V(A). Planned Program (Summary)

Program # 7

1. Name of the Planned Program

Sustainable Energy

V(B). Program Knowledge Area(s)

1. Program Knowledge Areas and Percentage

KA Code	Knowledge Area	%1862 Extension	%1890 Extension	%1862 Research	%1890 Research
204	Plant Product Quality and Utility (Preharvest)			7%	
206	Basic Plant Biology			69%	
511	New and Improved Non-Food Products and Processes			24%	
	Total			100%	

V(C). Planned Program (Inputs)

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

Year: 2009	Extension		Research	
	1862	1890	1862	1890
Actual	0.0	0.0	1.7	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	0	0
1862 Matching	**1890 Matching**	**1862 Matching**	**1890 Matching**
0	0	0	0
1862 All Other	**1890 All Other**	**1862 All Other**	**1890 All Other**
0	0	243567	0

V(D). Planned Program (Activity)

1. Brief description of the Activity

Scientists are currently determining best algal species (survival and growth rates, fat and starch content) for production of oils that will then be processed into biofuels. Investigation into Nevada's highly prolific rabbit brush as a good candidate for production of industrial (i.e., rubber, plastics, coatings, lubricants and adhesives) and energy feedstocks (i.e. biodiesel and cellulosic-based liquid fuels.

2. Brief description of the target audience

Those most effect by this research include the general public, energy and chemical industry, potential producers.

V(E). Planned Program (Outputs)

1. Standard output measures

2009	Direct Contacts Adults	Indirect Contacts Adults	Direct Contacts Youth	Indirect Contacts Youth
Plan	{NO DATA ENTERED}	{NO DATA ENTERED}	{NO DATA ENTERED}	{NO DATA ENTERED}
Actual	15	0	85	0

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

Year: 2009
Plan:
Actual: 0

Patents listed

3. Publications (Standard General Output Measure)

Number of Peer Reviewed Publications

2009	Extension	Research	Total
Plan			
Actual	0	9	9

V(F). State Defined Outputs

Output Target

Output #1

Output Measure

- Peer reviewed journal articles, publications in commodity group publications, presentations at scientific meetings, presentations at stakeholder, Native American and agency meetings.

Year	Target	Actual
2009	{No Data Entered}	37

Output #2

Output Measure

- Demonstrations and Field Days Conducted

Year	Target	Actual
2009	{No Data Entered}	4

Output #3

Output Measure

- Number of Graduate Students or Post-Doctorates Trained

Year	Target	Actual
2009	{No Data Entered}	3

Output #4

Output Measure

- Number of Undergraduate Students Involved in Research

Year	Target	Actual
2009	{No Data Entered}	0

Output #5

Output Measure

- Research Projects Conducted

Year	Target	Actual
2009	{No Data Entered}	1

V(G). State Defined Outcomes

V. State Defined Outcomes Table of Content

O. No.	OUTCOME NAME
1	Evaluating Algae's Potential as Feed Stock for Biofuel Production
2	Developing Better Plants for Biofuel Production
3	Rabbit Brush Potential for Domestic Rubber Production

Outcome #1

1. Outcome Measures

Evaluating Algae's Potential as Feed Stock for Biofuel Production

2. Associated Institution Types

- 1862 Research

3a. Outcome Type:

Change in Action Outcome Measure

3b. Quantitative Outcome

Year	Quantitative Target	Actual
2009	{No Data Entered}	3

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
Existing oilseed feed stocks derived from terrestrial plants (e.g. soybeans) cannot satisfy the current or projected transportation fuel needs of the U.S. Biofuel producing algae are one of the only avenues available for high-volume capture and reuse of CO2 generated in biomass-, coal- or natural gas-fired power plants and can provide the intermountain west with a major new "cash crop" without putting additional demands on freshwater supplies needed for residential, industrial and agricultural use. Nevada researchers and producers are uniquely enabled to leverage the geothermal, high solar radiation, ample land area, and salt basins to produce algae in a scalable and economically viable manner. The goal of this research is to explore alternative strains of algae for use as biofuel feed stocks and new nutrient mediums in which to grow algae.

What has been done
Building upon previous strains of salt-loving algae, analysis of growth rates, lipid content, and insoluble starch content has been completed for 11 strains of freshwater algae and 12 brackish water strains. In cooperation with Truckee Meadows Water Reclamation Facility (TMWRF) in Sparks, NV, large industrial centrifuges were used to remove wastewater sludge from Sparks' sewage. The water leaving a centrifuge after most of the solids have been removed (centrate) was then collected, added to algae growth solution. Research is in progress to identify the optimal centrate loading for algal use as well as water quality analyses to establish the product water quality following algae harvesting.

Algal stains have been tested and shown to be able to grow in up to 20% centrate while removing more than 50% of N and P content. Selection of algal strains that can tolerate higher concentrations of centrate is also in progress. Large-scale production trials of algae on centrate have been completed at the 2,000 L raceway ponds. Productivities are about 60% of those obtained by control fertilizer media. Kilogram quantities of algal paste have been produced for solvent extraction trials.

Genetic analysis of these strains is now in progress as are continued rounds of selection. Total genomic DNA and RNA from Dunaliella salina (a salt-loving, pink micro-algae) was submitted to the Joint Genome Institute for genomic sequencing and annotation. This information will be instrumental in improving the accuracy of genome annotation and mRNA expression analysis to test the effects of nutrient deprivation or other stress conditions on lipid production.

Results
The University of Nevada, Reno investigation team needing high concentrations of nitrogen, phosphorus and inorganic carbon, key nutrients required for algae growth, have setup an alliance with TMWRF to accept a secondary wastewater product (citrate). At present, TMWRF produces approximately 250,000 gallons of centrate per day, which is recycled to the head of the treatment facility. Utilizing this liquid stream for beneficial purposes as a nutrient source for algae growth would save the treatment facility about $4,400/day and $2,900/day for nitrogen and phosphorus removal, respectively, or $2.7 M annually while generating ~1,600 gallons of oil per day

assuming a 30% lipid content of the algae.

After 20 rounds of reiterative density gradient centrifugation, investigators have been able to increase lipid content by 39% and starch content by 227%. In addition, a room-size "demonstration" scale production facility is now operational to educate scientists, investors, and the lay public about the potential feasibility of the algae-to-biodiesel conversion process and related technologies.

4. Associated Knowledge Areas

KA Code	Knowledge Area
206	Basic Plant Biology
511	New and Improved Non-Food Products and Processes

Outcome #2

1. Outcome Measures

Developing Better Plants for Biofuel Production

2. Associated Institution Types

- 1862 Research

3a. Outcome Type:

Change in Knowledge Outcome Measure

3b. Quantitative Outcome

Year	Quantitative Target	Actual
2009	{No Data Entered}	1

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
The Department of Energy (DOE) has requested the scientific community to increase the energy density of nontraditional oil crops by understanding and manipulating the metabolic and genetic circuits that control accumulation of oils and other easily digestible, energy-rich compounds in plant tissues. In response, University of Nevada, Reno scientists are focused on how different cellular pumps can potentially be used to alter a plant cell's membrane structures and increase lipid content for use in biofuels. With one of the primary goals of the DOE's "Energy Mission" being to explore the potential of using lipid enzymes to trigger the formation of novel lipid storage organelles, this project is trying to understand the biological functions of different P-type ATPases (ion pumps or membrane pumps) in nontraditional crops.

What has been done
Using a genetic engineering technique call the "double knockout mutation" a vacuole calcium pump has been found to develop leaves in test plants with large numbers of necrotic lesions. This advancement is important for several reasons, including the potential to use these mutants as a screening/measuring technique for targeting information that directs calcium pumps to the vacuole. This will allow scientists to add or mutate potential targeting information to a cell's outer wall calcium pump and test whether it can be relocated to other areas within the plant.

In collaboration with the Palmgren group, the University of Nevada, Reno identified the first plant phenotype for a gene knockout of a lipid enzyme pump. Plants harboring a disruption of this gene grow more slowly and have reduced biomass. The team obtained evidence that the underlying cause of this defect is related to a defect in secretory activity of selected cell types.

Progress has also been made on determining the effects on membrane lipid profiles caused by gene disruptions in three different subfamilies of enzyme pumps; determining whether gene disruptions for supposed enzyme pump subunits will result in phenotypes that match earlier mutations; and, identifying changes in cellular functions caused by over-expression/activity of enzyme pumps.

Results
The success of the U.S. biofuel industry depends, to a large degree, on the quantity and quality of biomass available, and on the industry's ability to cost-effectively utilize biomass for energy production. Advances resulting from this project will provide the knowledge needed to develop new bio-based products, methods, and tools that the emerging biofuel industry can use.

Research efforts at the University of Nevada, Reno have opened the door to creating this genetically modified biofuel. If new invaginations (or membrane compartments) can be induced in lab plants, this represents a potential strategy to increase the lipid content of any photosynthetic cell, and thereby increase its value as a feedstock for biodiesel production.

4. Associated Knowledge Areas

KA Code	Knowledge Area
206	Basic Plant Biology

Outcome #3

1. Outcome Measures

Rabbit Brush Potential for Domestic Rubber Production

2. Associated Institution Types

- 1862 Research

3a. Outcome Type:

Change in Knowledge Outcome Measure

3b. Quantitative Outcome

Year	Quantitative Target	Actual
2009	{No Data Entered}	1

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
Global warming and decreasing oil reserves have mandated the development of renewable and sustainable alternatives to petroleum-based products. Plants, which produce zero balance carbon emission energy feedstocks, represent a particularly attractive alternative to fossil fuels. Furthermore, due the high degree of chemical diversity present in plant kingdom, many plant species are being investigated as replacement for the production of petroleum-based industrial chemicals.

One such plant species is Chrysothamnus nauseosus (rabbit brush). Rabbit brush is a highly prolific perennial shrub that is endemic to Nevada and the Great Basin region of the United States. This plant species produces significant amounts natural rubber, soft resins and plant based ethanol. As such, while most plants are utilized for single applications, rabbit brush could be exploited for both the production of industrial (i.e., rubber, plastics, coatings, lubricants and adhesives) and energy feedstocks (i.e. biodiesel and cellulosic-based liquid fuels).

The goal of this project is to examine the potential of rabbit brush as a multi-use industrial crop for biomaterial and bioenergy applications.

What has been done
Rubber was extracted by centrifugation and/or alginate-creaming agents. Rubber quantities were determined gravimetrically and molecular weights were determined by HPLC gel permeation chromatography. Rubber samples were also subjected to NMR analyses to determine sample purity.

Proteins were removed from extracted rubber and quantified. Rabbit brush rubber associated proteins were then

separated according to their molecular weight, and subjected to an analytical technique to detect specific proteins found in rubber tree latex allergens.

Analyses determined the antigenicity of rabbit brush rubber in regards to rubber trees latex allergies. In collaboration with the Desert Research Institute and Energy Parks Incorp., we were able to obtain and make operational a BIOMAX biomass gasifier and electrical generation system. The BIOMAX will be used in future studies to estimate the energy content of rabbit brush biomass.

Results
One of the major food safety and human health concerns raised in connection with agricultural biotechnology is the issue of allergenicity. These studies showed that rabbit brush rubber has a lower allergenicity and low protein content. "This is a good thing", said Dr. David Shintani, "indicating that it could serve as a source of hypoallergenic rubber".

Dr. Shintani: "There are about 300 medical devices that require natural rubber. Many occupations require people to wear gloves and the rabbit brush rubber does not appear to produce the allergic reactions because of fewer proteins."

4. Associated Knowledge Areas

KA Code	Knowledge Area
204	Plant Product Quality and Utility (Preharvest)

V(H). Planned Program (External Factors)

External factors which affected outcomes

Brief Explanation

{No Data Entered}

V(I). Planned Program (Evaluation Studies and Data Collection)

1. Evaluation Studies Planned

Evaluation Results

{No Data Entered}

Key Items of Evaluation

{No Data Entered}