Colorado State University

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Can we manage replant disorders without chemicals?

Our aim is to help grower's to manage their problems, especially the important ones such as cold damage, Cytospora canker, replant diseases, yellow peach syndrome, cherry rasp leaf virus and so on. These problems do not have an easy chemical solution, such as in many other problems. We have several ongoing research projects to address these grower's issues and have been sharing our research results with the growers through Western Colorado Horticultural Society conference as well as Annual Reports, Workshops, and Western Phytoworks.

Replanting of pome and stone fruit trees on a site where the same crop was grown previously may cause poor growth and reduction in productivity of trees often resulting in tree death called "Replant disease or disorder". Studies on "replant disorder" of fruit crops have found that a combination of several factors such as nutrient deficiencies and toxicities, soil pH, soil conditions, poor plant soil water relationships, and the biotic causes such as root and vascular pathogens, parasitic nematodes, and often their combinations contribute to poor tree growth. The biological component (mostly diseases and nematodes) is often, but not always, the primary cause of this complex phenomenon. However, in many cases no specific pathogen could be found associated with the poor growth of the replanted perennial fruit trees. It may be possible that microbes and the roots of the previous fruit trees are releasing products into the soil that are phytotoxic to root growth and development of young replanted trees of the same species. One theory is that replant disease is due to a whole menagerie of tree pathogens (biotic cause) such as fungi (*Phytophthora* species, *Verticillium* spp. *Fusarium*), bacteria, nematodes such as Root-knot (*Meloidogyne)*, Root lesion (*Pratylenchus)*, and ring (*Creconemoides*), viruses and other organisms. However, the biotic cause/s associated with such replant problem vary/ies with locations, soil types, previous crop rootstock planted and so many other factors.

Removing as much of the old roots of previous crop from sites that are to be replanted is important. In Colorado, often growers use soil fumigation to control plant parasitic nematode to cope with this problem irrespective of the presence or absence of nematode or their population levels. Our extensive surveys of tree fruit orchards in Colorado found that root lesion nematode is predominantly present in most all tree fruit growing areas in Colorado irrespective of a difference in crop production system including rootstocks grown. Root-knot and ring nematodes are present in many orchards with variable numbers. However, their population levels in many areas are not to a damaging level requiring nematicide fumigation.

Thus, application of nematicides is not justified unless we assess nematode populations in soil and populations of certain species are beyond economic threshold level. Further information on plant parasitic nematodes, especially for Colorado can be found on our website: http://www.colostate.edu/programs/wcrc/pubs/research_outreach/Nematode%20Problems%20_final%2012-20.pdf. The work on finding out the other causes in replant disease is ongoing at Western Colorado Research Center.

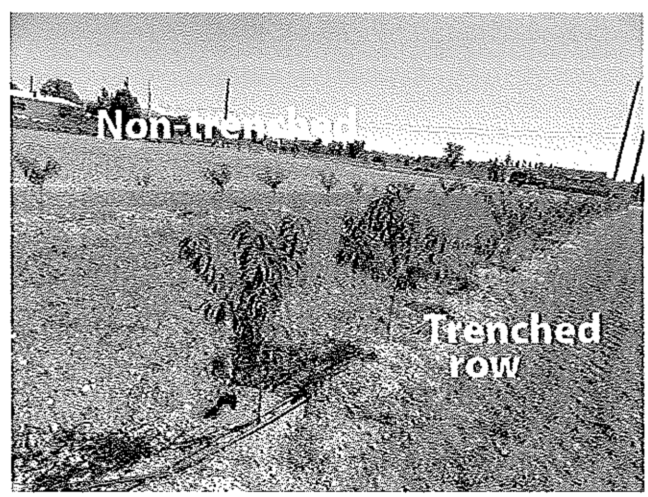


Figure 1

(Continued Page 4)

What is Second Generation Biofuel?

As the first-generation biofuel, ethanol is based on sugar and starch sources from food crops (i.e. corn and other grain crops). Ethanol production based on sugar and starch sources is not sustainable, and other feedstock sources are needed. Furthermore, ethanol, as a biofuel, has deficiencies. Ethanol has a lower energy content than gasoline, separates from gasoline in the presence of water, cannot be shipped through pipelines because it can become contaminated with water, and is corrosive to pipelines. Furthermore, ethanol contains dissolved impurities some of which are carcinogenic such as benzene, toluene, and xylene. Corn-based ethanol is often not an economically viable product, particularly in arid climates where water resources are scarce.

Biobutanol (n-butanol) is a preferred choice as a second-generation biofuel. Biobutanol, as a transportation fuel, has unique properties and is receiving significant attention from the research community. Unlike ethanol, butanol-gasoline blends do not have fuel blending restrictions. Because gasoline engines do not require modification to burn butanol this second generation biofuel is referred to as a "drop-in" fuel. The energy content of butanol is 86% compared to gasoline and butanol has a 30% higher energy content than ethanol.

A feedstock source of second-generation biofuels is lignocellulose. This biomass source can be processed by enzymatic conversion and fermentation. Rhizomatous, perennial grasses are a preferred feedstock because they have a neutral or negative carbon budget, require relatively few economic inputs, can be readily managed to be environmentally friendly, and can be grown on marginal, reclaimed, and underutilized land. The main technological challenges for lignocellulosic biofuel production are improving enzymatic treatments, fermentation, and process integration and efficiencies.

Biobutanol has superior fuel properties to ethanol because biobutanol:

- Has a higher energy content than ethanol.
- Can be readily blended with petroleum gasoline at higher concentrations than ethanol.
- Can be blended with gasoline at any concentration and at the refinery.
- Engines do not require modification for using biobutanol.
- Less susceptible to separation when mixed with water than ethanol.
- Less corrosive than ethanol.
- Reduces engine emissions (carbon monoxide and nitrous oxides).
- Suited for transport in pipelines.
- Low vapor pressure.
- Low solubility in water.
- Existing ethanol production facilities can be cost-effectively retro-fitted for production of biobutanol.

A benefit of biobutanol to agriculture is:

- Provides farmers with another market for agricultural products and by-products.

Biomass plots at the Colorado State University, Western Colorado Research Center at Fruita 2011. Photo by Calvin Pearson.

How will we make biobutanol?

A pilot-scale facility at Colorado Mountain College at the west campus in Rifle is under construction and nearing completion (see photo below). The biorefinery is capable of optimally producing 40 gallons of butanol per week from a range of lignocellulosic feedstocks. Briefly, the process stream involves grinding biomass in a chipper, dilute sulfuric acid pre-treatment, and transfer to a 430 gallon vessel for neutralization and enzymatic saccharification. The hydrolysate is passed through a filter press, in which solids are separated from the liquid phase. The liquid is heat-sterilized, deoxygenated, and introduced into a fermentation reactor. Anaerobic, fed-batch fermentation is conducted in a 400-gallon reactor with *Clostridium beijerinckii*. Product is removed intermittently by on-line gas stripping; H_2 and CO_2 are re-circulated through the fermentation broth while solvents (*n*-butanol, acetone, ethanol) are condensed and further purified by passage through a double-distillation unit in conjunction with a decanter. Probes for pH, dissolved oxygen, redox potential, and temperature provide real-time measurements in the fermentation, saccharification, and hydrolysate storage reactors. Sugars are monitored by high-performance liquid chromatography and butanol and other solvents are monitored by gas chromatography-flame ionization detection.

(Continued Page 3)

BioFuel (cont.)

The City of Rifle received a Rural Business Enterprise Grant in fall 2007 to conduct a feedstock study to gather information on the availability, costs, and feasibility of bioenergy production at the city-owned Energy Innovation Center site. In December 2008, City of Rifle officials contacted Dr. Calvin Pearson to discuss the potential for biomass and biofuel research and production in western Colorado.

In February 2009, phases of the feedstock study were completed and in spring 2009 Professor Jon Prater of Colorado Mountain College began construction of pilot biofuel production facility. Prater and City of Rifle officials discussed options for biofuel production and processing. In fall 2009, personnel from the City of Rifle, Colorado Mountain College, Colorado State University, and Flux Farm Foundation met and formed the Western Colorado Carbon Neutral Bioenergy Consortium (WCCNBC). Members of the consortium meet regularly.

In December 2009, Flux Farm Foundation was awarded a seed grant from the Colorado Department of Agriculture's Advancing Colorado's Renewable Energy (ACRE) program along with $25,000 in private support to conduct WCCNBC's first field study – "Evaluation of Perennial Plant Species and Production Inputs for Sustainable Biomass and Bioenergy Production in Western Colorado." Since that time others entities and companies have offered their non-financial support and encouragement to proceed with biomass and biofuels research and development in western Colorado. More information about WCCNBC is available at www.wccnbc.org.

On July 1, 2011, Drs. Calvin Pearson and Catherine Keske (CSU Assistant Professor/Agricultural Economist) were awarded a two-year competitive grant from the Sun Grant Initiative for $75,000 to:

1) evaluate grasses and legume species as single and mixed specie plantings and production input levels to identify those that are productive and profitable for biomass in this region,

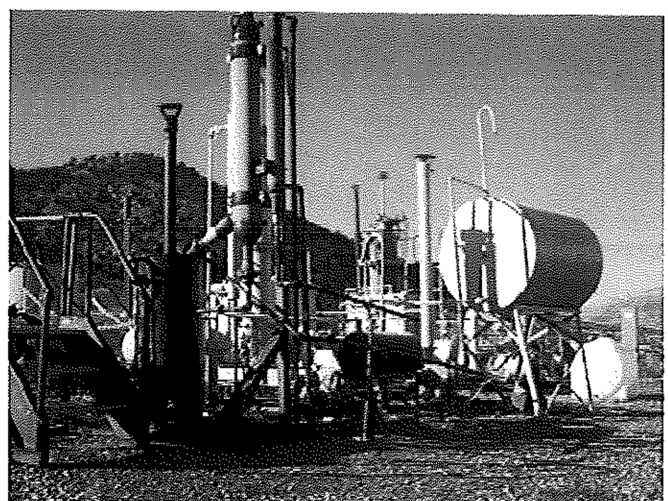
2) Assess carbon and nutrient cycling, carbon budgeting, and carbon sequestration in a biomass cropping system,

3) Perform economic, energy, and life-cycle analyses of the treatment variables to identify a sustainable biomass production system for the region,

4) Conduct outreach, public education, and technology transfer regarding biomass crop production along with associated technology (processing and conversion). Several other scientists and collaborators around the country are involved in this project and others aspects in our overall R&D efforts on biomass to biofuel in western Colorado.

A biomass-to-biofuel enterprise in our region and the West would create hundreds of new businesses and thousands of new jobs. For example, a reasonable goal is 3 dry tons/acre and a biofuel yield of 80 gal/ton of biomass, which would produce 240 gal/acre of biofuel.

Compared to much of the rest of the country, the Mountain States have (a) a large acreage of idle cropland, (b) a majority of this land in grassland, pasture, and range, and (c) one of the highest rates of crop failure in the country. Using sustainable cropping practices for biomass and planting, well-adapted biomass crops would reduce the incidence of crop failure. If 10 million acres of the 350 million acres of cropland, grassland, pasture, and range could be used for biomass, this land has the potential to produce 2.4 billion gallons of biofuel annually. This quantity of biobutanol produced in the western United States would make a significant contribution towards meeting the Congressional production goal of 16 billion gallons of biofuel per year by 2022.



The butanol biorefinery at Colorado Mountain College at Rifle.

For more information about this article contact Dr. Calvin Pearson at calvin.pearson@colostate.edu.

Replant Disorder (cont)

Ways to manage this problem are important for growers rather than knowing the cause. However, finding out the cause is important to effectively manage this problem. There is no silver bullet, like Methyl Bromide, which was available to manage this problem and is no longer available for use. We reported in 2008 that if the fumigation is done with optimal site preparation and application conditions, MIDAS 98 (1.0 or 0.5 lb/tree) could be as effective as chloropicrin (1 lb/tree) to manage the problem. Higher dose of MI (1.0 lb/tree) might be toxic to sweet cherry but not to peaches http://www.colostate.edu/programs/wcrc/pubs/publications/annrpt/cropyear06/tr07_08.pdf. With the increase in requirements for soil fumigation, application condition and soil preparation use for these chemicals use, use of these chemicals is problematic. Efforts to understand and manage this problem, especially finding non-fumigated strategies of replant management are underway at WCRC. Results of first year growth of four different types of peach rootstocks (Lovell, Nemagauard, St. Julian , and Viking) with different treatments indicated that we may be able to manage this problem without chemicals. The trees planted in a trenched and refilled replant site showed up to two fold increase in their first year's growth, as compared to trees planted in a non-trenched replant site (Fig 1), with minimal tree mortality, irrespective of compost application and rootstocks.

Trees planted in non-replant site had up to 4 fold increase without tree mortality irrespective of the rootstocks and varieties used as compared to replant site where the trees were planted almost 3 weeks after the replant site (Figure 2). The trees kept in a shade with enough water, with 3-4" root and shoot growth, were planted in non-replanted site as opposed to trees coming out from the cooler without new root and shoot growth planted in replant site. These studies need continuation to find a sustainable solution to the problem. These studies funded by EPA will be continued in future before we come up with some recommendations.

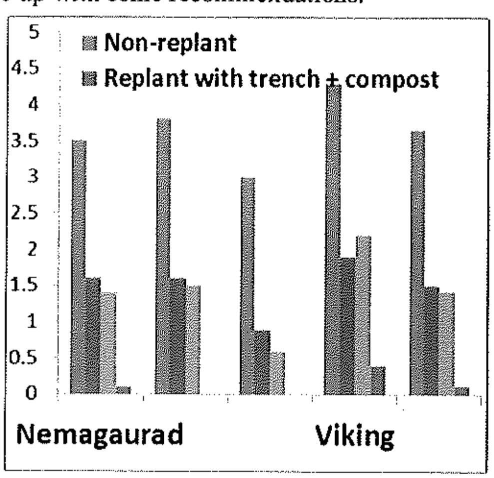


Figure 2

For further information on this issue contact rameshp@colostate.edu.

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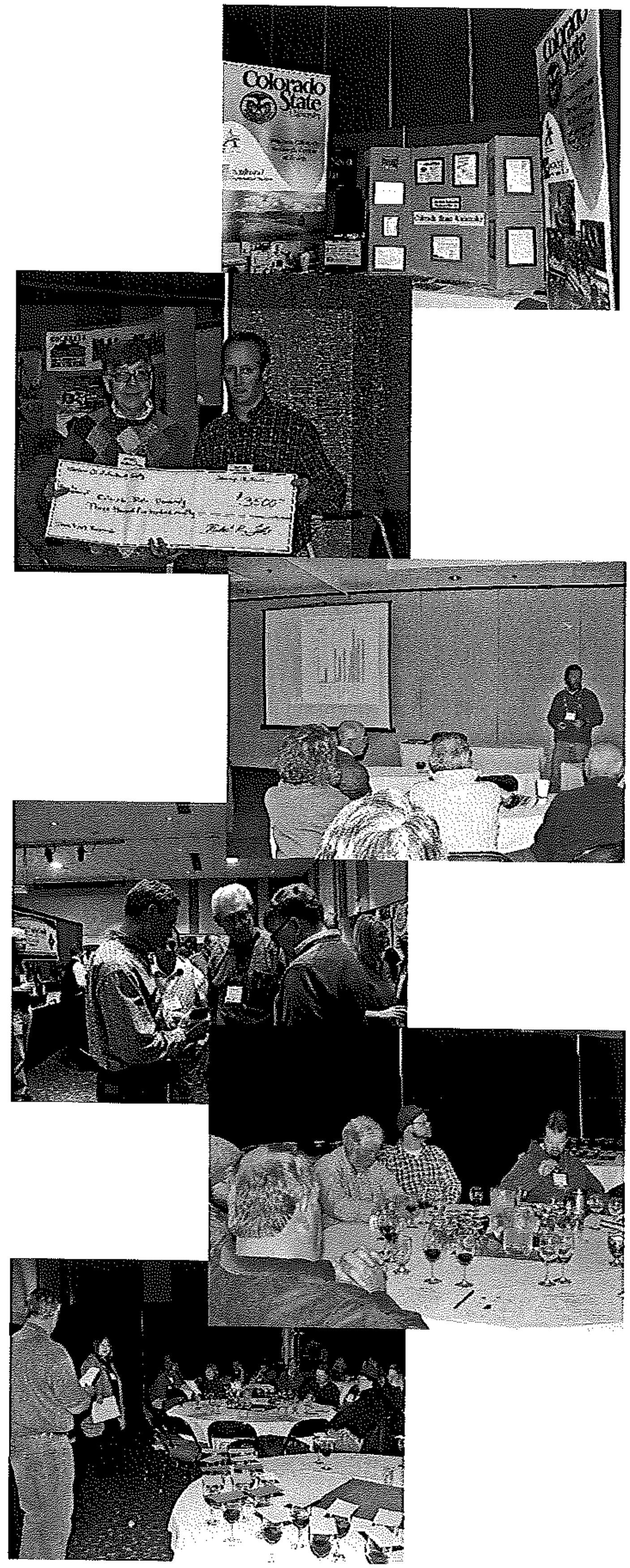


Nitrate Poisoning of Livestock

This past fall I've had inquiries from local clientele regarding nitrate poisoning of livestock and testing crops and crop residue for nitrates. Because of these inquiries I have prepared this article to provide people with some basic information about this topic.

Nitrate poisoning is a condition that affects ruminant animals (e.g., cattle, sheep, goats) when forages or water are consumed that contain an excessive amount of nitrate. High levels of nitrate in livestock can cause a disease called "nitrate poisoning." Although the term "nitrate poisoning" is often used, the toxicity is actually due to "nitrite."

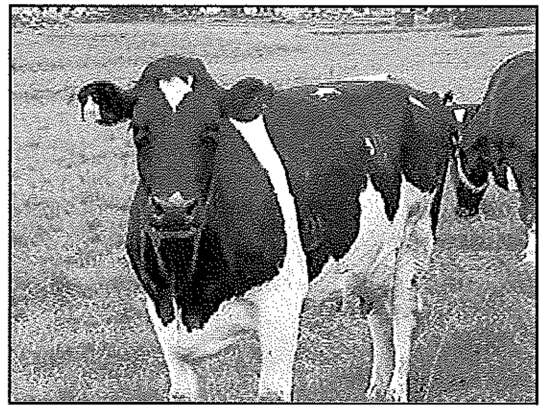
When animals ingest nitrate it is converted to nitrite, nitrite is converted to ammonia, and ammonia is converted to amino acids and then to protein. Nitrate is converted to nitrite faster than nitrite is converted to ammonia. Thus, when higher than normal amounts of nitrate are consumed, nitrite may accumulate in the rumen. The high amount of nitrite in the animal affects metabolic pathways and ultimately causes an inability to transport oxygen. Subsequently, the blood and tissues of the animal take on a blue to chocolate brown color and the animal is likely to suffocate and die.

Under normal conditions, nitrate is taken up by the plant in sufficient amounts for adequate plant growth and development and is utilized by the plant without nitrate accumulating within the plant. If nitrate is taken up faster than it can be utilized by the plant nitrate will accumulate in plant tissues. Excessive fertilization from commercial fertilizers or from manure can be a contributing factor for high rates of nitrate uptake by plants. Water sources can also be contaminated by excessive amounts of nitrate below or near manure piles, feedlots, cesspools, heavily manured or commercially fertilized fields, and others. Animals that drink from water contaminated with high levels of nitrate may also experience nitrate poisoning.

Environmental conditions can affect plant accumulation of nitrate including drought, sunlight, high temperatures, cool temperatures, frost, nutrient imbalances in the plant, and others (hail that reduces leaf area of the plant). Many growers recognize that when growth is arrested by a frost, nitrate can accumulate in parts of the plant. Thus, nitrate poisoning of livestock is often a concern in the fall, but nitrate poisoning can occur at other times of the year. Under adverse conditions such as drought, reduced sunlight, and unfavorable temperatures, roots can continue to absorb nitrate and it can accumulate in the lower portions of the plant, mainly stems and stalks when conditions do not allow the upper portions of the plant to utilize it. Seeds and flowers are typically not responsible for nitrate poisoning.

Some plant species including both crops and weeds are more likely to accumulate excessive amounts of nitrate than others. Examples of crops that are capable of accumulating high levels of nitrate are corn, small grains, sudangrass, sorghum, and sugarbeets. Examples of weeds that can accumulate high levels of nitrate are pigweed, lambsquarter, wild sunflower, field bindweed, nightshade, kochia, Canada thistle, and Johnsongrass.

Numerous factors can affect how an animal responds to ingesting feed or water containing high levels of nitrate. The factors include the age, health, species, feeding habits and history, animal hunger status, feed ration and quality, to name a few. Animals under stress are more susceptible to nitrate poisoning than healthy animals. Nitrate poisoning is also related



to the amount of total forage consumed, how quickly the forage is eaten, and how much nitrate is contained in the forage.

We do not perform plant tissue or water testing for nitrate at the Western Colorado Research Center. According to our associates at the CSU Veterinary Diagnostic Laboratory in Grand Junction, nitrate poisoning is a rare problem in western Colorado. Nevertheless, if you suspect a nitrate problem you should have your feed or water tested. A representative sample can be submitted to the CSU Veterinary Diagnostic Laboratory located in Grand Junction at 425 29 Road, phone 970-243-0673. Call the Veterinary Diagnostic Lab for details on sample size and sampling procedures. The sample will be logged at the Veterinary Diagnostic Lab at Grand Junction and then forwarded on to Fort Collins for analysis. The cost of nitrate analysis for feed and water at CSU is $10.00 per sample. Another alternative is to send samples to a reputable, certified laboratory. A list of certified laboratories in the U.S. is available at www.foragetesting.org.

For more information about this article see the additional reading below or contact Dr. Calvin Pearson at calvin.pearson@colostate.edu.

References and additional reading:

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Colorado Quality Wine and Grape Industry Program

A main goal of the Enology and Viticulture researchers at WCRC is to develop methodology that can help maintain and improve the quality of both the grapes used in the Colorado wine industry and the wines made from these grapes. To this end, a Colorado Quality Wine and Grape Industry Program has been created, under the guidance of the Colorado Wine Industry Development Board (CWIDB) and Associate Professor of Enology Stephen Menke, Department of Horticulture and Landscape Architecture (HLA). Dr. Menke has drawn up a 20 year multi-stage Vision Plan for quality improvement, including the following statements.

"Statement of Purpose: Colorado Quality Wine and Grape Industry Program

The Colorado wine and grape industry, in order to improve its quality, consumer approval, and sustainability, will institute a Colorado Quality Wine Industry Program, the purpose being to produce wines and grapes that meet high quality standards, to benefit the consumer, the industry, and the State of Colorado.

Mission Statement: Colorado Quality Wine and Grape Industry Program

A high standard of wine and grape quality, through a rigorous Wine and Grape Quality Testing Program, will be a continuing mission of the Colorado wine and grape industry. To attain a public and market image of quality wines, from quality wineries and vineyards, the Colorado wine and grape industry must institute testable standards, that foster a culture of producing fault-free, commercially acceptable wines and grapes, and drive the industry to produce world class wines and grapes. Education and training of the industry and the public will be consistent with a wine and grape culture that is driven by the pursuit of high standards of quality." [1]

The first step in this plan is to try to identify and describe characteristics of wines that do not meet commercially acceptable standards, by carrying out sensory quality assessments of commercial wines from the Colorado wine industry. To achieve this assessment, a pair of Colorado Wine Sensory Quality Assessment panels is assembled. The Wine Sensory Faults Panel consists of panelists trained to quantifiable aroma faults standards. The Wine Sensory Hedonistic Panel consists of nationally recognized wine competition judges. Each panel produces a score, the two scores are aggregated, and the aggregate score either exceeds, meets, or falls short of a passing score, which is set ahead of time by an advisory panel. Both panels also write comments on each wine assessed. The scores and the comments for each wine are sent confidentially to the submitting wineries. It is hoped that these scores and comments will provide essential information on wine characteristics, and that any further information or guidance on these wines can be provided by the Colorado State Enologist or other consulting experts.

The first joint Colorado/Nebraska Wine Sensory Quality Assessment was held in August of 2011, at the sensory laboratory of the Food Science Department at the University of Nebraska-Lincoln. The number of wines assessed was 148. Of these 58 were from 12 Colorado wineries and 81 were from 13 Nebraska wineries. As controls, 9 commercially purchased and nationally distributed wines in the same price range, none from Colorado or Nebraska were assessed. A perfect aggregate score is 32, and the passing score was set at 22.4. The overall rate of wines receiving a passing score was 73% and failing scores 23%. Of the Colorado wines, 91% passed and 9% failed. Of the control wines, 11% failed.

All results and comments for individual wines were sent to wineries submitting those wines. Initial informal feedback from the wineries is that the scores and comments are helpful to their winemaking operations, and should help in identifying sources of fault characteristics for wines that did not pass. In addition, comments on wines that passed, but did have some degree of flaws, are regarded as helpful by responding wineries. Lastly, comments describing positive characteristics of individual wines are regarded by wineries as helpful in defining wine styles.

[1] "VISION PLAN: COLORADO QUALITY WINE AND GRAPE INDUSTRY PROGRAM, by Stephen Menke, CSU Enology Program, 2009"