## V(A). Planned Program (Summary)

**Program # 1**

**1. Name of the Planned Program**

Global Food Security and Hunger - Agricultural Production in a Semi-Arid Environment

☑ Reporting on this Program

**V(B). Program Knowledge Area(s)**

1. Program Knowledge Areas and Percentage

| KA Code | Knowledge Area | %1862 Extension | %1890 Extension | %1862 Research | %1890 Research |
|---|---|---|---|---|---|
| 102 | Soil, Plant, Water, Nutrient Relationships | | | 25% | |
| 203 | Plant Biological Efficiency and Abiotic Stresses Affecting Plants | | | 38% | |
| 206 | Basic Plant Biology | | | 14% | |
| 502 | New and Improved Food Products | | | 5% | |
| 504 | Home and Commercial Food Service | | | 2% | |
| 511 | New and Improved Non-Food Products and Processes | | | 13% | |
| 701 | Nutrient Composition of Food | | | 3% | |
| | **Total** | | | 100% | |

## V(C). Planned Program (Inputs)

**1. Actual amount of FTE/SYs expended this Program**

| Year: 2014 | Extension | | Research | |
|---|---|---|---|---|
| | 1862 | 1890 | 1862 | 1890 |
| **Plan** | 0.0 | 0.0 | 1.5 | 0.0 |
| **Actual Paid** | 0.0 | 0.0 | 2.2 | 0.0 |
| **Actual Volunteer** | 0.0 | 0.0 | 0.0 | 0.0 |

**2. Actual dollars expended in this Program (includes Carryover Funds from previous years)**

| Extension | | Research | |
|---|---|---|---|
| **Smith-Lever 3b & 3c** | **1890 Extension** | **Hatch** | **Evans-Allen** |
| 0 | 0 | 264049 | 0 |
| **1862 Matching** | **1890 Matching** | **1862 Matching** | **1890 Matching** |
| 0 | 0 | 247981 | 0 |
| **1862 All Other** | **1890 All Other** | **1862 All Other** | **1890 All Other** |
| 0 | 0 | 0 | 0 |

## V(D). Planned Program (Activity)

### 1. Brief description of the Activity

Engineering Plants for Increased Nutrition and Improved Stress Tolerance
This year significant progress has been made in engineering thiamine production in the model plant Arabidopsis. The team engineered Arabidopsis seed with >8 fold increase in seed thiamine continent. Improved seed mass of thiamine enriched plants by 25% over wild types, increased seed oil continent by 10%, increased carbohydrates by 30%, and proved to be more tolerant to paraquate and salt induced oxidative stress. The team has now moved on to engineering the thiamine biosynthetic pathway into Camelina seeds.

Improving the Seed Set Yield of Plants under Heat, Cold and Drought Stress
Scientists at UNR have made significant progress in three areas of stress. The team tested whether stress tolerance can be improved by expressing two different transcription factors that regulate stress responses in vegetative tissues, but are not expressed in pollen. Secondly, they engineered CNGC16 (a cyclic nucleotide-gated channel, critical in stress tolerance of pollen) with mutations in its regulatory domain. Transgenic plants were made and tested for changes in pollen stress tolerance. And finally, transcriptome profiling was used to identify temperature-dependent changes in gene expression that occur in pollen. The team then performed RNA-sequencing analyses for pollen grains harvested from wild type Arabidopsis plants (and CNGC16 mutants), and two tomato cultivars (one being heat tolerant) under control and heat stress conditions.

Enhancing Cell Wall Digestibility To Increase The Productivity Of Biomass And Forage Feedstocks.
A new line of research was started this year with the addition of Dr. Ian Wallace. Wallace's lab has been busy gathering Opuntia (prickly pear cactus) pads for characterization of cell wall sugar composition and enzymatic deconstruction of lignocellulosic biomass using a fungus model. The team has genotyped Arabidopsis REF8 knockout lines to ensure that they are true mutants, and are currently growing mutants for preliminary phenotypic characterization as well as seed bulking for EMS mutagenesis. Additionally, the team has determined that alfalfa is sensitive to isoxaben.

Evaluation of Wine Grape Cultivars and Clones
Screening for genotype variation involved in water use efficiency were conducted on 13 varieties of wine grapes. From this massive undertaking one region was of the genome was discovered (NCED1) and sequenced. A rapid dehydration assay was developed that provides a simple and reliable way of phenotyping plant responses to water loss. Stomatal responses were then tested among known drought-tolerant varieties and compared to field trials.

Research was conducted to determine transcriptomics responses (the complete set of RNA transcripts that are produced by the genome, under specific circumstances) to dehydration of mature leaves of three varying drought tolerant genotypes.

The plant hormone abscisic acid was investigated to determine how it regulates various developmental process and stress responses over short time frames (i.e. hours). In order to understand the transcriptional regulatory, different organs of grape (berries, shoot tips, leaves, roots and cell cultures) were studied. Whole genome microarrays for genes of grape vines were used to determine the effects of ABA on organ-specific mRNA expression patterns. The biochemical pathways affected by ABA were analyzed using the Cytoscape program with the BiNGO plug-in software.

Hydroponics impact on Content of Bioactive Compounds
Work was conducted to determine whether culinary greens can be successfully grown using continuous flow systems (hydroponics) with varying levels of nutrients. Scientists examined the difference in yield at high, medium, and low levels of commercial hydroponic nutrient solutions in order to optimize the cost-benefit relationship in the growth of leafy greens. The team also determined the feasibility of an "organic" self-prepared nutrient solution for leafy green growth.

Optimizing High Desert Hoophouses
During the first year of this project, four hoophouse were construction and instrumentation. Though originally planned for year two, two warm season crops were planted. Production potential and efficiency were determined using soil water saturation when grown in an average humidity of 49.8%.

**2. Brief description of the target audience**

The scientific community, agriculture producers, local organizations, backyard hobbyists, as well as students taking classes or participating in research activities.

**3. How was eXtension used?**

eXtension was not used in this program

**V(E). Planned Program (Outputs)**

**1. Standard output measures**

| 2014 | Direct Contacts Adults | Indirect Contacts Adults | Direct Contacts Youth | Indirect Contacts Youth |
|---|---|---|---|---|
| **Actual** | 765 | 0 | 200 | 0 |

**2. Number of Patent Applications Submitted (Standard Research Output)**
**Patent Applications Submitted**

| | |
|---|---|
| Year: | 2014 |
| Actual: | 0 |

**Patents listed**

**3. Publications (Standard General Output Measure)**

**Number of Peer Reviewed Publications**

| 2014 | Extension | Research | Total |
|---|---|---|---|
| Actual | 0 | 7 | 7 |

**V(F). State Defined Outputs**

**Output Target**

**Output #1**

**Output Measure**

- Peer reviewed journal articles, chapters, or books

| Year | Actual |
|---|---|
| 2014 | 9 |

**Output #2**

**Output Measure**

- Non-peer reviewed publications

| Year | Actual |
|---|---|
| 2014 | 2 |

**Output #3**

**Output Measure**

- Presentations

| Year | Actual |
|---|---|
| 2014 | 44 |

**Output #4**

**Output Measure**

- Demonstrations and workshops conducted

| Year | Actual |
|---|---|
| 2014 | 6 |

**Output #5**

**Output Measure**

- Leveraged research funds

| Year | Actual |
|---|---|
| 2014 | 652415 |

**Output #6**

**Output Measure**

- Web sites created or updated

| Year | Actual |
|---|---|
| 2014 | 1 |

**Output #7**

**Output Measure**

- Number of graduate students and post-doctorates trained

| Year | Actual |
|---|---|
| 2014 | 7 |

**Output #8**

**Output Measure**

- Number of undergraduate students involved in research program

| Year | Actual |
|---|---|
| 2014 | 30 |

**Output #9**

**Output Measure**

- Databases, Models, Protocols, and Research Materials

| Year | Actual |
|---|---|
| 2014 | 1 |

**Output #10**

**Output Measure**

- New Germplasm

| Year | Actual |
|---|---|
| 2014 | 1 |

## V(G). State Defined Outcomes

### V. State Defined Outcomes Table of Content

| O. No. | OUTCOME NAME |
|---|---|
| 1 | New fundamental or Applied knowledge |
| 2 | Improved skills of students, post-doctorate, and stakeholders |
| 3 | New or improved methods |
| 4 | Adopt and use new methods or improved technology |
| 5 | Greater productivity in food provisions |
| 6 | Increase economic competitiveness |
| 7 | Adopt new improved skills |
| 8 | Apply improved fundamental or applied knowledge |
| 9 | Increasing Seed Set Yields in Stressed Plants |
| 10 | Urban Hydroponic Fruit And Vegetable Production: Optimizing Nutrient Solutions |
| 11 | Engineering Plants For Increased Nutrition And Increased Stress Tolerance |

**Outcome #1**

1. **Outcome Measures**

New fundamental or Applied knowledge

Not Reporting on this Outcome Measure

**Outcome #2**

1. **Outcome Measures**

Improved skills of students, post-doctorate, and stakeholders

Not Reporting on this Outcome Measure

**Outcome #3**

1. **Outcome Measures**

New or improved methods

Not Reporting on this Outcome Measure

**Outcome #4**

1. **Outcome Measures**

Adopt and use new methods or improved technology

Not Reporting on this Outcome Measure

**Outcome #5**

1. **Outcome Measures**

Greater productivity in food provisions

Not Reporting on this Outcome Measure

**Outcome #6**

1. **Outcome Measures**

   Increase economic competitiveness

   Not Reporting on this Outcome Measure

**Outcome #7**

1. **Outcome Measures**

   Adopt new improved skills

   Not Reporting on this Outcome Measure

**Outcome #8**

1. **Outcome Measures**

   Apply improved fundamental or applied knowledge

   Not Reporting on this Outcome Measure

**Outcome #9**

1. **Outcome Measures**

   Increasing Seed Set Yields in Stressed Plants

2. **Associated Institution Types**

   - 1862 Research

**3a. Outcome Type:**

Change in Knowledge Outcome Measure

**3b. Quantitative Outcome**

| Year | Actual |
|---|---|
| 2014 | 0 |

**3c. Qualitative Outcome or Impact Statement**

**Issue (Who cares and Why)**

Each year crop yields are severely impacted by a variety of stress conditions, including heat, cold, drought, hypoxia, and salt. Reproductive development in flowering plants is highly sensitive to hot or cold temperatures, with even a single hot day or cold night sometimes being fatal to reproductive success. In many plants, pollen tube development and fertilization is the ?weakest link?. The focus of this research is on developing two strategies to improve stress tolerance in pollen production.

**What has been done**

In the past year, NAES scientists tested whether stress tolerance can be improved by expressing two different transcription factors that regulate stress responses in vegetative tissues, but are not expressed in pollen. Cyclic nucleotide-gated channels (CNGC16) plants were made (transgenics) and tested for changes in pollen stress tolerance. Finally, profiling sets of RNA molecules to identify temperature-dependent changes in gene expression that occur in pollen. The team performed RNA-sequencing analyses for pollen grains harvested from wild type Arabidopsis plants (and CNGC16 mutants), and two tomato cultivars (one being heat tolerant) under control and heat stress conditions.

**Results**

NAES scientists in collaboration with an Italian lab, produced work that provides biochemical evidence to establish the auto-inhibited Ca2+-ATPases (ACA12) as the first example of an ACA-type calcium pump that appears to be constitutively active.

The discovery that knocking out an enzyme involved in cell wall functioning (TBL14) has provided the first genetic evidence that links a specific calcium signaling pathway with the function of a specific protein involved in plant cell wall biogenesis.

NAES scientists obtained genetic evidence that the complete loss plasma membrane calcium pumps is not lethal to the model plant Arabidopsis, but does result in a phenotype in which the plant fails to bolt and produce reproductive structures.

Finally, the team uncovered genetic evidence that increased stress tolerance can be obtained by expression of a galactinol synthase gene. This research has provided preliminary evidence that will allow scientists to manipulate the complete set of small-molecule chemicals found within pollen to improve reproductive fitness under stress conditions.

## 4. Associated Knowledge Areas

| KA Code | Knowledge Area |
|---|---|
| 203 | Plant Biological Efficiency and Abiotic Stresses Affecting Plants |
| 206 | Basic Plant Biology |

## Outcome #10

### 1. Outcome Measures

Urban Hydroponic Fruit And Vegetable Production: Optimizing Nutrient Solutions

### 2. Associated Institution Types

- 1862 Research

### 3a. Outcome Type:

Change in Knowledge Outcome Measure

### 3b. Quantitative Outcome

| Year | Actual |
|---|---|
| 2014 | 0 |

### 3c. Qualitative Outcome or Impact Statement

**Issue (Who cares and Why)**
Greenhouse agriculture has the potential for improving yield and vegetable quality in sub-optimal environments. Despite increased frequency of greenhouse vegetable production, basic information on optimal nutrient growing solutions and other environment factors for production of commercial vegetables is lacking. Nutrient and fertilizer delivery is a high-cost component to production, especially in hydroponic systems which often use proprietary blends of chemical fertilizers. Organic fertilizers are gaining in popularity, but need to be standardized and tested in hydroponic systems before they are widely used.

**What has been done**
Hydroponic systems were manipulated to investigate the influence of aqueous media on vegetable production. In this research, the team tested the feasibility of using low levels of commercial (General Hydroponics Flora series) nutrients on green leafy vegetable production. In subsequent tests, the team created an organic nutrient solution and compared the yield to both the high and low level conventional nutrient treatments.

**Results**
NAES scientists determined that regardless of level of commercial hydroponic nutrient solutions - 1,200 (manufacture recommendation), 600, 400, and 300 ppm - leafy greens growth was similar. Producers could reduce their nutrient input cost by 25% and should still see normal growth in both bok choy and lettuce.

### 4. Associated Knowledge Areas

| KA Code | Knowledge Area |
|---|---|

| | |
|---|---|
| 102 | Soil, Plant, Water, Nutrient Relationships |
| 203 | Plant Biological Efficiency and Abiotic Stresses Affecting Plants |
| 206 | Basic Plant Biology |
| 701 | Nutrient Composition of Food |

**Outcome #11**

**1. Outcome Measures**

Engineering Plants For Increased Nutrition And Increased Stress Tolerance

**2. Associated Institution Types**

- 1862 Research

**3a. Outcome Type:**

Change in Knowledge Outcome Measure

**3b. Quantitative Outcome**

| Year | Actual |
|---|---|
| 2014 | 0 |

**3c. Qualitative Outcome or Impact Statement**

**Issue (Who cares and Why)**
Thiamin (Vitamin B1) is an essential plant derived nutrient in human and animal diets. While not limiting in typical Western diets, thiamin deficiencies are common in developing countries where people subsist on diets consisting primarily of polished grains and seed products. By increasing thiamin levels in the seeds of important subsistence crop species such as rice and wheat, we can make a positive impact on human nutrition and global food security.

**What has been done**
Using standard molecular biology techniques, the open reading frames corresponding to the Arabidopsis enzymes (HET-P synthase, HMP-PP synthase and TMPPase), complementary DNA sequences were sub-cloned behind the rapeseed's napin and oleosin promoters, and the soybean glycinin promoter, respectively. Mature seed from each line were analyzed for seed thiamin content using reverse phase HPLC. The team obtained a cDNA clone corresponding to the sesame seed thiamin binding protein and created constructs that were transferred into first Arabidopsis and then into Camelina. To assess stress tolerance in the transgenic plants with elevated seed thiamin, the team used the highly sensitive root growth assay developed for Arabidopsis in an earlier NAES HATCH project.

**Results**
This year significant progress has been made in engineering thiamine production in the model

plant Arabidopsis. The team engineered Arabidopsis seed with 8+ fold increase in seed thiamine continent. Improved seed mass of thiamine enriched plants by 25% over wild types, increased seed oil continent by 10%, increased carbohydrates by 30%, and proved to be more tolerant to paraquate and salt induced oxidative stress. Finally, the team has genetically engineered False Flax (Camelina sativa) with three thiamine biosynthetic genes cloned behind seed specific promoters.

**4. Associated Knowledge Areas**

| KA Code | Knowledge Area |
|---|---|
| 203 | Plant Biological Efficiency and Abiotic Stresses Affecting Plants |
| 206 | Basic Plant Biology |
| 502 | New and Improved Food Products |
| 701 | Nutrient Composition of Food |

## V(H). Planned Program (External Factors)

**External factors which affected outcomes**

- Natural Disasters (drought, weather extremes, etc.)
- Economy
- Appropriations changes
- Competing Programmatic Challenges

**Brief Explanation**

The Experiment Station is still recovering from the 31 hard money positions lost after the 2010 circulation review. Until a number of critical areas are re-staffed the agricultural program will be severely hampered. Nevada annual average precipitation continues to decline, with past winter 6% of average surpassing the 1970's record. Work on the protective properties of abscisic acid on grape's tolerance to cold was completely ineffective, so research will now look at dehydrational effects on cold tolerance. The optimizing hoop house project has been impeded due to graduate student departure from the program during last year.

## V(I). Planned Program (Evaluation Studies)

**Evaluation Results**

Significant progress was made in the development of improved thiamine production in the model plant Arabidopsis. The team engineered Arabidopsis seed with a greater than 8 fold increase in seed thiamine continent. Improved seed mass of thiamine enriched plants by 25% over wild types, increased seed oil continent by 10%, increased carbohydrates by 30%, and engineered plants proved to be more tolerant to the herbicide paraquate and salt induced oxidative stress.

A rapid dehydration assay was developed that provides a simple and reliable way of phenotyping wine grape vines responses to water loss. Our varietal trials continue to produce cultivars with higher drought tolerance and lower demands for water inputs.

Commercial hydroponic nutrient solutions were shown to yield similar vegetative growth regardless of dilution levels.

Our forage biochemists have determined that Medicago truncatula (clover-like legume) is sensitive to herbicide isoxaben used in vineyards and orchards.

**Key Items of Evaluation**

- Three journal articles were published on grapevine responses to dehydration.
- One journal article was published on dilution effects of commercial nutrient solutions on the hydroponically grown vegetables.
- A new germplasm was genetically engineered with three thiamine biosynthetic genes