds	NS	NS	NS	NS	NS	NS	NS	NS				
Grasses	NS	NS	NS	NS	NS	NS	NS	NS				

^a Simazine was applied on November 1st, 2007 after mulching and soil hilling. The graft union was unburied from soil or mulch on

June 19th, 2008 and the same simazine treatment was repeated in the same plot after all weeds were removed.

^b Simazine was applied on November 16th, 2008 after mulching and soil hilling. The graft union was unburied from soil or mulch on

63

June 23rd, 2009 and the same simazine treatment was repeated in the same plot after all weeds were removed.

^c Mulch by simazine interaction was analyzed by ANOVA model. NS, not significant at P<0.05; *, significant at P<0.05; **,

significant at P<0.01.



Figure 3.1. The interaction of simazine rate and mulch on total weed control in July (a) and August 2008(b), and on broadleaf weed control in July of 2008 (c) and 2009 (d). Bars are standard deviations of the actual density data.

64

Chapter 4: Dynamics of Common Lambsquarters (*Chenopodium album* L.) Biotypes in Two Vineyards Determined with a Modified Molecular Method Targeting the *psbA* Gene

Linjian Jiang, Imed Dami, and Doug Doohan*

We modified a molecular method to efficiently and accurately differentiate triazine sensitive (TS) and triazine resistant (TR) biotypes, and used this method to monitor the dynamics of common lambsquarters populations in response to simazine treated mulches in two vineyards. Targeting the *psb*A gene, which determines TR in common lambsquarters, polymorphism was achieved on an agarose gel between TS control and putative TR after polymerase chain reaction (PCR). Sequencing of the *psb*A gene was included to validate the molecular method under different lab conditions without TR control. Local shredded wood bark and wheat straw were applied under the trellis in each vineyard and followed by simazine application at the rate of 2.7 and 5.4 kg ai/ha in November, 2007. The dynamics of the common lambsquarters populations were

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quantified in June, 2008. Simazine applied to bare soil significantly suppressed the total common lambsquarters population and resulted in a TR dominant population. The occurrence of TR biotype was unaffected by simazine rates when applied to bark-mulched plots. This result indicated that mulches could be a useful tool for management of TR weeds.

Nomenclature: Simazine; common lambsquarters, *Chenopodium album* L. CHEAL; wheat, *Triticum spp*.

Key words: Common lambsquarters, mulch, population dynamics, simazine, triazine resistance identification.

The photosynthesis inhibitor triazine herbicides, atrazine and simazine, have been widely used to control broadleaf weeds and grasses since the 1950s. Despite environmental concerns, triazine herbicides remain one of the major herbicide groups in use today because of low cost and a wide spectrum of weeds controlled. Corn (*Zea mays* L.) is the principal crop treated with triazines. According to the Agricultural Chemical Usage Database (USDA, 2009), 83 and 18 % of the total land planted to corn in Ohio was treated with atrazine and simazine, respectively, in 2007. Simazine is also extensively used in fruit production. In a vineyard survey conducted in 2004, we found about 23 % of grape growers in Ohio were using simazine (Jiang et al. 2008).

Extensive application of triazine herbicides has contributed to the development of resistance in many weeds and concomitant degradation of their value to agriculture. World-wide, 68 triazine resistant (TR) biotypes have been reported (Heap 2009). Herbicide resistant weeds are thought to occur due to selection on the wild population over several years by repeated application of herbicides with the same mechanism of action (Tranel and Wright 2002). Methods to confirm TR have contributed extensively to our understanding of the phenomenon, including the chlorophyll florescence method of Ali and Machado (1981) and the floating leaf disc method developed by Hensley (1981). However, these methods based on physiology are insufficiently robust to confirm resistance with absolute confidence. Since TR in common lambsquarters (*Chenopodium album* L.) is due to an A to G mutation and a subsequent serine₂₆₄ to glycine change in the *psbA* protein (Bettini et al. 1987), Tian and Darmency (2006) developed a bidirectional allele-specific PCR method to detect the TR biotype at the genetic level.

They reported that TS and TR common lambsquarters showed polymorphism on an agarose gel at the annealing temperature of 62 °C: the TS biotype had two fragments of 637 and 214 bp; whereas, the TR biotypes had two fragments of 637 and 447 bp.

Integrating herbicides with cultural practices and physical methods of control has been proposed as a systematic approach to improve weed control, and also to preserve herbicide efficacy by foiling selection of resistant biotypes. Mulch is one such physical control that affects weed community dynamics by altering the micro-environment in which seeds germinate and grow. Control of species occurs in response to physical suppression of germination and seedling growth, combined with attenuating the penetration of light required by many species to trigger germination (Wesson and Wareing 1969; Froud-Williams et al. 1984). Moreover, straw from cereal grains can release a variety of phytotoxic substances, including hydroxamic acids, phenolic acids and short-chain fatty acids (Wu et al. 2001). Release of allelochemicals was also reported from wood barks (Ortega et al. 1996; Parvez et al. 2004). In our preliminary trials simazine applied to bare soil at the rate of 10.8 kg ai/ha did not control the predominant species common lambsquarters, whereas, wheat straw, shredded wood bark and corn stubble treated with simazine did (Figure 4.1). This observation led us to hypothesize that control of TR weeds in the presence of simazine would be enhanced by the use of organic mulch.

We addressed the hypothesis by field and laboratory studies targeting the following two specific objectives: 1) to develop a robust molecular method to accurately and efficiently identify TR common lambsquarters, and 2) to monitor the dynamics of common lambsquarters in response to simazine treated mulches by using this molecularbased identification method in two vineyards.

Materials and Methods

Field trials. Experiments were conducted in vineyards of the Ohio Agricultural Research and Development Center (OARDC) at Wooster (40°46′43"N, 81°55′51"W) and Kingsville (41°53′5"N, 80°41′52"W). The soil types were a Wooster slit loam (pH 4.9, organic matter content 2.38 %, and CEC 12.8 meq/100g) at Wooster, and a Bogart loam at Kingsville (pH 6.1, organic matter content 1.98 %, and CEC 6.6 meq/100g). Each plot consisted of two grape vines spaced 1.2 and 1.8 m apart in the row, at Wooster and Kingsville, respectively. The mulch treatment extended approximately 50 cm each side of the center. Therefore, the plot size was 1 by 2.4 m at Wooster and 1 by 3.6 m at Kingsville. A mulch of mixed hardwood-species bark or wheat straw was applied to each plot to a depth of 5 cm. To increase the density of common lambsquarters, 18.9 g of locally-collected seeds were scattered into each plot before mulching in November, 2007. Simazine¹ was then applied as an aqueous emulsion to the top of the mulch or to bare soil at 0, 2.7 and 5.4 kg/ha on the same day mulch was applied in early November 2007. The CO_2 backpack sprayer used included a single 8003EVS nozzle² and delivered 230 L/ha at 240 kPa pressure. The field experiment was a complete randomized block design with four replications in both vineyards.

Method development for TR common lambsquarters detection. The gene psbA determines whether a common lambsquarters plant is TR or triazine sensitive (TS). A mutation from A to G resulting in a substitution of glycine for serine₂₆₄ confers TR (Bettini et al. 1987). The bidirectional PCR method targeting this allele-specific mutation developed by Tian and Darmency (2006) was used to identify TR biotypes in this study. However, both TR and TS biotype control were required by their method because polymorphisms may be different among different lab conditions. We modified their method by including *psb*A gene sequencing, and validated the method under different lab conditions even without a TR control.

Seeds were harvested from individuals that survived simazine applied in field trials at 10.8 kg/ha (Figure 4.1). This is twice the highest recommended rate for use in vineyards in the United States. Since TR is maternally inherited in common lambsquarters (Bettini et al. 1987), seeds collected from plots that survived simazine were assumed to be TR and were used in the experiments. TS common lambsquarters seeds³ were used as a negative control. DNA was extracted from common lambsquarters leaves according to the micro-prep method (Fulton et al. 1995). One leaflet was ground with DNA extraction buffer in a 1.5-ml centrifuge tube. A volume of chloroform:isoamyl alcohol (24:1) was added after incubating for 30 min at 65 °C. After thoroughly mixing the phases by inverting tubes four times, the samples were centrifuged at 10,000 rpm for 10 min. The

aqueous phase was transferred into a 1.5-ml centrifuge tube, in which DNA was precipitated by adding a volume of isopropanol. DNA was collected by centrifuging at 10,000 rpm for 10 min. The DNA pellet was washed with 70 % ethanol and resuspended in 200 µl of double distilled water.

Gradient PCR was conducted to find the optimum annealing temperature at which PCR products showed polymorphism between TS and the putative TR common lambsquarters samples on the agarose gel. Because a TR control was not available, further sequencing was conducted to confirm that the putative TR common lambsquarters that survived in our field trial were resistant. The DNA fragments were cut from the gel and purified using a PCR purification kit⁴ following the provided protocol. Sequencing was finished at the Molecular and Cellular Imaging Center at the OARDC.

The PCR mixture contained 50 mM KCl, 10 mM Tris-HCl, 0.1 mM dNTP, 2 mM $MgCl_2$, 0.12 μ M primer psbSF and 0.1 μ M of the other three primers. Primer sequences were: psbF, GTA GCT TGT TAT ATG GGT CGT GAG; psbR, TAC GTT CGT GCA TTA CTT CCA TAC; psbSF, GTC GAT TAA TCT TCC AAT ATG CTA; psbRR TAA AGA ACG AGA GTT GTT GAA ACC.

Dynamics of common lambsquarters biotypes. Common lambsquarters in each plot were counted in June 2008 and community dynamics (TR vs. TS) were quantified by collecting leaves from individual seedlings in each plot. The sample number from each plot was adjusted according to the density that occurred with different treatments (Table 4.1). This varied from 11 to 46 % of all individuals in bare soil plots, 24 to 83 % in bark

mulched plots and 15 to 73 % in straw mulched plots. Samples were stored on ice as they were collected and then transferred to a -20°C freezer until DNA extraction. The percentage of TR biotype in each plot was then calculated after separating TR and TS by the method described previously. Data were analyzed using GLM model in SAS⁵ and means were separated with a Student-Newman-Keuls test (P<0.05).

Results and Discussion

Identification of TR common lambsquarters. The annealing temperature at which TR and TS common lambsquarters provide different bands on the gel was determined by gradient PCR. A range of annealing temperatures from 46.5 to 64 °C was used. Three possible PCR products, 214, 447 and 637 base pair (bp) were observed on the gel. TR and TS biotypes showed polymorphism at several annealing temperatures. For example, at 64 °C (Lane 9; Figure 4.2a and 2b) the TS biotype had 1 band (637 bp) on the gel; whereas, the putative TR biotype had two bands (447 and 637 bp). At 57.3 °C (lane 5; Figure 4.2a and 2b), the TS biotype had 2 bands but the putative TR biotype had 3 bands. Because the specificity of the primer increases as the annealing temperature increases, we

decided to use 64 °C as the operative annealing temperature for identifying the TR biotype.

Regular PCR with an annealing temperature of 64 °C was then performed with DNA samples extracted from control TS common lambsquarters (n=2) and seedlings of common lambsquarters (n=60) that survived 10.8 kg/ha simazine (Figure 4.1). Because there was no TR control in this study, we further sequenced the 637-bp products purified from both control TS and the putative TR biotypes (Figure 4.2c). The *psb*A gene sequences matched the published *psb*A gene sequence of TR and TR biotype, confirming that these 637-bp products were part of the *psb*A gene (Bettini et al. 1987; Figure 4.3). Thus, this molecular method proved to be valid and efficient for identifying TR common lambsquarters at the genetic level.

We adopted the same primers as Tian and Darmency (2006) to discriminate the TR and TS biotypes. However, PCR at the annealing temperature used by the authors (62 °C) did no show polymorphism; DNA from both putative biotypes had two bands of 637 and 447 bp (Lane 7; Figure 4.2a and 2b). Moreover, the polymorphism apparent at other annealing temperatures (lanes 5 and 9; Figure 4.2a and 2b) did not show the same polymorphism reported by Tian and Darmency (2006). Failure of their method in our laboratory may have been due to slight differences in PCR conditions. This is a very common problem when a method is attempted in different labs. We modified Tian and Darmency's method to make it valid to study population dynamics under different lab conditions by including gradient PCR and sequencing. The modification included 1) gradient PCR to determine an appropriate annealing temperature that shows

polymorphism on the gel, 2) sequencing to confirm the polymorphism, and 3) application of regular PCR to analyze population dynamics. With this modified method, a suspected TR common lambsquarters can be determined by sequencing the 637 bp product of *psbA* gene without TR and TS controls. The sampling procedure was also simplified; a single leaf sample stored at -20 °C is sufficient for DNA extraction, whereas, the regular bioassay methods (Ali and Machado 1981; Hensley 1981) need seeds of suspected TR and known TR and TS controls to perform the assay.

The effect of simazine treated mulches on common lambsquarters control. Shredded wood bark reduced density of common lambsquarters at both vineyards (Table 4.1). Bark reduced common lambsquarters density by 70, 50 and 92 % when simazine was applied at 0, 2.7 and 5.4 kg/ha, respectively, compared with bare soil at Wooster. Wheat straw also decreased common lambsquarters density at Wooster regardless of simazine rate; however, at Kingsville straw controlled the weed only when simazine was applied at 5.4 kg/ha.

Common lambsquarters suppression by bark and straw was likely due to both physical factors and modification of the soil environment. Teasdale and Mohler (2000) demonstrated that common lambsquarters was vulnerable to the physical obstruction of mulch. Mulches also control weeds by releasing allelochemicals to the soil environment. Wheat straw and wood bark can release a variety of phytotoxic substances (Ortega et al. 1996; Parvez et al. 2004; Wu et al. 2001). Wu et al. (2001) reported that the allelopathic potential against weeds differed among wheat varieties and this may have been a factor

leading to the variable effect of wheat straw between the two locations. The wheat straw samples used in these experiments were locally acquired and therefore grown under different environmental conditions. Moreover, the variety used at each location was not determined. Alternatively, the porous characteristic of straw mulch and the low bulk density (0.04 g/cm³) might attenuate the physical suppression exherted on common lambsquarters.

Simazine reduced common lambsquarters density in bare-soil plots at both vineyards, as well as in the straw-mulched plots at Kingsville. However, control was more effective at Wooster than at Kingsville (Table 4.1). At Wooster, 2.7 and 5.4 kg/ha both reduced common lambsquarters density by about 80 % compared with the control; whereas, at Kingsville significant control was only observed when the herbicide was applied at 5.4 kg/ha. Sandier soil, lower CEC (6.6 vs. 12.8 meq/100g at Wooster) and lower organic matter content (1.98 % vs. 2.38 % at Wooster) were factors that likely contributed to reduced efficacy. Precipitation was also greater at Kingsville (591.1 mm) than at Wooster (505.7 mm) for the post-simazine application period of November 2007 to May 2008 (OARDC Weather System 2010). Therefore, it is likely that more leaching occurred at Kingsville resulting in a lower simazine content retained in the seed germination zone of the soil and less weed control than at Wooster.

In contrast to significant weed control with simazine applied to bare soil, simazine did not reduce weed density in bark- or straw-mulched plots at Wooster or bark-mulched plots at Kingsville (Table 4.1). The fact that mulches provided outstanding weed suppression on their own masked any effect of simazine on weed control, resulting in the apparent antagonism. For example at Kingsville, shredded wood bark reduced common lambsquarters density from 92 to 6 plants. At such a low density detecting a significant effect of simazine was improbable at the 0.05 level.

The effect of simazine treated mulch on the dynamics of common lambsquarters. A shift of the common lambsquarters population from one that was predominantly TS to one that was predominantly TR was observed when simazine was applied to bare soil in both vineyards (Table 4.2). For example, the occurrence of the TR biotype in the bare soil plots increased from 5 % when no simazine was applied, to 67 % when simazine was applied at 2.7 kg/ha, and to 100 % at 5.4 kg/ha at Wooster. This result confirmed that simazine controlled TS common lambsquarters (Table 4.1) and that surviving seedlings were TR. The incidence of TR in simazine-free bare-soil plots was very low, 5 % at Wooster and 0 % at Kingsville, probably because in the absence of triazine herbicides TR individuals were not competitive with their TS counterparts (LeBaron 2008). Parks et al. (1996) noted that under non-competitive conditions TR biotype common lambsquarters was less fit than the TS biotype. Leroux (1993) pointed out that a TR predominant common lambsquarters population would eventually become TS in the absence of triazine herbicides under competitive field conditions. Since the simazine-free bare-soil plots were also hosting many other weeds at high density, this created a very competitive condition likely responsible for their low abundance.

At Wooster, TR incidence in wheat straw plots did not change when simazine was applied at 0 or 2.7 kg/ha; neither TR nor TS biotypes established in straw mulched plots when treated with simazine at 5.4 kg/ha. However, simazine applied to wheat straw increased the TR biotype abundance as the application rate increased at Kingsville (Table 4.2). Frequency of the TR biotype was 8 % when straw mulch was applied alone; in contrast, 100 % were TR when straw had been treated with simazine at 5.4 kg/ha. Why straw was not effective in mitigating the selective pressure of simazine is not known. It may be that straw exerts less physical suppression on TR individuals than does shredded wood bark. The dry mass per m² of shredded wood bark applied to a depth of 5 cm was 13 kg compared to 2 kg for wheat straw.

In contrast to simazine applied alone or to straw, the abundance of the TR biotype did not increase when simazine was combined with shredded wood bark (Table 4.2). TR frequency was 8, 13 and 0 %, when simazine was applied to shredded wood bark at 0, 2.7 and 5.4 kg/ha at Wooster, respectively. These observations indicated that not only did shredded wood bark reduce total common lambsquarters density (Table 4.1), it minimized selection of TR biotypes when used in combination with simazine. Reduction of selection pressure on TR common lambsquarters could be contributed to the following two factors. Firstly, bark intercepted a large portion of simazine applied to the surface. Other researchers (Crutchfield et al. 1986; Banks and Robinson 1986) have shown that straw mulch reduces herbicide deposition to the soil surface by 4 to 9 folds compared to applications made to bare soil. Bark, with a less porous texture than straw, can be expected to reduce simazine concentration in the underlying soil, thereby reducing the selection pressure of simazine to TR weeds. Secondly, TR common lambsquarters are less fit than TS individuals (LeBaron 2008), making the physical suppression of bark very effective on TR common lambsquarters control.

In this research we modified the molecular method of Tian and Darmency (2006) by including gradient PCR and sequencing. This modification provides a system for identification of TR that is robust across diverse lab conditions. Applying this molecular method, we found that simazine applied to bare soil resulted in a predominantly TR population. When simazine was applied to wheat straw, both the total and TR common lambsquarters density was reduced although the frequency of TR biotype was either increased or unaffected. However, when simazine was applied to shredded wood bark, it did not affect the abundance of the TR biotype. Organic mulches have been well known to control weeds by physical smothering and possibly by allelochemicals, a different weed control mechanism from herbicides. Therefore, it is not surprising to observe that mulches suppressed herbicide resistant weeds. However, due to the difficulties to monitor the dynamics of weed population (in the aspect of herbicide resistance), convincing evidence has not been previously reported. The conclusive results of this study indicated that herbicide treated mulches are a potential new tool to manage TR common lambsquarters as well as other herbicide resistant weeds with different mode of actions. Further studies are needed to fully explore this possibility.

78

Source of Materials

¹ Princep® 4L, Syngenta Crop Protection, P.O. Box 18300, Greensboro, NC 27419.

² Spraying System Co., P.O. Box 7900. Wheaton, IL 60189.

³ Herbiseed, New Farm, Mire Lane West End, Waltham St. Lawrence, Reading, RG10 0NJ, UK

⁴ Qiagen Inc. 27220 Turnberry Ln # 200, Valencia, CA 91355

⁵ SAS 9.1, SAS Institute Inc. 100 SAS Campus Drive, Cary, NC 27513

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Figure 4.1. The effect of simazine treated-corn stubble, -wheat straw, simazine alone, and simazine treated-shredded wood bark on common lambsquarters on July 07, 2007 from left to right, respectively. Simazine was applied at 10.8 kg/ha in November 2006. Plot size was 1.5 by 1.5m. Common lambsquarters constituted 99 % of the weed community in the simazine-only treated plot.



Figure 4.2. Polymorphism in PCR products targeting the *psb*A gene in TR and TS common lambsquarters using primers psbF, psbR, psbSF, and psbRR. A 100 bp marker used for all agarose gels is indicated in lane 1.

(a) Amplification of the *psb*A gene of TS common lambsquarters purchased from
Herbiseed³. Annealing temperatures used in gradient PCR were 46.5, 50, 53.9, 57.3, 60.1,
62, 63.4, 64 °C from lane 2 to 9, respectively.

(b) Amplification of the *psb*A gene of the putative TR common lambsquarters. Annealing temperatures used in gradient PCR were 46.5, 50, 53.9, 57.3, 60.1, 62, 63.4, 64 °C from lane 2 to 9, respectively.

(c) Amplification of the *psb*A gene of the putative TR and TS common lambsquarters at an annealing temperature of 64 °C. Lanes 2 to 10 were common lambsquarter individuals that survived simazine; lane 11 was a water control; lanes 12 to13 were the control TS common lambsquarters from Herbiseed³.

TS biotype in this study [*]	CCGATTGATCTTTCAATATGCTAGTTTCAACAACTCTCGTTCTTTA
Sample 1	CCGATTGATCTTTCAATATGCTGGTTTCAACAACTCTCGTTCTTTA
Sample 2	CCGATTGATCTTTCAATATGCTGGTTTCAACAACTCTCGTTCTTTA
Sample 3	CCGATTGATCTTTCAATATGCTGGTTTCAACAACTCTCGTTCTTTA
Known TS biotype ^{**}	CCGATTGATCTTTCAATATGCTAGTTTCAACAACTCTCGTTCTTTA
Known TR biotype **	CCGATTGATCTTTCAATATGCTGGTTTCAACAACTCTCGTTCTTTA

Figure 4.3. Alignment of the *psbA* gene sequences in control TS common lambsquarters, the putative TR common lambsquarters

(samples 1 to 3), and that of the known TS and TR biotypes. The position where the A to G mutation occurs is indicated by the box.

^{*} Triazine sensitive common lambsquarters purchased from Herbiseed³ used as control.

98

** Known *psbA* gene sequences in TR and TS common lambsquarters reported by Bettini et al. (1987).

		Wooster		Kingsville							
Simazine	Bare	Shredded	Wheat	Bare	Shredded	Wheat					
-kg/ha	8011	WOOU Dark	plants/f	Four plots	WOOU DAIK	suaw					
8			I	I I							
0	122 A a ^{**}	37 A b	11 A b	92 A a	6 A b	62 A a					
2 5			2.4		z , 1						
2.7	26 B a	13 A b	2 A c	98 A a	5 A b	21 AB ab					
5 /	25 B a	2 A h	0 A b	56 B a	$0 \land c$	11 B b					
* Dlat airs w	25 D d	$\frac{2}{1}$ hr 26		JOD a							

Table 4.1. The effect of simazine treated mulches on total common lambsquarters density^{*}.

^{*} Plot size was 1 by 2.4 m and 1 by 3.6 m in Wooster and Kingsville vineyards,

respectively.

^{**} Values followed by the same uppercase letter in the same column or by the same lowercase letter in the same row were not significantly different from each other at P<0.05 level.

Kingsville Wooster Simazine Shredded Wheat Shredded Bare Bare Wheat soil wood bark soil wood bark straw straw -kg/ha-% TR common lambsquarters 0 8 A a 0 A a 5 B a 0 C a 11 A a 8 B a 0 A a 2.7 67 A a 13 A a 50 A a 38 B a 22 B a -* 100 A a $0 \, A \, b$ 83 A a 100 A a 5.4 -

Table 4.2. The effect of simazine treated mulches on percentage of triazine resistant (TR) common lambsquarters.

^{*} Samples were not collected because the population was 0.

** Values followed by the same uppercase letter in the same column or by the same lowercase letter in the same row at either location were not significantly different from each other at P<0.05 level.

Chapter 5: Soil Moisture, Winter Protection, and Grape Yield and Quality as Affected by Simazine Treated Mulches

Linjian Jiang, Imed Dami, and Doug Doohan*

Soil hilling around grapevines is required for winter protection of grafted and cold sensitive grape cultivars in Ohio. We investigated the effect of simazine treated mulches (STM) on soil water conservation and winter protection, as well as the effect of STM on grape yield and fruit quality. Simazine did not affect any variables measured in this study; however, mulches increased soil moisture content 45 to 75 % at Wooster and 8 to 23% at Kingsville compared with soil-hilling. Mulches also reduced scion rooting by a factor of 2 to 75 compared with soil-hilling. Mulches provided equally good winter protection and did not affect whole plant injury compared with the traditional soil hilling. Although the effect of mulches on grape yield varied on different cultivars from year to year, the overall effect was either positive or neutral. With the exception that sugar content of Auxerrois was increased by 5 % by straw treatment compared with soil-hilling in 2008,

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the fruit quality of grape was not affected by mulching treatment among different cultivars.

Nomenclature: Simazine; grape, Vitis spp.

Key words: Grape quality, grape yield, mulch, simazine.

Vinifera grapes are widely grown for making high quality wines in the North Central United States. However, vinifera grapes are very sensitive to phylloxera, an insect that severely parasitizes the root system. Grafting vinifera grapes onto a phylloxera-resistant root stock has been highly effective in managing this pest. Most vinifera grape cultivars are also cold-sensitive, and the graft union needs protection to survive through winter in most North Central states. Currently, winter protection is achieved by mounding soil, called "soil-hilling", to cover the graft union in the fall. Growers have to remove the hilled soil, called "dehilling", to prevent scion rooting and the subsequent phylloxera infection at the beginning of the next growth season (Dami et al. 2005).

Although hilling effectively prevents winter injury, experience suggests that the practice is not sustainable over the long term. Soil hilling requires fuel, labor and specialized equipment, increasing the cost and the carbon footprint associated with the final wine products. Soil hilling may also exacerbate pesticide offsite movement from vineyards. In previous studies, tillage increased pesticide runoff on various soils (Seta et al. 1993; Fu et al. 2006). Because vineyards usually have soil with good internal water drainage, the additional tillage required for soil hilling is likely to increase pesticide leaching (Gish et al. 1995). This is a particular concern because grape is a pesticide-intensive crop (180 kg ai/ha; USDA 2009). Moreover, soil hilling and dehilling are likely to increase soil erosion, since many vineyards in Ohio are located on hillsides. In the research vineyard at the Ohio State University/OARDC, we observed that soil hilling for 10 years resulted in a loss of the top 8 to 12 cm of soil in a field with a slope of less than 5 degrees. Finally, soil hilling complicates weed control. More weed species at higher

density were found in Ohio vineyards that had been hilled compared to those that were not (Jiang et al. 2008). An alternative to soil hilling that provides equivalent protection against cold injury but without the associated environmental costs is highly desirable.

Zabadal (2003) reported that mulch of wheat straw protected the graft union of vinifera grapes from winter injury as effectively as mounded soil. Mulches are known to effectively reduce soil erosion and chemical run-off (Maass et al. 1988; Smolikowski et al. 2001). Moreover, herbicide treated mulch provided better weed control than herbicide or mulch alone (Case and Mathers 2006). These features indicate that herbicide treated mulches may be a viable alternative to soil hilling and traditional herbicide-based approaches to weed control. However, the effect of herbicide treated mulches on the growth, yield and fruit quality of vinifera grapes has not been studied; moreover, because of the high C to N ratio, organic mulches may decrease N availability in soil and thereby affect the mineral nutrition of the grape plant (Eiland et al. 2001).

In this research, we applied simazine treated mulches (STM) in two vineyards for two years. We hypothesized that STM can replace soil hilling for winter protection and be a sustainable vineyard management tool for vinifera grape production. Our objectives were to determine the effect of STM on 1) winter injury and winter protection, 2) scion rooting, 3) soil moisture, 4) nitrogen level in plant tissues and 5) yield and fruit quality.

Materials and Methods

Field trials. Experiments were conducted in research vineyards of the Ohio Agricultural Research and Development Center at Wooster (40°46'43"N, 81°55'51"W) and at Kingsville (41°53'5"N, 80°41'52"W) in 2008 and 2009. The soil type was Wooster silt loam (pH 4.9, organic matter content 2.38 %, nitrogen content 0.11 % and CEC 12.8 meq/100g) at Wooster and a Bogart loam at Kingsville (pH 6.1, organic matter content 1.98 %, nitrogen content 0.09 % and CEC 6.6 meq/100g). The cultivars included in this trial were grafted Auxerrois (planted in 1994) for two years at Kingsville, own-rooted hybrid Seyval (planted in 2001) for two years at Wooster, and grafted Vitis vinifera Pinot gris (planted in 1994) for one year at both Wooster and Kingsville. The spacing between rows was 3 m for all cultivars, and the spacing distance within a row was 1.8 m for Auxerrois, 1.0 m for Seyval and 1.2 m for Pinot gris. Seyval has a high cordon training system and both Auxerrois and Pinot gris have a vertical shoot position training system. Spur pruning was performed before bud break for all three cultivars. Seyval, Auxerrois and Pinot gris at Kingsville were pruned to 25, 30 and 20 buds per vine, respectively. No cluster thinning was performed to Auxerrois or Pinot gris, while Seyval was thinned to one cluster (the basal one) per shoot. Because of severe bud injury (>95 %) to Pinot gris at Wooster during the winter of 2008/2009, all canes were hedged to have 5 buds. Fertilizers were not applied to vineyards in 2008, and urea was applied two weeks before bloom at a rate of 40 kg N/ha to all cultivars in 2009. Pesticides and fungicides were

applied as scheduled or needed following recommendations by OSU Extension (Dami et al. 2005).

Treatments consisting of mulches (hilled soil, wheat straw and wood bark) treated with simazine at 0, 2.7 and 5.4 kg/ha, were established in vineyards at Wooster and Kingsville. The area that received STM treatment extended approximately 50 cm from the center. Mixed hardwood-species bark (125 tons/ha), or wheat straw (20 tons/ha) was applied by volume using plastic buckets to a depth of 10 cm above the graft union and a depth of 5 cm for the rest of plot area in November each year. The 10 cm high cone around the grape vine had a diameter about 1 m. The standard method of winter protection, soil hilling, was included as the control and covered the graft union up to 20 cm. Prior to the second winter, additional bark or straw was added to the original mulch treatment to restore the mulch to the depths established the previous autumn. This required twenty L of straw or bark around each vine to cover the graft union. Soil was also hilled at the same time for the winter hilling treatment.

Simazine¹ was applied to the top of the mulch or soil at 0, 2.7 and 5.4 kg/ha as an aqueous emulsion on the same day mulch was applied. A CO_2 backpack sprayer with a single 8003EVS nozzle² that delivered 230 L/ha at 240 kPa pressure was used to apply the herbicide. In late June or early July the mounded soil and mulch that covered the graft union were removed from the graft union in each plot. Following the same regime used in autumn, simazine treatments were applied after hand weeding on the same day when the soil hill was removed in spring.
Soil moisture. Two soil cores, 5.3 cm in diameter and 10.5 cm in depth, were randomly taken from each plot and combined together into a zip-locked plastic bag to prevent moisture loss during sampling and transportation. For mulched plots, straw or bark was removed before collecting cores, and replaced after sampling. Soil dry weight was approximated by drying the cores to a constant weight. Constant weight was achieved at 50 °C after 3 days. Soil moisture content was expressed as the percentage of water relative to soil dry weight.

Thermal insulation assessment and whole plant injury evaluation. The probes of temperature loggers³ were placed on the surface or buried 10 cm deep under straw or bark and 20 cm deep under soil before the treatments were applied. Temperature was recorded once every 5 min until late June (2008) and early July (2009) at which time data were downloaded onto the computer. The winter of 2008/2009 was especially cold at Wooster, with an official low temperature of -28.1 °C (OARDC Weather System 2009); virtually, 100 % bud injury was incurred to Pinot gris at Wooster, while less than 10 % bud injury occurred with Pinot gris and Auxerrois at Kingsville and Seyval at Wooster. Therefore, whole plant injury was assessed on Pinot gris in Wooster at the pre-bloom (June 12th) and post-bloom stages (July 9th) according to the following scale: 1, no bud injury; 1.5, more than 75 % bud injury; 2, 1/3 cordon damage; 2.5, 2/3 cordon damage; 3, all cordons are dead; 3.5, top 1/2 part of trunk is damaged on phloem but suckers

emerge from this part; 4.5, scion dies back and root stock is alive; 5, the whole vine is dead.

Scion rooting. At the time of mulch or soil removal from the graft union, the number of new roots (longer than 0.5 cm) generated from the scion of Pinot gris and Auxerrois was recorded.

Nitrogen content. Thirty leaf petioles were collected from each plot, dried at 50 °C for 3 d and ground into a fine powder (<1 mm). Petioles of leaves opposite to the cluster were collected at one month after bloom in 2008 and at bloom stage in 2009 at both Wooster and Kingsville. At veraison, petioles were taken from fully expanded healthy leaves, approximately the fifth to seventh leave counting down from the growing tip. The total nitrogen was determined following the AOAC official methods of analysis (1989).

Grape yield and fruit quality. The harvest dates were September 17th 2008 and September 23rd 2009 for Seyval, September 19th 2008 at Wooster and October 1st 2009 for Auxerrois and October 1st 2009 for Pinot gris at Kingsville. Cluster number and total cluster weight were measured for each vine. A 100-berry sample was randomly collected from each plot at harvest for quantification of fruit quality. After taking the weight data, samples were juiced and centrifuged at 5000 rpm for 5 min. Sugar- and titratable acid (TA) content, and pH of the supernatant were measured. The sugar content was determined by a refractometer⁴. TA content and pH were measured by a pH/EP titration⁵ with 0.1 N NaOH up to pH 8.1 using 10 ml juice. Refractometer readings were expressed as percent soluble solids in aqueous solution at 20 °C and TA values were expressed as grams per liter.

Experimental design and data analysis. A complete randomized block design was adopted in both vineyards with four replications and two vines per treatment. Data were analyzed using ANOVA model by SAS^6 . Means were separated by t test at P<0.05 level.

Results and Discussion

The effect of STM on soil moisture. Simazine application rate did not affect soil moisture; however, mulches increased soil moisture compared with the soil-hilling treatment (Table 5.1). The highest soil moisture was observed under bark (15.9 %), followed by straw (14.0 %) and soil hilling treatment (12.9 %) at Kingsville. An identical pattern was observed at Wooster. Jones et al. (1968) reported that straw mulch increased soil moisture throughout the growing season by 20 to 30 % compared with an unmulched control. Organic mulches are well known for reducing soil water evaporation (Unger and Parker 1976) and therefore increasing soil moisture under the STM in this study. In addition, mulches reduce weed density; thereby, reducing soil water loss

through transpiration (Ateh and Doll 1996). The higher bulk density of bark $(0.25g/cm^3)$ compared with straw $(0.04 g/cm^3)$, and the closer contact of bark fractions were likely factors contributing to its greater resistance to water evaporation. The observed moisture conservation occurring in mulched plots is important because mulching can attenuate drought stress damage in rain-fed vineyards during dry years.

The effect of STM on soil temperature and grape injury. Bark and straw increased the minimum temperature around the graft union as effectively as did hilled soil (Table 5.2). In the winter of 2006/2007, the lowest temperatures recorded under 10-cm of bark and straw were -8.1 and -8.3 °C compared to -19.4 °C on the soil surface. A similar result was also observed at Kingsville in 2007/2008 and 2008/2009. For example, in the winter of 2007/2008, the minimum temperature under bark, straw and soil was 10.2, 9.7 and 10 °C, respectively, higher than that recorded at the soil surface. These results agree with those of Zabadal (2003), who reported that wheat straw could protect the graft union as effectively as hilled soil. Moreover, the thermal insulation by mulches also affected the maximum soil temperature as well as the average soil temperature (Figure 5.1). The monthly average temperature in plots covered by hilled soil, straw or bark was higher than soil surface from November to February; while, soil surface temperature was higher from April onward. The relative lower soil temperature caused by mulches during the growing season has a variety of impacts, including impacts on soil microbial activity and weed emergence (Hamdoun 1972).

STM had no effect on bud injury (data not shown), expect Pinot gris at Wooster in 2009 (over 99 % of primary bud injury). Ten healthy buds per vine were counted at the whole plant injury assessment date of May 11th (Table 5.3). By June a few more shoots had emerged from the trunk. On average, each vine only bore 4 to 5 clusters in July with no differences attributable to STM treatment. The whole plant injury assessed in both June and July varied from 2.3 to 2.9, suggesting that the cordons of most vines were severely injured. Simazine was found to have no effect on winter injury measurements of Pinot gris; neither did the mulch treatment (Table 5.3). However, the lowest number of broken buds, shoots and clusters per vine was observed with the soil hilling treatment; in addition, vines from the soil hilling treatment sustained a higher whole plant injury level. The difference could be significant if more replications were included.

The effect of STM on scion rooting. Simazine application rate did not affect scion rooting of Pinot gris or Auxerrois (Table 5.4). The photosynthetic apparatus in the chloroplast is the target site of simazine; therefore, this result is not surprising. However, mulches significantly affected development of roots from the scion when the hilled soil was removed from plants in mid-June. Fifteen roots developed from the scion of Auxerrois when covered by soil (2008), compared to 3.6 and 0.2 roots per vine when mulched with bark and straw, respectively. Similar results were observed with Pinot gris in 2009. Straw and bark have different physical characteristics than soil, which are apparently less favorable for scion root development. In fact, the respective bulk density was of bark and straw was 0.25 and 0.04 g/cm³, respectively, while the soil bulk density was

1.6 g/cm³. Higher bulk density of soil would have resulted in a closer contact between the vinifera scion and the soil that probably facilitated scion rooting. Though phylloxera infestation was not evaluated in this research it is probable that straw and bark mulch would reduce the chance for phylloxera to infect the scion and the need to remove mulch quickly in the spring relative to the urgency when soil hilling is used. Omer et al. (1995) reported that phylloxera infection promoted the infection of some soil-born fungal pathogens, resulting in serious damage to grapevines. Lotter (1999) pointed out that addition of organic materials into soil created a more favorable condition for the growth of beneficial fungi, which in turn suppress those soil-born pathogens. Therefore, as a means of adding organic matter to the vineyard, STM could play an important role in suppressing secondary fungal infections. Collectively, mulches could very likely reduce grape damage caused by phylloxera as well as the secondary fungal infection.

The effect of STM on nitrogen levels of grape. The N level for all cultivars during bloom or post-bloom stage varied from 0.89 to 1.14 % (Table 5.5), within the normal range for grapevines (Dami et al. 2005). The N level of Pinot gris and Auxerrois was higher at bloom than at veraison in 2009. Bates and Wolf (2008) had observed that such fluctuation was normal N dynamics as grape plants develop. However, N level was higher at veraison than at bloom for Seyval in 2009, suggesting that different cultivars had different N metabolism.

Simazine application rate did not affect petiole N level at either bloom or veraison. In contrast, petiole N content of each cultivar (Seyval, Auxerrois and Pinot gris) was

significantly affected by both wheat straw and wood bark mulches (Table 5.5). Since the C to N ratio for both bark and straw used in this study was over 100, it was expected that the application of bark and straw would decrease the soil N available for plants (Eiland et al. 2001). We observed that bark reduced N level at about two weeks after bloom for Auxerrois in 2008, corresponding well to this expectation. However, in 2009 when urea was included in the management regime, no reduction in petiole N was observed. Rather, straw increased N level at bloom for Seyval (8 % increase compared with bare soil treatment), and at bloom (16 %) and veraison (12 %) stages for Pinot gris. However, mulches had no effect on N level of Auxerrois at either bloom or veraison stage in 2009.

Because no N fertilizer was applied in 2008, biodegradation of straw and bark probably decreased the soil N available for grape (Eiland et al. 2001), resulting in the lower level of petiole N for Auxerrois in 2008. In 2009, the effect of mulch addition on petiole N level in Auxerrois appeared to have been offset by the application of urea (Table 5.5). Rainfall that occurred on the same day when urea was applied at both Kingsville (1.3 mm) and Wooster (15.5 mm) likely enhanced N efficiency. A year to year variation in grape petiole N level was also observed by Agnew et al. (2005) in response to organic mulch treatment, and they attributed this variation to the different amounts of rainfall that occurred 6 weeks before the sampling date in each year. The year to year differences we observed could also have been influenced by sampling dates. It is possible that cultivars may responds differently to mulch; for example, mulch significantly affected the N level in petioles of Auxerrois but not of Seyval in 2008 (Table 5.5). Higher N content in the Wooster soil (0.11 %) than in the Kingsville soil (0.09 %), as well as other different environmental factors between Wooster and Kingsville, were probably influencing factors.

Since mulch was applied only to the soil surface, decomposition was likely slow. Under these circumstances addition of N fertilizer appears to have been an effective tool to regulate grape N levels. Our data supported this notion: when urea was applied, straw increased N level at both sampling dates for Pinot gris and at one sampling date for both Seyval and Auxerrois. Suppression of weeds by straw may also have contributed to better crop nutrition because of less interspecies competition. Porosity of the straw enabled urea pellets to fall to the soil surface where the N content was protected from vaporization (data not reported). Furthermore, the soil under straw had a higher moisture content than bare soil (Table 5.1), facilitating mineralization of the N and assimilation by plants.

Bark also provided significant weed control, which would increase the availability of N for grapes. However, in this case the potential benefit of reduced nutrient competition may have been offset by bark preventing urea pellets from falling to the soil surface. Most of the pellets stayed on the surface of the bark mulch, reducing the amount of urea deposited to soil. This was a probable reason why the enhancement of N level by bark was not as significant as by straw.

Together our results indicated that the potential negative impact of mulches on grape N level could be offset by applying N fertilizer in a traditional vineyard. Mulch is likely to be an attractive management tool for organic vineyards in which case additional compost fertilizer may be required to prevent N immobilization by the mulch. Mulch has significant impact on soil moisture and temperature, which are important factors for the cycling of mineral nutrients in the soil. Therefore, the impact of mulch on other mineral nutrients needs to be explored in order to further understand the long-term effects on vineyard management.

The effect of STM on grape yield and fruit quality. Simazine application rate did not affect any variable measured: grape yield, berry size, sugar content, TA content or pH in these experiments (Tables 5.6, 5.7 and 5.8). With only a few exceptions (Auxerrois in Table 5.6; Seyval in Table 5.7) mulch did not affect yield and fruit quality. Variables affected were sugar content (Auxerrois), and yield and pH (Seyval). In 2008, the highest sugar content in Auxerrois berries was observed with the soil-hilling treatment (17.8 %) and was significantly higher than that observed in fruit from the straw mulch treatment (16.9 %). In 2009 fruit from the soil-hilling treatment also had the highest value in sugar content, although it was not significantly different from that measured in fruit from bark or straw mulched plots (Table 5.6).

Mulch increased yield of Seyval by 60 % and 44 %, respectively, in bark and straw treatments compared with the soil-hilling treatment in 2009 (Table 5.7). This effect was not significant in 2008. Further analysis of cluster number and size revealed that both yield components contributed to the increase observed in 2009 (Table 5.9). Bark and straw increased cluster number per vine by 32 % and 22 %, respectively, compared with the soil-hilling treatment. Similarly, both led to a 0.18 kg cluster, which was 20% heavier than that of the soil-hilling treatment. Mulches also significantly increased pruning weight when compared with the soil-hilling treatment (Table 5.9), 0.10 kg per vine with

the soil-hilling treatment, compared to 0.13 kg per vine with straw and 0.16 with bark. Since each vine was pruned to have the same number of buds, pruning weight was a good indicator of the growth vigor during the same growing season (Dami et al. 2005). These data indicated that mulch enhanced vine size during the growing season of 2008. Phadung et al. (2005) reported that grape shoot length was enhanced by straw mulch, also suggesting that mulches could promote grape vigor.

Conserving soil moisture, which guarantees a stronger growth during the drought period, is a likely explanation for the enhancement in grape vigor in response to mulch (Table 5.1). This was supported by the recorded precipitation data (OARDC Weather System 2009): a total of 472 mm rainfall occurred during the growing season of 2008 (May 1st to November 1st), but the entire August had only 32 mm precipitation. Similarly, in the growing season of 2009 a severe drought period was also recorded: the total precipitation was only 25 mm from June 21st to July 21st. Therefore, the enhancement of yield in Seyval by mulches in 2009 could be a two year effect of the treatments.

Mulches conserved soil moisture likely by reducing the evaporation from the soil surface and indirectly through suppression of weed competition for soil water. This effect enabled grape vines to better resist extreme drought stress in the growing season. Mulch provided winter protection that was as effective as the traditional soil-hilling. In contrast to soil hilling, mulches did not stimulate scion rooting; thereby, leaving a wide time interval for removal of mulches from the scion in spring. Although a reduction petiole N was observed one year, the subsequent year data indicated that the impact could be offset or even reversed by N fertilization. The response of yield and fruit quality to mulch treatments varied depending on cultivars and years. A long term study including more cultivars is needed to better understand the impact of mulches on the sustainability of grape production.

Source of Materials

- ¹ Princep® 4L, Syngenta Crop Protection, P.O. Box 18300, Greensboro, NC 27419.
- ² Spraying System Co., P.O. Box 7900. Wheaton, IL 60189.
- ³ StowAway® Tidbit® XT, P.O. Box 3450, Pocasset, MA 02559.
- ⁴ Reichert Inc., 3362 Walden Avenue, Depew, NY 14043.
- ⁵ Model 350, Denver Instrument, 1401 17th Street, Denver, CO 80202.
- ⁶ SAS 9.1, SAS Institute Inc. 100 SAS Campus Drive, Cary, NC 27513.

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Treatment	Kingsville	Wooster		
_		— % —		
Simazine				
-kg/ha-				
0	14.5 a^{**}		17.9 a	
2.7	14.1 a		17.9 a	
5.4	14.2 a		18.2 a	
Mulch				
Bare Soil [†]	12.9 c		12.8 c	
Bark	15.9 a		22.5 a	
Straw	14.0 b		18.6 b	

Table 5.1. The effect of simazine treated mulch on soil moisture (%) in 2008^{*}.

^{*} At Kingsville, soil samples were taken at 4 d after the most recent rainfall occurred from September 30th to October 3rd, totaling 49.3 mm; and at Wooster, soil samples were taken at 15 d after the most recent rainfall which occurred from 12th to 14th, September, totaling 41.9 mm.

^{**} Means followed by the same letter were not significantly different from each other at P<0.05 in the same column for either simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in June.

Treatment [¶]	Wooster	Kingsville				
	February 6 th 2007 *	February 11 th 2008	March 2 nd 2009			
Soil surface	-19.4 b ^{**}	-11.7 b	-11.5 b			
Bark	-8.1 a	-1.5 a	-1.5 a			
Straw	-8.3 a	-2.0 a	-7.0 ab			
Soil	-12.2 ab	-1.7 a	-5.7 ab			
Air temperature [†]	-21.3	-16.1	-12.8			

Table 5.2. The minimum temperature (°C) recorded during winter months (November to April).

^{*} Date when minimum temperature was recorded.

** Means followed by the same letter were not significantly different from each other at P<0.05 level in the same column.

[†] According to OARDC Weather Station, available at http://www.oardc.ohio-state.edu/newweather.

[¶] Temperature was recorded under 10 cm of bark, 10 cm of straw and 20 cm of soil.

	Broken Buds/vine	Shoot Number/vine	Cluster Number/vine	Whole Pla	nt Injury [*]
Treatment	Bud Break	Pre-Bloom	Post-Bloom	Pre-Bloom	Post-Bloom
	$(May 11^{th})$	$(June 12^{th})$	(July 9 th)	(June 12 th)	(July 9 th)
Simazine					
-kg/ha-					
0	$10.8 a^{**}$	18.0 a	4.0 a	2.3 a	2.4 a
2.7	10.3 a	15.7 a	4.4 a	2.5 a	2.8 a
5.4	12.5 a	15.9 a	5.0 a	2.8 a	2.7 a
Mulch					
Soil^\dagger	7.8 a	12.4 a	3.8 a	2.9 a	2.9 a
Bark	12.3 a	18.1 a	4.7 a	2.4 a	2.5 a
Straw	13.4 a	19.0 a	5.0 a	2.5 a	2.4 a

Table 5.3. Winter injury assessment of Pinot gris at Wooster in 2009.

* The whole plant injury was assessed based on the following scale: 1, no bud injury; 1.5, more than 75 % bud injury; 2, 1/3 of the cordon is damaged; 2.5, 2/3 of the cordon is damaged; 3, all cordons are dead; 3.5, top 1/2 part of trunk is damaged on phloem while suckers emerge from the top 1/2 part; 4, bottom 1/2 part of truck is damaged on phloem but suckers emerge from the bottom 1/2 part; 4.5, scion dies back and root stock is alive; 5, the whole vine is dead.

^{**} Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for either simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in the summer.

Treatment	Auxer	rois 2008	Auxerro	Pinot gris 2009		
			Root nur	mber/vine —		
Simazine						
-kg/ha-						
0	2.4	a**	1.3	a	7.0	а
2.7	7.6	a	0.8	а	4.6	а
5.4	9.0	a	2.7	а	6.8	а
Mulch						
Bark	3.6	b	1.6	ab	4.5	b
Soil^\dagger	15.1	a	3.2	а	11.2	a
Straw	0.2	b	0.1	b	2.8	b
Straw	0.2	b	0.1	b 2000	2.8	b

Table 5.4. The effect of simazine treated mulches on scion rooting at Kingsville, Ohio^{*}.

^{*} Soil or mulch was removed on 6-19-2008 and 6-23-2009.

** Means followed by the same letter are not significantly different from each other at

P<0.05 level in the same column for either simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in the summer.

Table 5.5. The effect of simazine treated mulch on total nitrogen content in leaf petioles of Seyval at Wooster, Ohio and Auxerrois and Pinot gris at Kingsville, Ohio.

	Auxerrois			Seyval	Pinot gris				
Tractmont	2008^{*}	4	2009**	2008	2	2009		2009	
Treatment	Post-Bloom	Bloom	Veraison	Post-Bloom	Bloom	Veraison	Bloom	Veraison	
	(July 22 nd)	(July 5 th)	(September 2 nd)	(July 21 st)	(June 24 th)	(August 10 th)	(July 5 th)	(September 2 nd)	
				% w/	/w				
Simazine									
-kg/ha-									
0	1.06 a [¶]	1.00 a	0.67 a	0.90 a	0.91 a	1.02 a	1.05 a	0.77 a	
2.7	1.04 a	0.97 a	0.65 a	0.92 a	0.89 a	1.03 a	1.06 a	0.80 a	
5.4	1.14 a	0.98 a	0.67 a	0.89 a	0.92 a	1.05 a	1.11 a	0.80 a	
Mulch									
\mathbf{Soil}^{\dagger}	1.14 a	0.91 a	0.64 a	0.89 a	0.86 b	1.02 a	0.99 b	0.76 b	
Bark	1.02 b	1.02 a	0.67 a	0.93 a	0.92 a	1.05 a	1.08 ab	0.77 b	
Straw	1.08 ab	1.01 a	0.67 a	0.89 a	0.93 a	1.03 a	1.15 a	0.85 a	

* No urea was applied.

^{**} Urea (40-0-0) at the rate of 40 kg N ha⁻¹ was applied to Seyval on June 1st, 2009 and to Auxerrois and Pinot gris on June 11th, 2009.

[¶] Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for either simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in the summer.

Tuestas	Yi	eld	Berry	y Size	Sugar C	Content	Tartar	ic Acid	pН	I
Treatment	2008^{*}	2009^{**}	2008	2009	2008	2009	2008	2009	2008	2009
	— kg/	vine —	— g/100	berries —	%	<u></u>	g	/L ——		
<i>Simazine</i> -kg/ha-										
0	7.9 a [¶]	2.8 a	201 a	179 a	17.3 a	18.8 a	0.54 a	0.62 a	3.37 a	3.31 a
2.7	7.1 a	2.9 a	211 a	177 a	17.3 a	19.0 a	0.54 a	0.63 a	3.39 a	3.30 a
5.4	8.3 a	2.3 a	198 a	177 a	17.4 a	19.0 a	0.53 a	0.63 a	3.42 a	3.34 a
Mulch										
\mathbf{Soil}^{\dagger}	7.3 a	2.8 a	201 a	178 a	17.8 a	19.1 a	0.53 a	0.62 a	3.41 a	3.32 a
Bark	7.5 a	2.2 a	205 a	176 a	17.2 ab	19.0 a	0.54 a	0.63 a	3.38 a	3.33 a
Straw	8.5 a	3.0 a	205 a	180 a	16.9 b	18.8 a	0.54 a	0.62 a	3.39 a	3.30 a

Table 5.6. The effect of simazine treated mulch on yield and fruit quality of Auxerrois grapes harvested in 2008 and 2009 at Kingsville, Ohio.

* No urea was applied.

^{**} Urea was applied at the rate of 40 kg N ha⁻¹ on June 11th, 2009

[¶] Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for either

simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in the summer.

Treatment	Yiel		Berry	y Size	Sugar	Content	Tartar	ic Acid	pI	H
Treatment	2008^*	2009**	2008	2009	2008	2009	2008	2009	2008	2009
	— kg/	′vine —	— g/100	berries —	0	%	g	/L		
Simazine										
-kg/na-	10 9	2.0	101	106	22.0	22.1	0.70	0.70	0.1.6	2.22
0	1.8 a	3.9 a	191 a	196 a	22.0 a	22.1 a	0.79 a	0.70 a	3.16 a	3.33 a
2.7	1.9 a	3.5 a	193 a	185 a	22.0 a	22.3 a	0.76 a	0.66 a	3.19 a	3.35 a
5.4	1.8 a	3.5 a	186 a	194 a	21.9 a	22.2 a	0.78 a	0.67 a	3.16 a	3.33 a
Mulch										
Soil^\dagger	1.6 a	2.7 b	194 a	186 a	22.0 a	22.3 a	0.78 a	0.66 a	3.14 b	3.33 a
Bark	2.0 a	4.3 a	185 a	193 a	21.8 a	22.0 a	0.77 a	0.69 a	3.18 a	3.34 a
Straw	1.9 a	3.9 a	192 a	194 a	22.0 a	22.3 a	0.78 a	0.67 a	3.18 a	3.34 a

Table 5.7. The effect of simazine treated mulch on yield and fruit quality of Seyval harvested in 2008 and 2009 at Wooster, Ohio.

^{*} No urea was applied.

^{**} Urea was applied at the rate of 40 kg N ha⁻¹ on June 1st, 2009

[¶] Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for either simazine or mulch treatment.

[†] Soil was mounded to cover the graft union over the winter and removed in the summer.

Treatment	Yield	Berry Size	Sugar Content	Tartaric Acid	pН
	– kg/vine –	— g/100 berries —	<u> </u>	— g/L —	
Simazine					
-kg/ha-					
0	$4.1 a^*$	132 a	21.9 a	0.87 a	3.27 a
2.7	4.2 a	135 a	22.3 a	0.85 a	3.30 a
5.4	4.0 a	130 a	22.0 a	0.89 a	3.28 a
Mulch					
Soil ^{**}	3.9 a	130 a	22.1 a	0.86 a	3.27 a
Bark	3.9 a	133 a	22.1 a	0.87 a	3.28 a
Straw	4.5 a	135 a	22.1 a	0.88 a	3.30 a

Table 5.8. The effect of simazine treated mulch on yield and fruit quality of Pinot gris at Kingsville, harvested on 10-19-2009.

116

* Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for

either simazine or mulch treatment.

** Soil was mounded to cover the graft union over the winter and removed in the summer.

Table 5.9. The effect of simazine treated mulches on cluster number, cluster size and pruning weight of Seyval at Wooster and Auxerrois at Kingsville in 2008.

		Seyval			Auxerrois				
Treatment	Cluster	Cluster Size	Pruning Weight [¶]	Y/P Ratio [†]	Cluster	Cluster Size	Pruning Weight [¶]	Y/P Ratio	
	-Number/vine-	-kg/cluster-	-kg/vine-		-Number/vine-	-kg/cluster-	-kg/vine-		
<i>Simazine</i> -kg/ha-									
0	$22 a^*$	0.18 a	0.14 a	14.6 a	65 a	0.11 a	0.82 a	9.7 a	
2.7	22 a	0.16 a	0.12 a	18.7 a	58 a	0.11 a	0.81 a	10.0 a	
5.4	22 a	0.16 a	0.12 a	17.0 a	67 a	0.10 a	0.98 a	8.6 a	
Mulch									
${\rm Soil}^{**}$	18 b	0.15 b	0.10 b	17.8 a	57 a	0.10 a	0.99 a	7.4 a	
Bark	24 a	0.18 a	0.16 a	14.2 a	66 a	0.11 a	0.78 a	9.7 a	
Straw	22 a	0.18 a	0.13 ab	18.2 a	67 a	0.11 a	0.84 a	11.2 a	

* Means followed by the same letter are not significantly different from each other at P<0.05 level in the same column for

either simazine or mulch treatment.

** Soil was mounded to cover the graft union over the winter and removed in the summer.

[¶] Pruning weight was collected in spring of 2009.

[†] Yield to pruning weight ratio = Yield per vine (kg) / Pruning weight per vine (kg).



Figure 5.1. Monthly maximum (a), minimum (b) and average (c) temperature recorded from November 2007 to July 2008 at Kingsville. A similar pattern was observed at Wooster.

Chapter 6: The Effect of Straw Mulch on Simulated Simazine Leaching and Runoff

Linjian Jiang, Imed Dami, Hannah Mathers, Warren Dick, and Doug Doohan*

In the Midwestern United States, winter hilling, consisting of two tillage activities per year, is required in vinifera vineyards for winter protection. However, this practice often causes severe soil erosion and pesticide offsite movement. The effectiveness of wheat straw mulch as a replacement for soil mounding was investigated as a way of not only providing winter protection but also for mitigating pesticide leaching and runoff. A laboratory simulation was conducted where simazine was applied to wheat straw or bare soil and then followed by simulated rainfalls. When compared with bare soil, straw reduced simazine leaching and runoff by 40 % and 68 %, respectively and the remainder was recovered from the straw at the end of the leaching and runoff trials. Absorption and/or interception of simazine by straw were responsible for this effect. Additionally, straw reduced soil erosion by 95 % which also contributed to decreased simazine runoff.

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The first simulated rainfall contributed 70 % and 34 % of total simazine runoff from bare soil and straw, respectively. In conclusion, mulching with straw during winter months to provide winter protection is also likely to be an efficient tool to control simazine offsite movement and soil erosion in vinifera vineyards.

Nomenclature: simazine; wheat, *Triticum spp*.

Key words: Leaching, runoff, simazine, straw mulch.

According to the Agricultural Chemical Usage Database (USDA, 2009) a typical vineyard in the US was treated with about 180 kg ai ha⁻¹ of pesticides in 2003. The area under the trellis is the target for most soil active herbicide applications and the ultimate depository of other pesticides applied to the foliage. Barber and Parkin (2003) estimated that about 30 % of all pesticides were deposited into soil under the trellis at the time of application. As time progresses, as much as 80 % of a given pesticide application could be deposited into soil under the trellis (Courshee 1960). Thus, the area under the trellis may function as a point source of various pesticides subject to leaching and runoff.

A common practice used by vinifera grape growers in regions where winter injury is a threat is to cover the graft union with an insulating layer of soil just before winter. This practice requires tillage under the trellis twice a year; first to mound the soil for winter protection and then to remove it in the spring in order to prevent scion rooting. These tillage practices are likely to increase soil erosion and pesticide runoff (Seta et al. 1993; Fu et al. 2006; Montgomery 2007). However, the effect of tillage on pesticide leaching varies depending on soil types (Sadeghi et al. 2000). It increased atrazine leaching in well-drained soils (Gish et al. 1995) while it reduced pesticide leaching in poorly-drained soils (Rothstein et al. 1996; Siczek et al. 2008). Vineyards are usually established on well-drained soils with excellent internal drainage or soils improved by installation of subsurface tiles (Dami et al. 2005). Since increased internal water flow could also increase chemical leaching (Rothstein et al. 1996; Gish et al. 1995), pesticide leaching in vineyards has a great potential to cause groundwater contamination.

One major group of pesticides frequently found in ground and surface water is the triazine herbicides (Ritter 1990). Simazine is the only triazine herbicide registered for grape and was extensively applied to over 20 % of vineyards in the US in 2005 (USDA 2006). Spurlock et al. (2000) reported that 97 % of wells (n=33) were contaminated by simazine in two counties of California where grapes were a major crop. Various mitigation methods have been employed to reduce pesticide offsite movement, including grassed buffer strips, conserved tillage, constructed wetlands and mulching (Reichenberger et al. 2007). Grassed strips have been widely used in vineyards between rows to protect the soil and reduce pesticide movement (Dami et al. 2005; Reichenberger et al. 2007). However, grass growth in the area under the trellis is not an option because grapes will not tolerate competition for nutrients and water. Additionally, soil tilth is required under the trellis in northern states so that soil can be mounded for winter protection.

Mulching with straw interested us because previous research had shown it to be an effective substitute for mounding soil under the trellis for winter protection in vinifera vineyards (Zabadal 2003). Moreover, simazine treated straw improved weed control in our field trials (Jiang et al. 2008). This enhancement in weed control by straw may be due to increased simazine retention by straw relative to soil, suggesting in turn that straw mulch may be an effective tool to reduce offsite movement of the herbicide. Enhanced weed control would be an additional benefit for using straw mulch in vinifera vineyards. However, the literature indicated little information is available regarding simazine absorption and desorption from straw. Previously, Martin et al. (1978) had shown that

over 20 % of the triazine herbicide cyanazine could be recovered from corn stubble at the end of a laboratory washoff trial. Similarly, Dao (1991) reported that straw also absorbed another triazine herbicide, metribuzin, and its *S*-ethyl analog. However, metribuzin and *S*-ethyl metribuzin were absorbed differently by straw although they have a very similar structure. Therefore, it is likely that each triazine herbicide will interact independently with different mulch materials.

We hypothesized that straw mulch applied as a winter protection treatment, could also be a useful tool for reducing pesticide offsite movement from vineyards. To test this hypothesis, we conducted a laboratory simulation study to investigate the dynamics of simazine when applied to straw and subjected to simulated rainfalls. The objective of this research was to examine how simazine leaching and runoff were affected by straw mulch and simazine application rate under controlled environmental conditions.

Materials and Methods

Location, soil and wheat straw. Simulated simazine leaching and runoff experiments were conducted during the summer months in an unheated building located at the Ohio Agricultural Research and Development Center (OARDC) in Wooster, Ohio. The soil used in both trials was Wooster silt soil with the following profile: pH 7.2, organic matter content 4.01 %, and CEC 10.7 meq 100g⁻¹. Wheat straw was grown on the OARDC farms. To assure good uniformity, all straw used in this trial (about 15 kg) was taken

from the same single bale. Milled straw had a pH of 7.08, nitrogen content of 0.54 % and carbon content of 46.9 %.

Leaching trials. Soil was sieved through a 6 mm screen and well mixed to achieve a uniform texture. It was then placed into a polyvinylchloride (PVC) column used to simulate herbicide leaching or a plastic tray used to simulate runoff. Wheat straw cut to a uniform length of 2 to 4 cm was placed on the surface of soil in columns or trays to simulate mulching. Straw was omitted from columns or trays that simulated the unmulched treatments. Both leaching and runoff studies were conducted on specially designed tables, that allowed leachate or runoff water to be collected from beneath.

Leaching trials used a PVC column (25 cm in diameter 15 cm in length) with a 1 mm grid sieve glued to the bottom to retain soil while allowing water to leach through. Soil was compressed to form a 9 cm thick soil column with a bulk density around 1 g cm⁻³. Soil columns were initially tested for internal drainage. The columns used in this experiment were those that retained some water on the soil surface 4 h after application but were well drained after 24 h. Thirty g of wheat straw were uniformly spread over the soil surface. This amount of straw is equivalent to an application of 6000 kg ha⁻¹. Soil and straw in the columns were then saturated by water and drained for one d before simazine was applied. A water suspension of simazine¹ was applied to the surface of the straw or bare soil at rates of 5.4 (26.5 mg pure simazine per column) and 10.8 kg ha⁻¹ (53.0 mg per column). Ten ml of the simazine water suspension (2000 L ha⁻¹) was applied gravimetrically to each column. Twenty-four h later, a 20 mm rainfall was simulated by

sprinkling 1 L of tap water into each column at the rate of 500 mm h^{-1} using a watering can. The simulated rainfalls caused ponding water, which leached through the soil column within 24 h. The simulated rainfall was applied to each column every other d for a total of 3 times. The total volume of leachate after every 3 simulated rainfalls was recorded and water samples were collected to represent the overall leaching in response to these 3 simulated rainfalls. A total of 12 simulated rainfalls (20 mm each) were applied. By allowing every third rainfall to drain for 2 d, the sample collection was adjusted to be on a weekly basis and continued for 4 wk. After wk 4, all straw was collected from straw mulched PVC columns. Two soil cores through the entire depth of the soil layer were then taken from each column with a 53-mm diameter probe. Soil and straw samples were air dried under an electrical fan for 3 d at room temperature in the lab. All samples were stored at -20 °C until extraction for simazine quantification.

Runoff Trials. Plastic trays (51 by 25 by 6 cm) were used to hold 1.5 kg of soil in simulated runoff trials. Trays were adjusted to have a 10 % slope along the 51 cm axis of the tray. Soil was put into trays starting from the lower 25 cm long wall and covered an area of 25 by 20 cm in each tray. A v-shape notch was made at the 25 cm long wall for collection of runoff. The surface of the soil was level with the bottom of this notch. A 6 by 8 cm plastic screen (1 mm mesh) that allowed both water and fine particles to run through was placed between soil and the notch. Mulch and simazine were then applied following the procedures outlined for the leaching trial. One d after simazine treatment and daily thereafter, a 10 mm simulated rainfall was applied to the surface of each tray

using a watering can. This was equivalent to a simulated rainfall of 500 mm h⁻¹. The volume of runoff water 24 h after each simulated rainfall was recorded and water samples were collected 6 times on a daily basis. Two soil cores through the entire depth of the soil layer were taken from each plastic tray with a 53-mm diameter probe. Straw on the soil surface was also collected. Both soil and straw samples were dried under a fan in the lab for 3 d and stored at -20 °C until simazine quantification.

Simazine extraction and quantification. To extract simazine, 1 ml of toluene and 1 ml of water were mixed together in a sealed glass vial, and shaken at 100 rpm overnight. Simazine was extracted from straw or soil by mixing 1 g of material with 10 ml of deionized water and 10 ml of toluene in a sealed plastic beaker and shaking at 100 rpm overnight.

Water samples collected from the leaching columns were clear with little visible soil sediment. Water from the runoff trays carried a significant amount of soil sediment. In the case of the runoff trays, simazine in the soil:water suspension and in clear water (that is the supernatant after centrifugation of the original sample at 5000 rpm for 5 min) were analyzed separately for simazine content. The difference in simazine content between the soil:water emulsion and clear water was defined as suspended simazine, including simazine absorbed by soil sediment and fine particles of formulated herbicide.

Two μ l of toluene were pipetted from the upper layer of each extract and directly subjected to gas chromatography². The instrument was equipped with a capillary column and nitrogen-phosphorus detector in which simazine was detected at 300 °C. The carrier

gas was helium at a constant rate of 1.2 ml min⁻¹. The injector temperature was 280 °C and the oven temperature was programmed as following: (1) 90 °C for 0.5 min, (2) increase at 15 °C min⁻¹ until 160 °C, and (3) increase at 25 °C min⁻¹ until 280 °C and held for 5 min at 280 °C. The retention time of simazine was 8.45 min under these conditions. A standard curve for the range of 0.1 to 5 mg L⁻¹ was prepared using analytical-grade simazine (99.9 %)³. The average recovery rate was over 95 % from water, soil and straw.

Soil erosion quantification. Ten ml of original runoff water was centrifuged at 5000 rpm for 5 min, and was dried in an oven at 50 °C after discarding the supernatant. The soil sediment was then weighed using a scale with a limitation of 0.0001 g.

Statistical analysis. A complete randomized design was used in this trial with 4 replications. Mulch (straw-mulched or bare soil) and simazine application rate (0, 5.4, 10.8 kg ha⁻¹) were the two independent variables, and simazine leaching and runoff were dependent variables in this trial. Data were subject to an ANOVA model⁴ and means were separated by a t test at 0.05 level.

Results and Discussion

Effect of mulch and herbicide rate on simazine leaching. Straw modified the simazine leaching pattern due to the interception and/or absorption of simazine. Even after extensive simulated precipitation (a total of twelve 20-mm rainfalls within 4 wk), the straw mulch treatment still reduced the cumulative simazine leaching by almost 50 % compared with the un-mulched treatment (Figure 6.1b and Table 6.1). The reduction in leaching due to the straw mulch was most pronounced during the first 3 wk after herbicide application and declined over the course of the experiment. By the fourth wk simazine content was similar in leachate from mulched and non-mulched soil columns. The average simazine leaching during wk 1 through 4 from straw mulched columns was about 1/10, 1/3, 1/2 and 1 times the average simazine leaching from bare soil columns, respectively (Figure 6.1a). Over the course of the experiment, straw reduced the cumulative quantity of simazine that leached from the soil columns by 10, 3.3, 2.5 and 2 times at wk 1, 2, 3 and 4, respectively, when compared with un-mulched treatments (Figure 6.1b). The total simulated precipitation (240 mm) leached through soil columns in 4 wks is similar to the average total 3-month cumulative rainfall for Columbus, Ohio (NCDC 2009). These results indicate that straw is likely to reduce cumulative simazine leaching for several months under field conditions where rainfall intensities and amounts would be much less.

Increasing simazine application rate did not enhance leaching (P<0.05) on either bare soil or straw mulch treatment (Figure 6.1). An effect of application rate was likely masked by 1) the packed conditions in the soil columns and 2) the low water solubility of simazine. The packed columns in this trial lacked macropores, such as earthworm channels, known to play a major role in the vertical movement of pesticides (Shipitalo et al. 2000). The low solubility of simazine has also been previously shown to limit vertical movement in the soil. Cogger et al. (1998) reported that most applied simazine was restricted to the top 15-cm layer of soil.

Increasing simazine application rate did increase the quantity of simazine retained by soil after 12 simulated rainfalls. When simazine was applied to bare soil at 10.8 kg ha⁻¹, 15.1 mg of simazine were recovered in contrast to 5.1 mg of recovered simazine from bare soil columns that had been treated with 5.4 kg ha⁻¹ (Table 6.1). Straw also reduced the quantity of simazine that remained in soil compared to an application to bare soil. However, this affect was only significant at the 10.8 kg ha⁻¹ application rate (Table 6.1). Our data reported here and research by others (Crutchfield et al. 1986; Banks and Robinson 1986) demonstrate that straw can reduce herbicide deposition to the soil surface by a factor of 4- to 9-fold. This is much greater than the 2 x difference in application rate.

In addition to physical interception, slow release of simazine from straw, was likely another important factor. Dao (1991) demonstrated that straw could be used as a slow release carrier for herbicides. The capability of straw to absorb metribuzin was about 10 times that of soil. Similarly, in our runoff experiment, we observed that simazine was primarily released from straw in the dissolved form (Table 6.2). However, because simazine has a low water solubility (5 mg L^{-1} at 20 °C), its release from straw was limited. Extensive interception of the herbicide, coupled with slow release endowed straw with a significant capacity to reduce herbicide offsite movement (Dao 1995; Sigua et al. 1993).

The average simazine recovery rate by toluene extraction was over 95 % from water, soil and straw; however, only 40 to 67 % of total applied simazine was recovered from the pooled experimental fractions. The reported half life of simazine varies from 28 to 149 d (Wauchope et al. 1992). Thus an estimated 50 % degradation of the simazine applied is a reasonable explanation for the simazine recovery observed in this experiment (t=29 d). This explanation is also supported by the higher simazine recovery rate in our runoff experiment which varied from 63 to 89 % over the course of only 7 d (Table 6.3). Interestingly, the overall simazine recovery rate from the straw mulched treatment was significantly higher than that from un-mulched treatment, suggesting that straw protected simazine from degradation. For example, at 10.8 kg ha⁻¹ in this leaching trial, 67 % of applied simazine was recovered from straw mulched columns, whereas only 40 % of applied simazine was detectable from bare soil columns (Table 6.1). A similar pattern was also observed in the runoff trial (Table 6.3). Soil microorganisms play an important role in simazine biodegradation (Kodama et al. 2001). The higher C to N ratio in straw (87:1) may have suppressed microorganism activity (Eiland et al. 2001). Moreover, straw intercepted a large portion of applied simazine; thereby, straw physically isolated the intercepted simazine from soil microbes, contributing to the higher simazine recovery rate under mulching treatment.
The effect of application rate and mulch treatment on simazine runoff. Simazine runoff was affected by both simazine rate and straw (Table 6.4). Straw reduced runoff regardless of simazine rate because 1) it intercepted the herbicide at the time of application, 2) released simazine slowly to the soil and 3) reduced soil erosion and thereby simazine runoff associated with soil erosion.

Due to the interception of simazine by straw, the magnitude of decline in runoff from straw mulched treatments was much less than that of un-mulched treatments from d 1 to d 2 (Table 6.2). Specifically, 1.33 mg of simazine ran off from the straw mulched treatment on d 1, followed by 0.55 mg on d 2. In contrast, simazine runoff from bare soil was 7.24 and 1.3 mg on d 1 and d 2, respectively.

Statistically, the same amount of dissolved and suspended simazine ran off from straw mulched treatments on d 1. However, there was always more dissolved simazine runoff from straw than suspended simazine for the balance of the trial (Table 6.2). Cumulatively, dissolved simazine runoff accounted for over 70 % of the total simazine runoff from straw, indicating total simazine runoff from straw occurred primarily in the dissolved form. Since simazine has low water solubility (5 mg L^{-1} at 20 °C), the release of simazine from straw was also at a low but constant level. This was because straw intercepted and bound most of the applied simazine.

Straw mulch reduced total simazine runoff on d 1 and 2 relative to applications made to bare soil. However, total simazine on d 5 and 6 was greater in runoff from straw mulched soil than in runoff from bare soil. This pattern was also observed with dissolved and suspended simazine (Table 6.2). The reduction of simazine runoff by straw on d 1 and 2 was likely due to the interception of simazine by straw (Table 6.3). However, the increase of simazine runoff from straw during the late stage of the experiment (d 5 and 6) was most likely due to the following two factors. (1) Simazine that was prone to wash off (mainly suspended simazine particles) was rapidly removed from bare soil as time progressed. Therefore, total simazine runoff declined to a low level by d 3 at which time it was primarily occurring in the dissolved form. (2) Straw retained more than 50 % of applied simazine even at the end of the experiment, facilitating a more consistent release of dissolved simazine at a higher concentration than from the un-mulched treatment by d 5 (Table 6.2).

In contrast to the column leaching study, herbicide rate influenced simazine runoff. By the end of the experiment more simazine runoff had occurred at the higher application rate of 10.8 kg ha⁻¹ than at the 5.4 kg ha⁻¹ rate (Table 6.4). Application rate also influenced the quantity of simazine found in suspended and dissolved simazine runoff. However, it affected the quantity of suspended simazine more than the quantity of dissolved simazine. Dissolved simazine was 65 % higher (from 2.34 to 3.86 mg) when applied at 10.8 kg ha⁻¹; whereas, suspended simazine was 187 % higher (from 2.08 to 5.97 mg). Cumulatively, total simazine runoff was increased by 123 % (from 4.41 to 9.85 mg) by the higher application rate. Our results agree with those of Reichenberger et al. (2007) who recommended reducing herbicide rate as an effective way to minimize simazine runoff.

Surface runoff of simazine following heavy rainfall is likely to play a more important role in offsite movement than leaching. After 60 mm of simulated rainfall less than 1 mg

of simazine was detected in leachate from un-mulched columns treated with 5.4 kg ha⁻¹ (wk 1; Figure 6.1). In contrast, 6.3 mg of simazine were detected in runoff water and sediments following similar simulated rainfall (Table 6.3). Others have reported corroborating data. Cogger et al. (1998) found that simazine was not prone to leaching since most applied simazine was restricted within the top 15-cm layer of soil under field conditions. In contrast, up to 6.5 % of applied simazine ran off in response to 2 simulated rainfalls under field conditions (Liu and O'Connell 2002).

The pattern of simazine runoff. Since runoff water contained a significant amount of soil sediment we analyzed two fractions: dissolved simazine and suspended simazine. Suspended simazine consists of formulated simazine particles plus soil absorbed herbicide (Figure 6.2). The greatest amount of total simazine runoff was in the suspended fraction and this occurred primarily on d 1 (Figure 6.2) coinciding with maximum soil erosion (Figure 6.3). Runoff dramatically dropped on d 2, gradually decreased during d 3 and 4, and stabilized on d 5 and 6 (Figure 6.2). Suspended and dissolved simazine runoff followed a pattern similar to total runoff. However, suspended simazine decreased more quickly than did dissolved simazine. Most simazine was in the suspended fraction on d 1. This fraction declined rapidly from 76 % of the total on d 1 to 14 % by d 4 (Figure 6.2).

Suspended simazine runoff due to the first simulated rainfall played a major role in total simazine runoff. Specifically, 5.88 mg of suspended simazine runoff occurred from the un-mulched treatments following the first simulated rainfall, which accounted for 57 % of the cumulative total simazine runoff (10.3 mg) from the un-mulched treatments

throughout the entire experiment (Table 6.2). We attribute this observation to most simazine remaining as small particles of formulated herbicide at the time of application. This becomes apparent when considering that simazine was applied at $5,230 \text{ mg L}^{-1}$ and the water solubility is only 5 mg L^{-1} . Like fine soil sediment, these small simazine particles are very prone to runoff during heavy rainfalls. In addition, eroded soil sediment that absorbed simazine was also a contributor to simazine in the suspended fraction. However, free simazine particles were likely to be a more important contributor to suspended simazine runoff than soil absorbed simazine. This is supported by the observation that soil erosion from bare soil did not change much in response to the first and second simulated rainfalls (Figure 6.3), but the suspended simazine runoff from bare soil decreased about 9 times from d 1 to d 2 (Table 6.2). Averaged across mulched and non-mulched treatments, the first simulated rainfall resulted in 4.28 mg of simazine runoff, or 60 % of the cumulative total simazine runoff of 7.13 mg (Figure 6.2). These results correspond well with those of Glenn and Angel (1987) and Gaynor et al. (1992) who found that maximal herbicide runoff in the field was observed in response to the first rainfall.

The simazine application rate did not affect the amount of simazine in soil when treated with straw mulch. However, when applied to bare soil, the lower application rate (5.4 kg ha^{-1}) significantly reduced the amount of simazine retained in soil by 25 % compared with the 10.8 kg ha⁻¹ rate (Table 6.3). Straw had an even greater influence on simazine retained in soil than did application rate. Straw significantly reduced the amount of simazine in soil by 47 % and 53 %, respectively, at the 5.4 and 10.8 kg ha⁻¹ rates

(Table 6.3). This could be attributed to the effective interception and slow release of simazine by straw. As a result, less simazine was detected in soil covered with straw treated with simazine at 10.8 kg ha⁻¹ than in bare soil treated with simazine at 5.4 kg ha⁻¹. Reduced simazine concentration in mulched soil arguably would lead to less weed control. However, physical weed control by straw is likely to offset the effect of a lower herbicide interception in the soil. Crutchfield et al. (1986) observed no increase or decrease in weed control in straw-mulched relative to non-mulched plots 4 months after herbicide application even though the concentration of herbicide in the soil was lower. Our results also suggest that mulching may give growers the option of applying higher rates of simazine without increasing environmental or crop injury concerns. Dao (1991) proposed that straw could be possibly used as an herbicide slow release carrier, reducing herbicide offsite movement but not necessarily affecting weed control. Moreover, Case and Mathers (2006) reported that herbicide treated mulches provided better weed control than herbicide or mulch alone. In the experiments reported here, straw retained over 50 % of the applied simazine after 6 simulated rainfalls in the runoff trial supporting the concept that it acts as a slow release carrier.

The effect of straw on soil erosion. Straw reduced soil erosion in the runoff experiment by over 95 % (Figure 6.3). Similar results reported by Maass et al. (1988) and Döring et al. (2005) also indicated that mulch could reduce soil erosion by more than 90 % under field conditions. Moreover, straw also significantly reduced the initial simazine concentration in soil by 6 to 9 times due to the interception and/or absorption (Crutchfield et al. 1986). Collectively, by reducing both the amount of soil erosion and the concentration of simazine in soil, straw mitigates soil absorbed simazine runoff, thereby reducing simazine offsite movement (Table 6.3).

Soil erosion from straw mulched treatments was practically zero throughout this trial. However, it declined as time progressed from un-mulched treatments (Figure 6.3). It is widely accepted that newly disturbed soil is more vulnerable to erosion during heavy rainfalls (Langdale et al. 1979; Bradford and Huang 1994). Soil erosion is an important factor that limits the sustainability of vinifera grape production in cold climates because growers have to conduct tillage at least twice a year to build the soil hills needed for winter protection and to removed hilled soil for prevention of scion rooting.

In these experiments straw mulch retained about 40 % and 57 % of applied simazine at the end of leaching and runoff trials, respectively, and significantly affected the pattern of herbicide leaching and runoff. Respectively, straw reduced simazine leaching and runoff by 41 % and 62 % compared to the bare soil. The interception of simazine by straw and the slow release of simazine from straw were two important factors that contributed to these observations. The effect of straw in reducing soil erosion as well as simazine runoff associated with soil erosion was an additional factor. Straw reduced simazine concentration in soil by about 50 % at the end of both leaching and runoff trials. These results suggested that wheat straw would be a useful tool for management of simazine offsite movement and soil conservation in vinifera vineyards and other settings. The potential for straw as a mitigation tool for the offsite movement of other pesticides still remains to be explored.

136

Source of Materials

¹ Princep® 4L, Syngenta Crop Protection, P.O. Box 18300, Greensboro, NC 27419.

² Varian CP-3800, Varian Inc., 3120 Hansen Way, Palo Alto, CA 94304.

³ Ultra Scientific, 250 Smith St North Kingstown, RI 02852.

⁴ SAS 9.1, SAS Institute Inc. 100 SAS Campus Drive, Cary, NC 27513.

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Figure 6.1. The effect of straw mulch and simazine application rate on weekly simazine leaching (a) and cumulative simazine leaching (b). Three 20 mm simulated rainfalls were applied each wk and leachate from each was combined together to represent the entire wk. Bars marked by the same letter are not significantly different from each other within the same wk at the 0.05 level.



Figure 6.2. Daily suspended, dissolved and total simazine runoff, averaged across straw mulched and bare soil treatments and herbicide rate, following a simulated 10 mm rainfall. Dissolved simazine was that recovered from supernatant after centrifuging runoff water at 5000 rpm for 5 min. Suspended simazine was defined as the difference between simazine recovered from runoff water and from the supernatant after centrifuging. Bars indicate the standard deviation of total simazine runoff.



Figure 6.3. The effect of straw mulch on daily soil erosion in response to a simulated 10 mm rainfall. Standard deviation is indicated by bars.

Mulah	Data	Simazine	Simazine recovered				
Mulch	Kale	applied	Water	Soil	Straw		
	$-kg ha^{-1}-$ -		mg				
Soil	5.4	26.5	6.3 a [*]	5.1 bc	-		
Straw	5.4	26.5	2.6 b	3.9 c	5.2 b		
Soil	10.8	53.0	6.2 a	15.1 a	-		
Straw	10.8	53.0	3.8 b	5.7 b	26.1 a		

Table 6.1. The effect of straw mulch and simazine application rate on simazine recovered from leached water, soil and straw after 12 simulated, 20 mm, rainfalls.

* Means followed by the same letter are not significantly different from each other at

P<0.05 level.

Mulch	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Cumulative
				– mg			
		— Total simazine in runoff water —					
Soil	$7.24 a^{*}$	1.30 a	0.87 a	0.33 a	0.33 b	0.24 b	10.31a
Straw	1.33 b	0.55 b	0.66 a	0.36 a	0.50 a	0.56 a	3.96 b
	Dissolved simazine						
Soil	1.35 a	0.65 a	0.55 a	0.29 a	0.32 b	0.23 b	3.39 a
Straw	0.71 b	0.44 b	0.45 a	0.31 a	0.44 a	0.48 a	2.83 b
	Suspended simazine						
Soil	5.88 a	0.65 a	0.32 a	0.05 a	0.01 a	0.01 b	6.92 a
Straw	0.62 b	0.11 b	0.21 a	0.05 a	0.06 a	0.08 a	1.13 b

Table 6.2. The effect of straw mulch on daily simazine runoff in response to a simulated 10 mm rainfall.

* Means followed by the same letter are not significantly different from each other within

the same column and simazine type.

Mulah	Rate	Simazine	Sim	Simazine recovered			
Mulch		applied	Runoff Water	Soil	Straw		
	$-kg ha^{-1}-$ -		mg				
Soil	5.4	26.5	6.3 b [*]	14.4 b	-		
Straw	5.4	26.5	2.5 c	7.7 c	13.3 b		
Soil	10.8	53.0	14.3 a	19.3 a	-		
Straw	10.8	53.0	5.4 b	9.0 c	32.1 a		

Table 6.3. The effect of straw mulch on simazine recovered from runoff water, soil and straw after 6 simulated, 10 mm, rainfalls.

* Means followed by the same letter are not significantly different from each other at

P<0.05 level.

Simazine	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Cumulative	
-kg ha⁻¹-	mg							
	Total simazine in runoff water							
5.4	2.67 b^*	0.58 b	0.39 b	0.24 b	0.28 b	0.25 b	4.41 b	
10.8	5.89 a	1.26 a	1.14 a	0.46 a	0.55 a	0.55 a	9.85 a	
	Dissolved simazine							
5.4	0.88 b	0.39 b	0.33 b	0.22 b	0.27 b	0.25 b	2.34 b	
10.8	1.19 a	0.69 a	0.67 a	0.38 a	0.48 a	0.45 a	3.86 a	
	Suspended simazine							
5.4	1.80 b	0.19 b	0.06 b	0.02 b	0.01 b	0 b	2.08 b	
10.8	4.70 a	0.57 a	0.46 a	0.08 a	0.07 a	0.09 a	5.97 a	

Table 6.4. The effect of simazine application rate on daily simazine runoff in response to a simulated 10 mm rainfall.

* Means followed by the same letter are not significantly different from each other within

the same column and simazine type.

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