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2011 Challenges and Opportunities for Industrial Crops



Fargo, North Dakota

September 11-14th, 2011

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ABSTRACTS

Keynote Speaker

THE DYNAMIC WORLD VEGETABLE OIL MARKET: ARE MORE ADAPTATIONS POSSIBLE

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Oilseeds have been the most adaptable crop group due to ability of breeders to alter fatty acids and the market looking for alternate sources of supply. Some of the changes, such as avoiding hydrogenation, have been consumer and market driven. Vegetable oil as a substitute for petroleum based products continues to get market and political attention. Biodiesel has been largely government driven around the world which leaves this industry somewhat vulnerable. New-to-market oilseed crops continue to be researched for unique fatty acids and market applications. The challenge may be increasing for market introductions of new oilseed crops or oils with altered fatty acids in major oilseed crops. There is a considerable market cost and inefficiencies for identity preserved production. Yet consumers want 'natural' products and the environmental community wants 'green' products. The speaker will review the world oilseed complex, adaptations that have occurred, changes that may be expected, market challenges and finally consumer acceptance, and government influences that affect this dynamic industry.

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Plenary Session

CHANGING CROP COMPETITIVENESS IN NORTH AMERICAN AGRICULTURE: BACKGROUND, CHALLENGES, AND OPPORTUNITIES

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There are major changes occurring in world and North American agriculture. Most important is the acceleration in demand for most crops, and that crop productivity has been lagging. This is compounded by differential rates of research and development expenditures across crops, and technologies resulting in differential growth rates. As a result of these changes, there have been shifts in area planted toward crops with greater productivity growth rates, reduced risks and ultimately higher profitability.

The objective of this presentation is to provide background to the above. It will include a review of changes in major world agricultural fundamentals, and expenditures on public and private sector research and development. In addition, the challenges and opportunities are discussed, as well as some economic analytical methodologies (real options) will be used to illustrate the value of new crops and/or traits.

The methods will draw upon the presenters existing work and published studies in areas of new crop development, R&D, and agbiotechnology.

The organization of the presentation includes discussions on 1) Changes in agricultural supply/demand fundamentals; 2) R&D expenditures in competing crop technologies (specifically, biotechnology and conventional); 3) Changing competitiveness of crops in terms of yields, crop profitability, risk and spatial geography; 4) Economic implications for farmers to end-users including discussion on marketing, information, and agronomics for established versus industrial crops. Finally, these will be summarized in terms of opportunities and challenges for new crop development.

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MENON BIOCONVERSION PROCESS FOR FUELS

Peter V. Czipott

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Menon began evaluating strategies to apply microbiology, which is its core technology focus, to the problem of renewable fuels development in 2007. This evaluation looked at fermentation to alcohols and synthesis to triacylglycerides (TAG). After studying the capabilities of different microbes from its library of environmental samples from around the globe, Menon identified economically promising microbes that digest cellulosic biomass and store excess energy as TAG. This pathway has the greatest chance for commercial success due to the emergence of viable processes to transform TAG into hydrocarbon fuels and our nontoxic, aerobic microbial strains that can (1) efficiently produce TAG, (2) produce high-value co-products, and (3) be easily dewatered and processed for oil extraction.

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THE FATTY ACID BIOREFINERY – A COMMERCIALY VIABLE CONCEPT FOR THE PRODUCTION OF RENEWABLE

TRANSPORTATION FUELS AND CHEMICALS

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There are currently many different approaches being proposed to replace petroleum-derived fuel and chemical products with renewable alternatives. The fatty acid biorefinery is a suite of technologies that allows a variety of renewable feedstocks, including crop oils, algal oil, and lignocellulosic biomass, to be converted into a suite of fuel and chemical products as well as other valuable by-products. The heart of this pathway is the noncatalytic cracking of triacyl glyceride oils which produces a suite of organic chemicals that can then be efficiently processed into drop-in compatible transportation fuels and chemicals. Results from a wide variety of feedstocks will be presented.

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CHALLENGES IN SEED GERMINATION AND DORMANCY FOR NEW INDUSTRIAL CROPS

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Plants provide a unique source of carbonaceous compounds for industrial applications including carbohydrates for cellulosic biofuel production, and lipids for biodiesel or other industrial uses. The primary advantage is that plants are renewable source of these raw materials and thus provide a desirable 'green' alternative to petro chemicals. Industrial crop plants can be grouped into different categories: 1) sexual (seed) or asexually propagated and 2) non-agronomic or agronomic crops. The focus of this discussion is on seed propagated, non-agronomic crops as seeds from this group may have seed dormancy and variable seed quality. These seed attributes can reduce both the uniformity and percent germination, thus negatively impacting field seedling establishment.

Seed testing provides information on the seed quality of a particular lot including purity, germination and dormancy with the sum of the latter two comprising viability. Nonviable seeds do not contribute to seedling establishment, so can be factored out; however, an increase in the percent nonviable seeds is associated with seed aging and the deterioration of the seed lot. Dormancy is more problematic as these seeds are living, and may germinate over time resulting in non-uniform stands.

Most potential industrial crops discussed at this conference are seed propagated, non-agronomic crops. Selected species are found in five plant families: Poaceae, Euphorbiaceae, Brassicaceae, Asteraceae and Lamiaceae, and all families have in common the seed dormancy type 'physiological dormancy'. Physiological dormancy is caused by a physiological inhibiting mechanism of the embryo that prevents radicle emergence. However, structures that cover the embryo, including endosperm, seed coats, and indehiscent fruit walls, may play a role in preventing germination (Baskin and Baskin, 1998).

The depth of seed dormancy for a particular genus species is dependent on variety, and seed lot, and examples of each will be illustrated for switchgrass (*Panicum virgatum*). 'Espresso' switchgrass, developed by B. Baldwin was selected for low dormancy. Our results confirm little dormancy, even when tested under a stress temperature of 30°C. Four commercial seed lots of 'Kanlow' switchgrass were obtained and the percent germination ranged from 83 to 20, while the percent viable seed ranged from 94 to 23. Seed vigor tests can be adopted for industrial crops to provide supplemental information for the seed label. This information can be helpful in determining seeding rates compared to the use of PLS (pure live seed). Physiological dormancy largely attributed to the seed coat acting as a physical barrier will be illustrated for castor (*Ricinus communis*) seeds. Removal of the caruncle did not affect germination, while scarification beneath the caruncle enhanced both the percent and uniformity of germination. Scarification improved germination under cool temperatures and in the field.

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SUSTAINABLE APPROACHES TO BIONENERGY R&D

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The Food Conservation and Energy Act of 2008, also known as the Farm Bill, describes all the funding authorities and programs under which USDA operates through 2012. The Farm Bill established the National Institute of Food and Agriculture (NIFA) with the mission to advance knowledge for agriculture, the environment, human health and well-being, and communities by supporting research, education, and extension programs in the land grant university system and other partner organizations. Within this mission, bioenergy has been designated as a priority topic area and is part of the Institute of Bioenergy, Climate and Environment, one of four institutes in NIFA. The goal of the Institute is to promote energy independence through clean, biobased energy systems, and to ensure sustainable and adaptive agro-ecosystems in response to climate change. In order to accomplish this goal, bioenergy research and development is supported in the context of a sustainable supply chain.

Sustainability is an overarching theme for all bioenergy programs in NIFA. From an agricultural perspective, sustainability is defined as satisfying America's needs for food, fiber, feed, and fuel, while at the same time maintaining or enhancing environmental quality, rural economic viability, and quality of life. In order to address sustainability in its fullest sense, two programs have taken a holistic approach to bioenergy and biobased products R&D, and they are the Agriculture and Food Research Initiative (AFRI) Sustainable Bioenergy Challenge and the Biomass Research and Development Initiative (BRDI). These two competitive grant programs encourage the formation of multidisciplinary teams that can move technologies to implementation and successful commercialization.

This presentation describes the status of the programs and how sustainability will be quantified. A framework of interest to NIFA is the Global Bioenergy Partnership (GBEP), an internationally accepted program that identifies criteria and indicators for measuring sustainable biofuels production.

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Oilseeds

CANOLA PRODUCTION AND PROCESSING IN THE NORTHERN PLAINS

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Production of canola in the Northern Plains has increased dramatically in the last decade due to increased demand for the use of canola oil for food and fuel purposes. North Dakota leads the nation in the production of canola, accounting for nearly 90% of U.S. production. End use demand for canola oil continues to increase primarily from food demand driven by the nation's desire to eliminate *trans* fats from consumer diets. Having the lowest saturated fat content of common vegetable oils, canola oil is capturing a growing share of the food market.

North Dakota has several canola processing plants adding value to the crop grown in the Northern Plains. It is also home to a large canola biodiesel processing plant. Investment in the processing sector has increased rapidly in the last decade as more plants are processing canola and plants have expanded production capacities. This has resulted in a greater demand for more canola acres in this region of the U.S. as well as Canada.

Canola oil demand will continue to increase as new varieties are continually developed that offer favorable fatty

acid profiles for consumers and increased yields for canola growers. The canola oil marketplace will include both traditional canola as well as 'designer' canola, each having fatty acid profiles specific to end use needs. Food demand will continue to be the highest value market for canola oil, while industrial use will be dependent on biofuel subsidies and the market value of canola RINS in the biofuel marketplace.

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CAMELINA: A DESIGNER OILSEED CROP FOR METABOLIC ENGINEERING OF IMPROVED BIOFUELS AND BIOLUBRICANTS

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Camelina sativa (false flax) has been cultivated for over 3,000 years, but is an emerging Brassicaceae oilseed crop for semi-arid regions, including the Great Plains and Pacific Northwest of the United States. The growing interest in camelina is due largely to its potential for biodiesel and jet fuel production in geographic areas that are not well-suited for cultivation of established oilseed crops such as soybean. Camelina is also not widely grown in the United States and most of the world for food use. The cultivation of camelina for biofuels will therefore not compete with food crops for prime agricultural land. Our research is focused on developing camelina as an industrial oilseed crop for the production of designer vegetable oils for lubricants, biofuels, and high-value industrial oils. Camelina is an ideal crop plant for metabolic engineering because it can be genetically transformed by a simple floral vacuum infiltration of agrobacterium. With this method, metabolic engineering of camelina can be conducted in a non-labor intensive manner to rapidly develop new traits.

A biotechnological pipeline has been initiated to generate camelina lines with improved fuel and lubricant properties (e.g. enhanced oxidative stability). We are also developing field capacity to evaluate the agronomic properties of engineered lines and to generate sufficient quantities of seeds for oil functionality testing.

Single and multi-gene oil traits engineered to date include high oleic acid, high vitamin E, high saturated fatty acids, and wax esters, an alternative fatty acid storage form. Progress has also been made in introducing pathways for short- and medium-chain fatty acids into camelina for jet fuel-type functionality of the seed oil. In addition, research has been conducted with 454 DNA pyrosequencing to identify nearly all of the expressed oil and seed storage protein genes in camelina seeds. This information will facilitate efforts to genetically improve camelina as a superior industrial oilseed crop by breeding and biotechnology.

The availability of an easy and rapid genetic transformation protocol, an extensive metabolic engineering toolbox, and access to genetic resources make camelina an attractive crop platform for the production of biobased industrial compounds, including biofuels and biolubricants.

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WHAT ARE MAXIMUM YIELDS OF CASTOR SEED?

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Castor (*Ricinus communis* L.) is an endospermic, dicotyledonous plant adapted to both tropical and temperate regions. The extensive endosperm of castor gives this plant the unique ability to accumulate seed oil contents up to 60%. The oil of castor is rich in ricinoleic acid which is both a valuable industrial and an energy dense biofuel feedstock. The combination of 1) very high potential seed yield, 2) extremely high oil content, and 3) adaptation to mechanized production allows castor to produce the highest oil yield of any temperately adapted oilseed crop. Our hypothesis was that by using varieties with semi-dwarf internodes and carefully selected input resources, castor seed production approaching 5 Mg/ha could be obtained. Five semi-dwarf internode varieties are being grown at optimized plant populations, fertilization, irrigation, and plant growth regulators at Lubbock, TX to develop the technology required to grown in excess of 2500 liters/ha of oil. This region is virtually free of pathogens and insect pests that attack castor. Production of castor at 5 Mg/ha on 4 million hectares (10 million acres) would supply the U.S. 15 billion liters (3.8 billion gallons) of renewable fuel and the high protein meal necessary to finish 130 million kilograms (285 million pounds) of beef annually.

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EFFECTS OF PLANT GROWTH REGULATOR ON CASTOR (*RICINUS COMMUNIS* L.) DEVELOPMENT AND SEED YIELD PRODUCTION ON THE TEXAS HIGH PLAINS

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Petroleum is an essential commodity needed by man for food production, as well as to keep our energy based economy operating at optimum performance and efficiency. Castor (*Ricinus communis* L.) has tremendous potential as an oilseed crop to produce biofuels and industrial oils. However, castor plants can reach heights of over three meters in a single growing season limiting mechanical harvest. The purpose of this research was to evaluate the effect of plant growth regulator application on mature castor plant height and seed yield. Castor (cv. Hale) was grown over a three year period on sub surface drip irrigation. Mepiquat chloride plus cyclanilide (2007-2009) and mepiquat chloride alone (2008-2009) were applied at four rates (0, 380 ml, 511 ml, and 731 ml/ha⁻¹) to castor at five stages of growth (6th node, 8th node, first flower, 1st node past flower, and second flower). There was a strong interaction between environment (year) and the effects of both mepiquat chloride plus cyclanilide and mepiquat chloride alone on plant height, raceme numbers, and yield. In 2007 and 2009, application rate and timing had significant effects on these measures. However in 2008 excessive rain in September resulted in no significant differences using Mepiquat chloride plus cyclanilide and Mepiquat chloride alone. The results of this research will allow farmers to enhance the production and mechanical harvest efficiency of castor grown as vegetable oil for feedstock for biofuels and industrial oils.

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WEIGHT OF INDIVIDUAL CASTOR SEEDS BY POSITION ON THE RACEME AND IN THE CAPSULE

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Castor (*Ricinus communis* L.) is an industrial oilseed crop. India, China, and Brazil produce around 1.6 million tons of castor seeds each year. We conducted a broad investigation on the ability of the castor plant to adjust yield components as a strategy to maximize seed yield under drought stress. It was found in a previous study that individual castor seed weight does not follow a normal distribution, but there is a noticeable amount of light seeds in the distribution of seeds produced during drought stress. Our hypothesis is that those light seeds play a role on the adjustment of yield components under drought stress.

In this study we aimed to assess if the individual seed weight is evenly distributed along the raceme and among the three fruit locules, and if light seeds can germinate. This information is useful for planning harvest, managing pests/diseases, and selecting genotypes.

Racemes were obtained from an experiment on drought tolerance (irrigation levels vs. genotypes) conducted in 2010 at the Experimental Farm of Texas Tech University (Lubbock, TX). Four replications of primary mature racemes were harvested from two genotypes (BRS Energia and AL Guarany) and two water conditions (rainfed and 6 mm of daily irrigation). Position of each fruit on raceme was recorded, and each seed was weighed. Relative seed weight was the ratio of seed weight and the maximum seed weight on the same raceme. Germination was measured in seeds classified into seven groups by relative seed weight.

In racemes of water stressed plants, the seed weight was found to increase from the bottom to the top, while in racemes of irrigated plants the seed weight increased from the top to the bottom. When all treatment are analyzed together there is no significant trend, and we concluded that changes in the seed weight along the raceme can occur randomly. The probability of finding a fruit with at least one light seeds is higher at the bottom (18.6%) than at the top (6.9%) of BRS Energia racemes, but in racemes of AL Guarany the probability is lower in the bottom (59.2%) than in the top (75.1%). Fruits with 4 seeds are rare (1.6%). Light seeds (relative seed weight below 0.6) have very poor germination capacity. The frequency of fruits with at least one light seed (relative seed weight < 0.4) was different between genotypes (32.3% in racemes of BRS Energia, 55.6% in racemes of AL Guarany), but it was not influenced by water availability. When a fruit has one light seed, the other seeds in the same fruit are not favored. Thus, there is no compensation in this yield component at the fruit level.

We conclude that along the castor raceme there is no clear trend to change the weight of individual seeds or the probability of finding fruits with light seeds. The wt of a seed is not increased if occurs a light seed in the same fruit. The frequency of fruits with light seeds was influenced by genotype but not by the drought stress. Light seeds germinate poorly.

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BIOTECHNOLOGY DEVELOPMENT FOR LESQUERELLA FENDLERI TO IMPROVE HYDROXY FATTY ACID SYNTHESIS

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The conventional source of hydroxy fatty acid (HFA) is ricinoleic acid (18:1OH) from castor seeds. Ricinoleic acid and its derivatives are used as raw materials for numerous industrial products, such as lubricants, plasticizers and surfactants. The production of castor oil, however, is hampered by the presence of the toxin ricin and hyper-allergic 2S albumins in its seed. Seed oil of *Lesquerella fendleri* (*Lf*) contains a valuable HFA, lesquerolic acid (20:1OH), and does not have such toxic compounds, thus its oil represents an alternative source of HFA. Lesquerolic acid is derived by a 2 carbon elongation of 18:1OH. By suppressing the elongation step through genetic engineering, it is possible to generate a new *Lf* crop that could produce 18:1OH.

To understand HFA synthesis and regulation in *Lf*, we investigated morphological and physiological changes, as well as temporal details of fatty acid composition and gene expression during seed development. We found the synthesis of 20:1OH regulated mainly by gene transcription of a 3-ketoacyl-CoA synthase in *Lf*.

We also developed an efficient procedure to eliminate the chimeras and established a stable genetic transformation system in *Lf*. With aid of a β -glucuronidase gene that confers blue color in transgenic cells, we demonstrated a seed-specific promoter from *Brassica napus* that is activated only in seed of *Lf*. In addition, we determine the copy number in the transgenic *Lf* using a qPCR assay. The qPCR method allowed the selection of desirable transgenic lines through screening of a large population and saved greenhouse space, which increased the capacity of the transgenic production pipeline.

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SEED GERMINATION STUDIES ON VARIOUS PHYSARIA SPECIES

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Physaria species are valued for novel hydroxy fatty acids in their seed oil. Considered among the new crops for commercialization, the National Plant Germplasm System has a valuable array of germplasm to support cultivar development activities in the primary species lesquerella (*P. fendleri*).

In this study, two separate experiments comparing seed germination response among different species of conserved *Physaria* (*P. argyraea*, *P. fendleri*, *P. gracilis*, *P. rectipes*, *P. recurvata*, *P. sessilis*, and *P. thamnophila*), as well as materials from *P. fendleri* and *P. gordonii* collected from natural populations were conducted.

Freshly harvested seeds were subjected to different afterripening durations (4, 8, and 12 weeks) and six storage conditions utilizing combinations of saturated salt solutions (LiCl and MgCl₂) and temperatures (5, 25, and 35°C). The germination assays were conducted with light (1,052 lux) and gibberellic acid (GA₃) (100 ppm) and without them.

In both experiments using materials from the conserved accessions as well as those from the wild, higher germination percentages were observed on assays conducted with light and GA₃. There were slight differences among the total germination percentages after the afterripening durations detected on *P. fendleri*, *P. recurvata*, and *P. thamnophila* with higher values after the longest storage duration considered in the study.

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DOUBLE-CROPPING CAMELINA AND SOYBEAN IN THE NORTHERN CORN BELT

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Growing camelina as a winter annual crop provides potential economic and environmental benefits that may make producing this relatively new industrial oilseed crop attractive to farmers. Previous research showed that winter camelina in the northern Corn Belt can be harvested early enough to allow producing a second crop in the same season. Soybean serves well as a second crop following camelina. However, further work is needed to develop this cropping system to optimize yields of both crops to improve economic returns. Another issue of double-cropping (DC) is seasonal water use.

The objectives of the following study were to determine the best method of DC winter camelina and soybean in west central Minnesota to improve crop yields while minimizing inputs and to determine seasonal crop water use.

Double-cropping treatments included sequentially following camelina with an early maturing soybean (MG 00) and inter-seeding a full-season soybean (MG I) into camelina (relay-cropping) early in the spring. Swathing and

herbicide treatment of camelina were also evaluated as methods to hasten camelina harvest. Total crop water use was assessed in all treatments including full-season mono-cropped soybean.

Camelina seed yields ranged from 1106 to 1393 kg ha⁻¹ among treatments, but were not significantly different, and oil content ranged from 40 to 42% (wt wt⁻¹). Moreover, camelina provided good weed suppression throughout the early growing season. Soybean yields ranged from 1765 kg ha⁻¹ (26.5 bu acre⁻¹) for sequential DC to 2764 kg ha⁻¹ (41.5 bu acre⁻¹) for relay-cropping. As expected, crop water use was higher for the DC treatments, but not greatly different than a full-season soybean crop.

Results indicate that a winter camelina-soybean DC system is feasible for the Corn Belt region and may be a sustainable means of producing biofuel feedstock without sacrificing food/feed production.

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GENETIC IMPROVEMENT OF CAMELINA FOR USE AS BIOFUEL FEEDSTOCK

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Camelina-derived jet fuel has emerged as an attractive option in the aviation industry's efforts to reduce its dependence on fossil fuels. Increased productivity of the feedstock is of importance for the economic viability of the initiative. Development and use of genetically superior seed constitutes a practical strategy to achieve that goal. In this study agronomic data collected from an applied breeding program in camelina implemented by Sustainable Oils, LLC was used to determine the levels of adaptability of camelina to the northern US and southern Canada, to understand the causal mechanisms associated with the response, and to evaluate the response to conventional selection efforts.

Data from two groups of tests were used in this study. Group 1 consisted of four sets of field experiments conducted from 2006 to 2009 across 19 locations in the US and 3 locations in Canada (51 environments) in which a total of 79 camelina accessions were evaluated. Group 2 consisted of one set of experiments conducted in 2009 and 2010 across 8 locations in the same area (11 environments) in which 7 F2:4 and 15 F2:5 breeding populations obtained using conventional breeding protocols were evaluated. In each experiment the plant materials were tested using a randomized complete block design with four or three replications under dryland, low moisture conditions. Data was subjected to standard statistical analysis, response to selection was calculated using the variance component estimate approach and the impact of genotype-by-environment interaction was assessed by measuring the degree of crossover interaction observed for seed yield using a balanced data set from Group 1

Seed yield among accessions ranged from 1375 to 1721 lbs/ac with an average of 1545 lbs/ac and it was demonstrably higher among populations, ranging from 1418 to 1743 lbs/ac with an average of 1628 lbs/ac. The levels of variation in seed yield among environments indicated that populations were more responsive to environmental fluctuations than accessions. A similar trend in response between these genotypic groups was observed for seed weight, oil content and oil yield (1.09 and 1.27 g/1000 seed weight, 36.6% and 37.6% oil content, and 557 lbs/ac and 639 lbs/ac). Consistently, materials with increased seed and oil yield had shorter bolting and flowering days and increased seed filling period and seed weight. Although a relatively large genotype-by-environment interaction variation was detected, which appeared to be of cross-over type, broad-sense heritability estimates were of magnitude (0.72, 0.98, 0.84, and 0.76 for seed yield, seed weight, oil content, and oil yield). Response to direct selection for seed yield corresponded to 80 lbs/ac (4.9%) and correlated response on oil yield, seed weight and oil content was high (0.91, 0.60, and 0.65). Predicted seed and oil yield of select populations were 1708 lbs/ac and 618 lbs/ac, which were ~11% higher than the average observed for accessions. The levels of adaptability and the amount of genetic gains observed indicate that development of camelina varieties with superior oil productivity for the northern-tier US & southern Canadian prairies should be highly feasible.

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THE EFFECT OF SEEDING DATE AND SEEDING DEPTH ON CAMELINA IN SASKATCHEWAN, CANADA

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Currently, there are three potential markets for oil from camelina, human consumption, biofuel, and as a crop that could be genetically modified to produce specialty oils for high value industrial products. If demand for camelina oil increases there are issues around production practices that need to be addressed for the successful commercial production of camelina. The objective of this research was to evaluate the effect of two production practices seeding date and seeding depth on the production of camelina. Two experiments were used to conduct this research at several locations across Saskatchewan. The first experiment consisted of eight seeding dates with four in the fall and four in the spring starting in early October and ending in June. No camelina was seeded between the middle of November until the middle of April. The second experiment consisted of two seeding dates, one in the fall and the other in the spring and four seeding depths, 0, 0.635, 1.25, and 2.5 cm. All camelina seeded in the fall started to germinate as soon as soil moisture was available and at most sites in most years this occurred at seeding. The grain yield of the fall treatments has been more variable than the spring seeded treatments. The last seeding date in the fall which usually germinated but did not emerge until spring usually had grain yield that was similar to the early spring seeded camelina. Camelina seed consistently emerged from a depth of 0 to 2.5 cm. The seed placed on the surface was packed into soil disturbed by the fertilizer portion of the opener on the seeder. Therefore it is recommended that producers seed camelina in late fall or early spring with a seeding depth of 0.635 to 1.25 cm.

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WINTER CANOLA IS A POTENTIAL SOURCE TO DEVELOP HIGH PERFORMANCE SPRING CANOLA GERMLASM

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Canola is a strategic crop of great importance to North Dakota, is the largest producer of canola in the USA. Increasing seed yield in canola is an important breeding objective for the breeders to satisfy the demand of the growers and seed processing industries. On the basis of growing season, rapeseed (*B. napus*) is classified into two groups, such as annual type and biennial type. The annual type is also known as spring type and biennial type is known as winter type. The yield potentiality of winter type rapeseed is over two-fold compared to spring type canola. Because of severe winter hardiness, winter type canola is not possible to grow in North Dakota.

The objective of this study is to develop high seed yield spring canola utilizing winter type rapeseed.

Crosses were made between winter (cv. ARC-97108) and spring (cv. Regent) type canola. The winter parent ARC-97018 was vernalized to get flower in the vernalization chamber of the North Dakota State University. The F₁, F₂, and backcross progenies were grown in the greenhouse of the North Dakota State University. Data on day to flower,

plant height, number of branches/plant, number of pods/plant, pod length, root length, dry stem weight, dry root weight, and seed yield/plant were taken to study the correlation of coefficient among the characters using SAS 9.1 software (SAS Institute Inc., Cary, NC, USA).

Vernalization requirement to flower has been identified as a recessive trait. Two genes loci are responsible for the vernalization requirement of *B. napus*. Significant positive correlations were found between branches vs. number of pod (0.58***), branches vs. dry stem weight (0.38*), branches vs. seed yield (0.49**), number of pods vs. root length (0.41*), number of pods vs. dry stem weight (0.43*), root length vs. dry root weight (0.46*), root length vs. seed yield (0.37*), dry stem weight vs. seed yield (0.54**).

Two recessive genes are responsible for vernalization in *B. napus*. Plants with vigorous root system showed a positive correlation with number of pods and seed yield.

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SEEDING DATE INFLUENCE ON CANOLA PERFORMANCE IN NORTH DAKOTA

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Poor field conditions, related to precipitation and temperature, often prevent planting of crops with prolonged delays compromising performance. Knowing when crop performance begins to decline would help growers target when to have completed planting to achieve highest crop performance.

The objective of this study was to determine the influence of seeding date on canola (*Brassica napus* L.) performance at several locations with different geography.

The study was a RCBD, with four replicates, in a split-plot arrangement where the main plot was seeding date and the subplot was hybrid. Seeding dates began in late April or early May, were spaced at approximately 10 d intervals, and concluded about 10 June for a total of five dates. A Roundup Ready and Liberty Link hybrid were seeded at 5.6 kg/ha in solid-seeded plots approximately 1.5 x 6.2 m with row spacing of 15 cm. The study was conducted at four North Dakota locations in the 2010 season.

At the northern locations yield was maintained at the first two seeding dates before decline at later dates, but at the southern Hettinger location yield decline occurred with each successive seeding date. Seed yield reduction from date 1 to date 5 ranged from 50 to 60% at three of the four locations. Seed oil content decrease across seeding dates ranged from 4 to 8.5% among locations, but oil content was similar at dates 1 and 2 followed by decline at later seeding dates. Days from seeding to maturity decreased from 95 (Date 1) to 75 days (Date 5) at the Carrington location with duration of vegetative and reproductive periods decreasing equally as seeding date was delayed. At two locations the seeding date by hybrid interaction indicated hybrids responded differently at early and late seeding dates.

Based on these results seeding before 15 May and 1 May, at northern and southern locations, respectively, is recommended for achieving highest yield.

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VARIATION IN SEED LIPIDS IN CALENDULA GERMPLASM

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Calendula officinalis (pot marigold) has considerable promise as an industrial crop. It has a long history both as an ornamental and a medicinal plant. In addition, it is marketed as an ingredient in cosmetics and as a colorant. It also displays unusual seed-lipid chemistry, which can provide an additional market for commercial *Calendula* producers, potentially spurring expanded production. More than 80 diverse accessions of *Calendula*, including wild relatives, have been assembled at the North Central Regional Plant Introduction Station (NCRPIS) and are available for research. This collection and a survey of its uses will be presented in our talk.

The objective of our preliminary study was to sample all available *Calendula* seed lots, primarily regenerations made at the NCRPIS, but including some original lots received from other donors or collectors for comparison, with the goal of documenting the range in variation in seed oil percentage and fatty-acid composition. Since these seeds were produced between 1965 and 2010, results are likely to be affected by differential stability in cold storage, but our results can be used as a starting point for future, replicated analyses of samples produced in a common environment. We extracted 88 samples and used pulsed NMR calibrated with calendula oil to determine total oil percent reported on a dry weight basis. Fatty acid methyl esters were made directly from the seeds using 0.5M sodium methoxide and analyzed by gas chromatography with an SP2330 30m×0.25mm i.d. column for isolation, and a flame ionization detector for quantification based on equivalent chain length values.

From an oilseed perspective, our *Calendula* samples generally were low in total seed oil, ranging from 5.7 to 16.5%, due in part to the morphology of their extensive seed coats. However, the primary fatty acid was the unusual omega-6 compound, calendic acid (8*trans*, 10*trans*, 12*cis*-18:3), which ranged from 31.7 to 67.8% of total seed oil. In five comparisons between paired samples of single accessions regenerated at the NCRPIS in Ames, IA with those grown elsewhere, four Ames regenerations displayed 6.3 to 15.7% lower proportions of calendic acid, compensated by increased proportions of linoleic and/or oleic acids. Wild *Calendula* species often presented fatty-acid profiles beyond the range found among the 40 *C. officinalis* samples. For example, the highest proportion of calendic acid (67.8%) was found in *C. maroccana* PI 607417, of linoleic acid (39.6%) in *C. arvensis* PI 597586, and of oleic acid (24.0%) in *C. stellata* PI 649651. These wild taxa may be useful in breeding *Calendula* cultivars with modified fatty-acid composition.

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POST-EMERGENCE HERBICIDES USEFUL IN CALENDULA

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Easy and effective weed control is required by growers who are considering new industrial crops. Post-emergence herbicides typically are the products of choice by today's growers. Unfortunately, post-emergence herbicides with proven safety margins are not known for calendula (*Calendula officinalis*), whose seed oil has applications as a replacement for volatile organic compounds used in paints and other finishes.

Our immediate objective was to examine the tolerance of calendula to a number of post-emergence herbicides commonly used for broadleaf weed control in other crops. The longer term objective was to include calendula on the label of at least some of these products.

Fifteen postemergence herbicides were tested under greenhouse conditions and six of these were tested further under field conditions. In all cases a dilution series of concentrations were examined. The concentrations ranged from 1% to 1000% of the normal label rate in greenhouse tests, and 25% to 100% of the normal label rate in field tests. Plants were at the 2- to 4-leaf pair stage at the time of treatment. Plants were scored for damage and weighed about two weeks after treatment in the greenhouse. Field-grown plants were scored for damage while immature and, subsequently,

harvested for yield at seed maturity.

Only three post-emergence herbicides appeared to be tolerated by calendula. These were desmediphan + phenmedipham, imazamethabenz, and MCPB. These herbicides are the active ingredients in commercial products such as Betamix, Assert, and Thistrol, respectively. Calendula seedlings exhibited no damage symptoms with imazamethabenz treatments, slight deformity with MCPB, and substantial but ephemeral leaf damage with desmediphan + phenmedipham. Plants treated with these herbicides appeared normal at seed maturity. Seed yields of plants treated with desmediphan + phenmedipham, imazamethabenz, or MCPB did not differ appreciably from those of non-treated control plants.

At present desmediphan + phenmedipham, imazamethabenz and MCPB are three post-emergence herbicides that appear to be tolerated sufficiently by calendula to merit further testing. If tolerance is confirmed, the activity spectra regarding susceptible weed species is complementary among the three herbicides. For instance, of locally important broadleaf weed species, imazamethabenz controls only kochia (*Kochia scoparia*), wild mustard (*Sinapis arvensis*), and wild buckwheat (*Polygonum convolvulus*). Desmediphan + phenmedipham control the three latter weeds as well as buffalo bur (*Solanum rostratum*), cocklebur (*Xanthium strumarium*), common lambsquarters (*Chenopodium album*), common ragweed (*Ambrosia artemisiifolia*), eastern blacknightshade (*Solanum ptycanthum*), and various pigweeds (*Amaranthus* spp.). MCPB adds control of thistles (*Cirsium* spp.), field bindweed (*Convolvulus arvensis*), and sowthistles (*Sonchus* spp.). Although many other broadleaf weed species exist, control of the above species by desmediphan + phenmedipham, imazamethabenz and MCPB may facilitate the commercialization process.

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INFLUENCE OF ENVIRONMENT AND AGRONOMIC PRACTICES ON EUPHORBIA SEED AND OIL YIELD

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In the late 1950s the USDA first recognized that *Euphorbia lagascae* was nearly unique among plants in that its seed oil contained high levels of vernolic acid, an epoxidized fatty acid of interest to the paint and coating industry as a drying solvent in alkyd resin paints. Euphorbia is a drought-tolerant native of Spain whose seed contains about 45%-50% oil, of which 60%-65% is vernolic acid. Using “non-shattering” mutants developed in Spain, previous research in Oregon’s Rogue Valley demonstrated that euphorbia could be grown and harvested in the US, and that it required much less irrigation water than typical crops.

The objective of these multiyear studies was to evaluate the effect of agronomic factors on crop growth, seed yield, and seed oil production under varying climatic conditions.

Factors evaluated in these field studies included planting date, irrigation, fertilizer, seeding density/row spacing, harvest method, seed type, and herbicide tolerance. Studies were conducted over several years in two differing climate areas (Rogue Valley and Klamath Basin) to gauge euphorbia’s climatic limits and response to varying weather patterns.

Euphorbia was able to grow and produce a seed yield of approx. 100-200 kg ha⁻¹ with no irrigation and only a few cm of natural rainfall, but yield increased three-fold when approx. 15 cm of irrigation was applied. Maximum seed yield in these studies approached 1000 kg ha⁻¹. Plants grew more upright with fewer side branches when in narrow rows and were broader with more branches when in wider rows, but Euphorbia seems to be morphologically “plastic” and thus seed yields were not well-correlated with row spacing. Early seeding dates (April or earlier) enhanced branching, earlier maturity and good seed formation. Euphorbia did not show a significant response to added N fertilizer. Because euphorbia’s seed do not mature uniformly, it was beneficial in some cases to direct combine-harvest when approx. 1/3 of the seed pods were brown, and then re-combine the same material a week later to thresh the remaining seeds after drying. These later seeds had slightly lower oil content than fully mature brown seeds. Several pre-plant incorporated (PPI) and pre-emergence (PRE) herbicides appear to hold promise for weed control programs in euphorbia, including benefin, ethalfluralin, trifluralin, and pendimethalin, while other PPI and PRE herbicides are clearly toxic to euphorbia. While many common post-emergence (POST) herbicides damage or kill euphorbia, euphorbia tolerated some POST herbicides well, including clopyralid, chloridazon, alachlor, oxyfluorfen, bromoxynil, and acifluorfen, although euphorbia

appears to be sensitive to specific rates in some cases.

Euphorbia's ability to produce a reasonable seed yield, as well as its ability to be grown using standard farming equipment with limited modifications, suggest it could be a viable crop in arid regions, including much of the western US. Its climatic limits need to be further evaluated to avoid problems such as the immature or abnormal seed heads we have observed when tested under a short growing season.

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COLLECTION AND EVALUATION OF FIELD PENNYCRESS (*THLASPI ARVENSE* L.) POPULATIONS

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The common winter annual weed *Thlaspi arvense* L. (field pennycress) has been recently identified as a potential new source for fuel and industrial products. Field pennycress offers a very short life cycle allowing for opportunities for double cropping in the Midwest region. The current USDA collection has less than 14 accessions of *Thlaspi arvense* available for distribution.

The objective of this study was to collect and evaluate diverse wild populations of field pennycress (*Thlaspi arvense* L.) for potential inclusion in a plant breeding program.

In the springs of 2009-2011, wild populations of field pennycress seed were collected from 3 distinct geographical regions; 12 local populations in McDonough County, Illinois, 30 wild populations from across the Central United States, and 18 accessions from other regions of the world. A total of 60 populations were evaluated including 12 accessions from the North Central Regional Plant Introduction Station in Ames, IA. Populations were evaluated for total seed oil and fatty acid methyl ester content. Total oil content was determined by nondestructive pulsed NMR on whole pennycress seed, while fatty methyl esters were quantified utilizing gas chromatography.

Total oil content ranged from 25.9% to 37.2% across all the populations. The variation in total oil was not significantly different between each of the three geographical regions. The highest total oil content was found in populations from Midwest, while the lowest line was collected from Tibet, China. Erucic acid C22 levels ranged from 30.3 to 37.5%, while Oleic acid C18:1 ranged from 7.8 to 11.9%. In the fall of 2011, all 60 lines will be planted in Illinois and evaluated for seed yield and for important agronomic traits.

Pennycress's unique short growing season, high levels of total oil, and erucic acid content increases its viability for commercialization and potential for plant breeding programs.

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DEVELOPING CASTOR COMMERCIAL CROPPING SYSTEMS

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Castor (*Ricinus communis* L.) is an ideal candidate for the production of renewable fuels because it can be grown under fully mechanized production on soils that do not support traditional crop commodities. Castor has very high seed oil content (48-60%); a high life cycle analysis (88 to 97%); a very positive energy balance; and produces high levels of flexible feedstock (150-300 gal. fuel/acre). Historically, commercial production of castor in this country has been limited by the lack of mechanized technologies to produce and process the crop. Additionally, a deterrent to commercial castor production is the potential contamination in a grain crop such as corn. Prior to commercial production, it is essential that

we determine the best methods for volunteer castor management as well as weed management within a castor crop. Thus, the main objectives of this study were to identify optimum management practices for volunteer castor control and identify appropriate herbicide chemistries and application timings for weed management within castor. In 2011, two volunteer castor experiments were setup to compare different herbicide chemistries, multiple rates and three application timings. Likewise, a separate weed management study was conducted to compare multiple herbicides and two application timings within castor. The experiments were setup in a randomized complete block design and replicated four times. Application timings included a pre-plant treatment and post-emergence treatments at 2 and 4 leaves. Regarding volunteer castor control, both Atrazine and Direx applied prior to planting provided 98% and 100% control, respectively. Roundup Powermax, Ignite 280, 2,4-D and Clarity have also provided acceptable post-emergence control. Regarding weed management within a castor crop, preliminary results suggest that Prowl H20, Dual Magnum, Linex, Layby Pro and Select all have potential. Detailed results and application information will be discussed further. It is clear from these initial results that volunteer castor management and weed management within a castor crop will not be limiting factors to commercial castor production.

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NILA BICENTENARIA: A COLOMBIAN CASTOR NEW VARIETY

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A Colombian castor new variety "NILA BICENTENARIA" (*Ricinus communis* L.) was released in March 2011 and it is recommended to plant from 1.800 to 2200 masl (6000 to 7300 feet) for Colombian Andean region. This variety was developed by the Breeding Program of Colombian Agricultural Research Corporation (CORPOICA). This andean variety was obtained by recurrent population improvement, using Half-sib selection with ear (spike)-to-row method cycle III. The original accession of castor coded ILS3078, was donated to the Colombian Castor Collection (CCH) by NOPCO SA Company.

Parental germplasm evaluation was carried out at Corpoica La Selva and Nataima Agricultural Research Centers in 2007A, and replicated trials for selection and recombination were grown at La Selva Center in 2008A and 2009A. 46 Half-sib families were planted in isolation plot at La Selva Center. A combined selection pressure of 10% was made (50% among families and 20% within families, for a total of 23 families and one individual by family were selected). The traits for selection included earliness, disease performance, grain yield and plant type which was carried out through 2010. Over 500 kg of seed were harvested in March 2011, for use in regional and field-scale trials and for releasing a foundation seed.

Nila Bicentenaria plants have reddish stems without wax. The inflorescence is normal monoecious. Stigma color is red. The primary spike is cylindrical in form. Capsules spines and pedicels are long, semi-indehiscent, and hull easily when dry. Seeds are elongated, medium size, brown striped and mottled. The weight of 100 seeds is 59 to 71 g. Mature plant height to the primary raceme is 1.05 to 1.27 m (3.5 to 4.2 feet). Slender stems and branches produce spikes of the second, third and fourth order. The primary raceme is normally produced after 18 to 20 nodes are formed. Plants are tolerant to capsule mold. Oil content is 48%. Nila Bicentenaria yields around 3,500 kg ha⁻¹ yr⁻¹ which were greater than other commercial (i.e. BRS-Nordestina) or spontaneous varieties.

Breeder seed will be maintained by the Castor Breeding Program of the Colombian Agricultural Research Corporation and it will be incorporated to the Commercial Seed National Program in the Colombian Agricultural Institute (ICA).

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SCREENING OILSEED FEEDSTOCKS FOR IMPROVED BIODIESEL QUALITY

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One of our goals is to improve our understanding of genetic and environmental factors impacting quality of biodiesel from canola and other oilseeds. This presentation will provide an overview of several NDSU biodiesel research projects. The ability to screen oilseed feedstocks for biodiesel quality factors including oxidative stability and cold flow properties resulted in the development of a direct (*in situ*) alkaline transesterification procedure where 40 lines /per week can be evaluated. Implementing this procedure, canola biodiesel quality was evaluated across several ND growing locations, years, and varieties and significant differences in oxidative stability and cloud point temperature were detected. The development of NIR spectroscopic methods for rapidly assessing biodiesel quality including oxidative stability, acid value, total glycerin, and cloud point will be discussed. Ongoing research is aimed at identifying minor seed constituents that negatively impact biodiesel quality.

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RENEWABLE OILS FOR USE IN COMPOSITE MATERIALS

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The public increasingly seeks products derived from sustainable, bio-based alternatives to petroleum and produced through environmentally-friendly processes. Our objectives are to (1) produce composite materials with a high content of renewable epoxy resin, and (2) attain thermal and mechanical performance similar to or better than those produced exclusively from petroleum-based epoxies. Resins were produced from refined oils using hydrogen peroxide and catalyst (acetic acid and an ion-exchange resin) in an optimized process. Epoxy resins (epoxidized vegetable oil or EVO) which have been evaluated to date include those from canola and soybean oil, as well as high-oleic canola and high-oleic sunflower oils. The resin was cured and reinforced with E-glass fiber for application in composite materials. EVOs were blended with petroleum-based resin at high levels of 30 to 40% of the matrix before curing. The composite specimens were then tested for flexural properties and subjected to dynamic mechanical analysis. To improve thermal and mechanical performance, we evaluated several parameters, such as the effects of various curing agents and the ratio of curing agent to epoxy resin. Performance of these EVO composites was similar to, but slightly lower than, performance of the petroleum-based controls. Among amine curing agents studied, bis (p-aminocyclohexyl) methane (PACM) at amine: epoxy ratios of 0.8 and 1.0 showed superior performance. E-glass fiber-reinforced composites with PACM/EVO showed thermal and mechanical performance slightly lower than the composites with 0% EVO. Epoxy resins can potentially be produced from a variety of other renewable resources, especially other unsaturated vegetable oils; the approach used here can be readily extended to these other feedstocks.

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Fiber and Cellulosics

AN INDUSTRY PERSPECTIVE ON THE APPLICATION OF LIGNO-CELLULOSIC FIBERS IN THE THERMOPLASTIC COMPOSITE PRODUCTS – FIBER SOURCES, PROPERTIES, AND INNOVATIONS

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Global environmental concerns and issues related to sustainability, recyclability and biodegradability of commercial products have rekindled the interest in natural and agricultural ligno-cellulosic fibers. There is a renewed effort by researchers and industry for identifying raw materials that can be successfully utilized for designing innovative products. Some of these applications include building materials, automotive parts, packaging materials, and OEM products. In the last two decades building products industry has taken significant lead by launching several composite products made from natural fibers.

There are several factors that dictate the acceptance of natural fibers into composite materials or products. Some of the factors for consideration are key physical, chemical and mechanical properties of the desired products, durability characteristics, economics, efficient processing technologies, raw material availability, material consistency, price stability and transportation costs. Case studies of kenaf, flax, and cotton burr fibers have shown that physical properties such as fiber aspect ratio, particle size distribution, moisture content, bulk density, chemical constituents (cellulose, hemicelluloses, lignin, etc), contamination, and silica content are important characteristics. Key mechanical properties for the composite are flexural modulus and strength, tensile/compressive modulus and strength, resistance to moisture, thermal and dimensional stability, fastener holding capacity, impact resistance, UV resistance, and resistance to biodegradation under various temperature and humidity conditions. Developing technical specifications of new materials, and collaborating with focused industrial partners could provide an edge while launching a new product.

Currently there are numerous opportunities for natural fiber filled composite materials. Federal mandate to increase fuel efficiency of motor vehicles by year 2015 is one major factor driving the usage of low cost, light weight, high strength cellulosic fibers in composites. Industry sustainability score card is also promoting the usage of natural fibers. Building material, and paper and packaging industry are now accepting agricultural fibers as an alternative to traditional wood fiber. They are also being researched as a potential source for pure cellulose and carbon fibers.

Significant progress in patents and trademark activity related to natural fibers is also a good indicator of strong future for agricultural fibers. From 2000-2011, more than 100 patents have been granted in the US for biofiber composites.

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PHYSICAL AND MECHANICAL PROPERTIES OF COMPOSITES MADE FROM COTTON BURS, COTTON STALKS, KENAF, AND SOUTHERN PINE BLENDS

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Previous studies evaluating physical and mechanical properties of composites produced from blends of cotton gin byproducts (CGB), which consisted of cotton carpel (burs), and small sticks, and guayule bagasse (GB) indicated these fibers were a viable cost effective replacement for wood fiber. However, water absorption for the CGB and GB blends was greater than the 100% wood composites used as standards. As a continuation of the previous CGB and GB studies, this study was undertaken to evaluate composites produced from blends of cotton burs (B), cotton stalks (S), kenaf (K) and southern yellow pine (P). The objective of this study was to evaluate physical and mechanical properties of composites produced from various blends of B, S, K, and P compared to commercial hardboard and particleboard

standards.

Experiments were conducted on 13 composite boards and compared to standards for hardboard (C-5) and particleboard. The CGB, kenaf, and P were processed through a hammermill and a shaker table so that all particles were within 0.42 to 4mm. All fibers were dried to less than 5% moisture content and blended with a binding agent comprised of 10% melamine modified urea-formaldehyde adhesive with 1.5% wax emulsion. Board construction was accomplished using a 91-Mg capacity oil-heated hydraulic press. Composite boards, 1.1 cm by 40.6 cm by 43.2 cm, were produced using the following blend of fibers: 100% B, 75B/25K, 50B/50K, 25B/75K, 75B/25P, 50B/50P, 25B/75P, 75B/25S, 50B/50S, 25B/75S, 25K/50B/25K (3-layer composite, kenaf face), 25S/50B/25S (3-layer composite cotton stalk face), 25P/50B/25P (3-layer composite southern yellow pine face). The standards used for comparison came from the American National Standards Institute (ANSI A208.1-1993 PB; ANSI A208.2-2002 MDF) for Interior Applications.

Results indicate improved physical and mechanical properties in almost every variable measured for the blended composites compared to composites consisting of 100% B. Most of the blended composites exhibited equal or superior properties than the particle board or MDF except for water absorption. For water absorption, most of the tested blends exhibited significantly higher values than the standards. The most promising blends, for water absorption, were the 25B/75P and the 25P/50B/25P (3-layer composite southern yellow pine face). The data indicate that B can be a good fiber substitute when blended with wood and used in concentrations of 50% or less.

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FORMULATION OF A BIODEGRADABLE, ODOR-REDUCING CAT LITTER FROM SOLVENT-EXTRACTED CORN DRIED DISTILLERS GRAINS

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Cats are among the most popular pets in the U.S., and the majority of these animals are kept indoors where litter boxes containing some type of absorbent litter material are needed. Dried distillers grains (DDGs) are one of the two major co-products (with carbon dioxide) of the corn ethanol industry, and these DDGs are primarily sold as animal feed. Our research group has been studying value-added uses for DDGs by extracting valuable phytochemicals from them with a variety of organic solvents. After extraction, it was found that these extracted DDGs exhibited enhanced water absorption capacity.

The objective of this research was to determine if the extracted DDGs could be utilized as a biodegradable cat litter.

DDGs from whole-kernel corn were extracted for 24 h with hexane using a Soxhlet apparatus to remove oil and other non-polar compounds, and dried for 24 h at 45°C to remove residual hexane. A suspension of 50 g of guar gum (commonly used as a clumping agent) in 500 g glycerol were added to 100 g samples of extracted DDGs at rates of 10, 25, 50 and 100 g, respectively. Clumping activity was determined by adding 5 g of each treatment into 60 mm x 15 mm plastic petri dishes, and allowing 5 ml of water to drip into each plate from a 100 ml burette placed 10 cm above the plates to simulate cat urination. These samples were placed in a drying oven set at 30°C for 24 h. The contents of each plate were emptied onto a 6-mesh sieve (large enough to allow all non-clumped extracted DDGs to fall through) and placed on an orbital shaker set at 250 rpm for 1 min. Clumping percentage was calculated as the weight of clumps retained on the top of the sieve divided by the total weight (5 g) multiplied by 100. Because copper compounds have been found to complex thiol compounds such as the specific cat urine odor compound, a guar/glycerol suspension was also prepared as before with the addition of 1 mg copper sulfate/ml glycerol, and a litter formulation was prepared as before containing 25 ml solution/100 g extracted DDGs. Headspace analysis vials containing litter formulations (1 g) with and without copper sulfate were incubated for 24 h with 0.2 ml of a solution of 3-mercapto-2-butanol before SPME-GC headspace analysis.

Increasing levels of guar/glycerol significantly increased clumping percentage; however, the rate of 25 g solution/100 g extracted DDGs were determined as the most desirable formulation tested. Addition of copper sulfate substantially reduced the volatilization of 3-mercapto-2-butanol into the headspace of the vials.

Our results indicate that extracted DDGs formulated with guar/glycerol/copper sulfate have excellent potential as commercial cat litter.

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LONG-TERM PERFORMANCE OF THERMOPLASTIC COMPOSITE MATERIAL WITH COTTON BURR AND STEM (CBS) AS PARTIAL FILLERS

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Past research has shown good potential for using cotton burr and stem (CBS) fraction of cotton gin byproducts as a fiber filler in thermoplastic composites. However, water absorption of this new composite was a problem, with significantly higher values than thermoplastic composites containing oak wood fiber (OWF) as the filler. Lab-scale studies showed that adding 30% of guayule bagasse (GB) to CBS may reduce water absorption significantly. There are also well recognized coupling agents that would reduce the water absorption and enhance the strength properties.

The objective of this study was to evaluate 3 different methods to reduce the water absorption of thermoplastic composites containing CBS fillers.

An experiment was conducted with 6 different treatments to reduce water absorption of composite containing CBS. The six treatments were T1: 50% OWF + no CBS, T2: 25% CBS + 25% OWF, T3: 25% CBS + 25% OWF +1% compatibalizer; T4: 25% CBS + 25% OWF+1% coupling agent, T5: 17.5% CBS+ 7.5% GB + 25% OWF, and T6: 17.5% CBS+17.5%OWF+15%GB. All fiber materials were ground to a size of 20-60 mesh. The composite contained 50% by weight of fiber filler. Samples were manufactured in to a commercial decking profile of 25 x 152 mm with a single screw extruder. The extruded samples were water cooled, conditioned, and then tested for water absorption and flexural strength based on ASTM D1307-2006 standard.

All the three fiber treatments reduced the long-term water absorption of the composite samples containing CBS fibers. The coupling agent and the two GB treatments were equally effective in reducing the water absorption of composite containing CBS by 24-26%. Fiber treatment with compatibilizer, coupling agent and guayule at 7.5% were all very effective in increasing the flexural modulus of elasticity significantly. All four treatments also increased the flexural strength of the composites.

This study has shown that the guayule bagasse, an agricultural processing waste stream, is equally effective as the expensive chemical additives such as coupling agent and compatibilizer in reducing water absorption and enhancing flexural properties of thermoplastic composites containing CBS by 25% of weight.

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THE INCORPORATION OF WASTE STREAM AGRICULTURAL BY-PRODUCTS IN THE DEVELOPMENT OF THERMOPLASTIC BIOCOMPOSITES

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In the last several years the use of biobased materials as fillers in thermoplastics has seen a remarkable increase. The desire to produce lighter, stronger, and more ecologically friendly materials are the key driving forces behind the movement. While the focus has thus far been on producing biobased composites out of commodity polyolefins and harvested fibers such as hemp, there is some merit in utilizing waste stream products from such operations as ethanol production as fillers. These new biocomposites show much promise in a wide variety of industrial applications.

This study looks at the incorporation of several fillers from commodity processing waste streams into polypropylene, a high-volume commodity thermoplastic. Testing was carried out on the biocomposites blends to determine the resulting effects of the filler on mechanical properties. Additional thermo-mechanical and thermo-rheological tests were also performed to determine material heat stability and processability.

Fillers were acquired from commodity processors and reduced in nominal size through the use of grinders. A process called torrefaction was also used to prepare some of the fillers in a preliminary study leading to the use of bio-fillers in high-temperature applications. As polypropylene lacks good interfacial bonding with natural fibers, the use of a maleated chemical modifier (MAPP) was used to improve the matrix-fiber bond. Biocomposite blends were melt compounded in a co-rotating twin-screw extruder and then injection molded into test specimens. Tensile testing was carried out in accordance with ASTM D638. Flexural testing was also performed under ASTM D790, Procedure A. Notched Izod impact testing was conducted, as prescribed by ASTM D256. Melt Flow Index (MFI) of each compound was determined according to ASTM D1238. Heat distortion temperatures (HDT) were found under the guidelines of ASTM D648.

The use of the chemical modifier was shown to increase the ultimate tensile strength of the biocomposites over those without the modifier, approaching the strength of the neat matrix. With some further investigation, the ultimate tensile strength could reach or surpass the neat matrix with the proper fiber loading. The filler also produced an increase in elastic modulus and flexural performance. These improvements do however come at the cost of impact resistance, which drops below the performance of the neat matrix. The filler also increased the HDT and decreased the MFI of the material.

Viable biocomposites have been produced from the incorporation of waste stream agricultural by-products into polypropylene. The successful addition of torrefied filler opens the door to future work with high-temperature thermoplastics. These new composites have various industrial applications from handles to engine compartment components.

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EVALUATION AND OPTIMIZATION OF BIOFUEL PRODUCTION FROM SUNFLOWER HULLS

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Sunflower is a widely adapted crop and is now grown in every temperate region. In the U.S., 2 million acres were grown in 2010, about 75 percent of which was oilseed sunflower. This results in large amount of sunflower hulls during the industrial processing. Sunflower hulls have little commercial value and decompose very slowly. Sunflower hulls are rich in 5-carbon (C-5) and c-carbon (C-6) sugars that are ideal for the production of advanced biofuels. The objective of this project is to evaluate and optimize the biofuel production from sunflower hulls as a non-food resource and thus improve the local economy and reduce the dependence of our nation on foreign sources of energy. This project is divided into two tasks 1) pretreatment and 2) enzymatic saccharification to evaluate the sunflower hull as a potential biofuel feedstock.

Extractives such as nitrates/nitrites, waxes, protein, ash, chlorophyll were removed through exhaustive extraction process. The components of sunflower hulls are glucan (39.16 wt%), lignin (25 wt%), xylose (23.42 wt%) and traces of galactan, arabinan and ash. The sunflower hulls pretreatment was evaluated by response surface design methodology. Three factors were considered for the pretreatment: 1) Acid concentration were 0.5, 1.25 and 2% (w/w) 2) reaction temperatures were 140,150 and 160 °C 3) reaction times were 10, 20 and 30 minutes. The pretreatment was performed in

steam jacketed batch reactor. Pretreated samples were vacuum filtered and separated into solid and liquid fractions. Solid fraction samples were air dried for 4-5 days and analyzed for carbohydrates and lignin in the biomass. Liquid fraction samples were analyzed for monomeric, total sugars and inhibitors using HPLC. Enzymatic saccharification was conducted with solid fraction samples to get fermentable sugars. Accellerase 1500 (manufactured by Genecor) were used as an enzyme for enzymatic hydrolysis. The enzyme saccharification conditions were 20 mg enzyme/g cellulose, 50°C, 250 rpm for 72 hours.

All the results were evaluated according to the combined severity factor (CSF). The results showed that increase in CSF increased the amount of monomeric and total sugars liberated in the liquid fraction with little or negligible oligomeric sugars.

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KENAF: A RENEWABLE SOURCE FOR “GREEN” CHEMICALS AND FUELS IN NORTH DAKOTA

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Kenaf (*Hibiscus cannabinus* L.) is a warm-season annual. Kenaf fibers are used for paper pulp and cordage, but it is also an interesting lignocellulosic feedstock for bioenergy. The objectives of this study were to: 1) determine the best plant density and fertilization of kenaf to maximize biomass and ethanol yield; 2) evaluate the potential of kenaf as a dual crop used for summer supplemental forage and as a biomass.

Three field studies, plant density, nitrogen fertility, and harvest date and frequency were conducted at Fargo and Prosper, ND in 2010. Plant density treatments included two in-between row spacing (30 and 60 cm), four plant density sub treatments (32, 16, 8, 4 plants/m²). N fertility treatments were of a non-fertilized check treatment and N treatments of 75, 100, 150, and 200 kg N ha⁻¹ (soil N + fertilizer N). Evaluations included nitrogen content of tissue at two phenological stages: 8-leaf stage and at flowering, plant height, stem diameter, biomass yield, and conversion to ethanol. The harvest frequency experiment had three treatments: a two-cut system (19 July and 20 Sept), one harvest in 10 August, and one harvest in the fall (20 Sept). Experimental units were 5-m long and 3-m wide. Plant height, stem diameter, biomass yield, and conversion to ethanol were evaluated. Ethanol yield was calculated using the conversion factor of 113 mg ethanol g⁻¹ of dry biomass.

As plant density increased biomass and estimated ethanol yield increased. Maximum biomass and estimated ethanol yield was obtained with the two highest plant densities of 16 and 32 plants m⁻². Maximum biomass yield was 11.8 Mg ha⁻¹ and 1688 L ha⁻¹ or 143 L of ethanol per Mg of dry matter.

Kenaf had a positive response to nitrogen for biomass and ethanol yield. Crops with potential as feedstock's for bioproducts must be efficient in the use of nitrogen, an expensive and non-renewable energy input. Maximum biomass yield was 12.6 Mg of dry matter ha⁻¹ and 1800 L ethanol ha⁻¹. Although kenaf responded positively to increasing rates of nitrogen, as most crops do, the biomass yield was 8.8 Mg ha⁻¹ with no fertilizer application. Stem diameter increased linearly as the N rate increased. Nitrogen deficient plants had thinner stems which could increase plant lodging.

The harvest frequency experiment indicated the highest dry matter yield was obtained with only one harvest in the fall. The yield in this experiment was the highest for all experiments indicating the great potential of this crop as a feedstock for cellulosic biofuel and bioproducts.

Ethanol yield was also highest with only one harvest at the end in the fall. The maximum biomass yield obtained in the experiments conducted at Fargo and Prosper, ND, 2010, was 17.1 Mg ha⁻¹, equivalent to 2440 L ha⁻¹ of ethanol annually. Kenaf has a tremendous potential as a cellulosic feedstock for biofuel and bioproducts in North Dakota.

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PRETREATMENT AND ENZYMATIC HYDROLYSIS OF KENAF BIOMASS AS A RENEWABLE SOURCE FOR BIOFUELS AND GREEN CHEMICALS

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The effect of dilute acid pretreatment on biomass from corn (*Zea mays* L.) stover and switchgrass (*Panicum virgatum* L.) is very well studied. However, there are other potential biomass crops in which pretreatment have not been studied. Kenaf (*Hibiscus cannabinus* L.) has myriad of applications. It is a fiber crop mainly used in the pulp and paper industry across the globe. It can grow up to 3 to 4 m tall. Kenaf can yield approximately around 6 to 12 Mg ha⁻¹ of dry biomass. The main objective of this project was to increase the yield of fermentable sugars in kenaf biomass after enzymatic hydrolysis and also find an optimum condition for the pretreatment.

Raw kenaf was pulverized using Wiley mill to an average particle size of about 0.1 to 0.5 mm. Kenaf major components are glucan (42.5 wt%), lignin (17 wt%), xylan (16.2 wt%) etc. Glucan content in kenaf is higher compared to corn stover. The pretreatment of kenaf has been investigated using response surface methodology. Kenaf was pretreated with dilute sulfuric acid at 0.5, 1.25, and 2% (w/w). Reaction temperatures were 150, 155, and 160°C and reaction times were 10, 20, and 30 minutes. The pretreatment was performed using steam jacketed batch reactor. The reactor is made of hastelloy C276 which is more resistant to corrosion at high acid concentration and high temperature environment. The pretreated slurry samples were vacuumed filtered and separated into liquid and solid fractions. Liquid fractions were analyzed for monomeric sugars, total sugars, and inhibitors using Agilent 1200 series HPLC. Solid fractions were air dried for 4 to 5 days at room temperature and measured for carbohydrates and lignin. Enzymatic saccharification was performed on solid fraction of the pretreated biomass. Acellerase 1500 enzyme (supplied by Genecor) was used in enzymatic hydrolysis process. Enzymatic hydrolysis was carried out with 20 mg of protein/ml enzyme loading for 72 hours at 50°C in an incubator. The liquid fraction was analyzed in HPLC for amount of cellulose digested after the enzymatic saccharification.

The results showed a significant reduction in oligomeric sugars liberated in the liquid fraction at higher acid concentrations during the pretreatment. The conversion of cellulose into fermentable sugars showed a positive trend with increase of acid concentration. At 0.5% (w/w) acid concentration the yields were very low. The maximum cellulose to glucose yield was 87% (w/w). The optimum conditions were at 0.2% (w/w) acid concentration, 155 °C, and 10 minutes reaction time. A higher acid concentration leads to the degradation of glucose to hydroxymethylfurfural and xylose to furfural, but not very significantly. From the data obtained kenaf can be used as a potential resource for producing renewable fuels.

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NITROGEN UPTAKE AND BIOMASS AND ETHANOL YIELD OF FORAGE CROPS AS FEEDSTOCK FOR BIOFUEL

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Ethanol production is projected to increase with the utilization of forage crops for biomass production. Nitrogen fertilizers are extensively used to enhance the growth of biomass crops, but the current trend towards greater sustainability in agriculture is leading towards increased research to reduce the use of fertilizers. This study was conducted to determine the forage crop that produces the most biomass that can be utilized for bioenergy and to determine the effect of different levels of nitrogen fertilizer on biomass yield and nitrogen uptake in the different forage crops.

The effects of nitrogen fertilization on the nitrogen uptake and the biomass yield of forage sorghum [*Sorghum bicolor* L. Moench], sudangrass [*Sorghum sudanense* (Piper.)], reed canarygrass [*Phalaris arundinacea* L.], and kenaf [*Hibiscus cannabinus* L.] were investigated at the North Dakota State University research sites in Fargo and Prosper, ND. The experimental design in both locations was a randomized complete block design with a split-plot arrangement with three replicates. The different crops constitute the main plots and the nitrogen rates were regarded as subplots. The independent variables in this study were the four biomass crops and the five levels of N fertilization (0, 75, 100, 150, and 200 kg N ha⁻¹). Soil samples were taken for analysis in the spring before the crop was planted and in the fall immediately following harvest. Soil samples from both 0 to 15 cm and 15 to 60 cm were taken from each subplot and analyzed for N, P, K, OM, and pH. Aboveground portions of whole plants were collected at three developmental stages: vegetative (V8), flowering (VF), and right before harvest for plant NO₃-N determination. Biomass yield, forage quality, estimated ethanol yield, and nitrogen uptake were recorded. An economic analysis was conducted to determine the most economic rate of N fertilizer to obtain the greatest yield.

In 2010, the plant NO₃-N levels increased for all crops as N rates increased. Forage sorghum had the greatest dry matter biomass yield (21.28 Mg ha⁻¹) whereas reed canarygrass had the least biomass yield (8.35 Mg ha⁻¹), though reed canarygrass had the greatest N uptake (18.73 kg N ha⁻¹). Estimated ethanol yield was highest for forage sorghum (3381 L ha⁻¹) at the 150 kg N rate. The results indicate that forage sorghum has greatest potential as a lignocellulosic feedstock for biofuels.

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BIOMASS CROPS PRODUCTIVITY AFFECTED BY COVER CROPS

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Forage crops have gained interest as a potential source of biomass. As a biomass source however, more focus should be on cropping systems which include annual cover crops, to increase their productivity. This study was done to identify the agronomic of five different cover crops on four different annual biomass crops, as a source of lignocellulosic feedstock to produce ethanol. The experiment was conducted according to a randomized complete block design with a split-plot arrangement where the cover crop was in the main plot and the biomass crop in the sub-plot with three replicates. All five cover crop species were planted 9 August 2010. Just before the expected hard frost (October 14 according to previous data), foliar samples were collected to evaluate forage yield and quality. Results across locations indicated that forage pea (*Pisum sativum* L. cv. Arvika) had the highest dry matter yield (3238.3 kg ha⁻¹) followed by forage turnip Pasja [hybrid forage brassica (forage turnip X forage rape)], forage radish (*Raphanus sativus* var. niger cv. Daikon) and purple top turnip (*Brassica rapa* var. rapa). There was no significant difference in yield among the above mentioned cover crops. Further, forage pea N uptake was 126 kg N ha⁻¹ and forage radish, forage turnip and purple top turnips N uptake was 65 to 68 kg N ha⁻¹. This indicates that forages peas fixed about 60 kg of N ha⁻¹ in only 40 days in the fall. In the spring of 2011, four different biomass crops were grown and their biomass yield and forage quality parameters will be analyzed. From those crops forage sorghum (*Sorghum bicolor* L. Moench) is expected to have the highest biomass yield and forage barley (*Hordeum vulgare* L.) will be highest in forage quality.

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MECHANICAL HARVESTING OF CATTAILS FOR BIOMASS

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Cattails (*Typha latifolia*) have the potential to be an excellent source for biofuel due to its composition, ease of production, and abundance across North America. Organizations in North Dakota have been exploring the possibilities of producing ethanol from cattail.

The objective of this study is to investigate the feasibility of harvesting cattails in North Dakota and to explore the possible mechanical harvesting device to cut and collect the cattail plant materials.

Cattails grow in muddy wet soil up to a depth of 20 inches in water, though harvesting is likely to be less than this depth. Matured cattails have a feathery seed on the head that is easily blown by wind. Moist soil condition and possible buildup of seeds on the air intake of the power unit during harvesting was considered in designing the mechanical harvesting device. Three different possible designs were studied.

The three designs studied were: 1) Mounting a cutting bar to the rear of a tractor with a three-point hitch, similar to a sickle mower, 2) Cutting head mounted on the front of a skid steer loader with high flotation tracks and 3) A bio-baler for harvesting woody stemmed plants with a front mounted mulching head to cut and bail in a single pass. All the concepts were analyzed with engineering design principles.

A single pass bio bailer might be a possible harvesting device based on its weight, cost, efficiency, risks and safety factor. The design would also depend on availability of information on cattails' ethanol content, its yield, and transportation to nearest processing plant.

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Natural Rubber and Resins

PRODUCTION OF CROP-BASED MEDICAL AND INDUSTRIAL POLYMERS AND FUELS: FROM BENCHTOP TO BIOREFINERY

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As global population continues to increase, and fossil fuel resources become depleted, or impose an intolerable, environmental burden on our planet, efforts are underway to move sustainable biorefineries, powered with renewable fuels, from a subsidized concept to commercial reality. Biorefineries currently tend to produce a very limited array of materials or fuels, not a wide range as seen from petroleum refineries. Although efforts to convert biomass to biofuel via either enzymatic or thermochemical processes will continue to contribute towards energy independence, this process alone is not enough to achieve target goals. Instead, a combination of products and fuels, from opportunity feedstocks, is needed for profitability. Thus, the production of rubber, resins, bioproducts and fuels from guayule, or rubber, inulin and fuels from Russian Dandelion, are both attractive to a true biorefinery. However, as both are new industrial crops, scale-up issues are extremely complex. The expansion of acreage needs to be intimately tied to expansion of local processing capacity and to geographically-specific markets. Expansion must be accomplished in the face of limited resources, and the traversing of multiple "valleys of death" is required before sustainable establishment can be achieved. However, at the current time, increasing demand is causing global shortfalls in rubber and resin supplies and is creating reinvigorated interest from government and industry and new opportunities for researchers.

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TOWARD A STANDARD GUAYULE RUBBER

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Quality standards for guayule rubber (GR) are derived from those for natural rubber (NR), yet the chemical and physical properties of GR can differ from those of both NR and its synthetic analogs (IR). These differences include *cis*-1,4 content, gel content, non-rubber extractables (resin) content, protein content, bulk viscosity, oxidation stability, and cure chemistry.

The resin in GR acts as a plasticizer, reducing bulk viscosity. The low protein content of solvent-extracted GR decreases gel, reduces oxidative stability, and affects cure chemistry. Resin and low gel tend to reduce mixing energy. Lower levels of non-rubber components such as proteins can reduce the efficiency of sulfur cross-link formation. These particular characteristics have an impact on the performance of GR in commercial applications. As a result, GR has not been a universal replacement for NR.

Could standards specific to GR assure a commercially-acceptable product? Answering that question requires both an understanding of how shrub production and processing influences rubber composition and an evaluation of rubber performance in finished goods.

Aircraft tires were built from 100% GR. The dynamic test performance — a most important specification - of GR tires did not match that of NR tires even though the GR used in fabrication had met all applicable NR standards. Light truck tires were built from 100% GR and 50:50 blends of GR and NR. The proprietary wire skim and tread stocks used in these tires had no significant deficiencies in properties. There were deviations from standard performance, most particularly in ply-to-ply adhesion and tear strength, but it was anticipated that adjustments in cure package composition would overcome these relatively minor deficiencies. At the conclusion of testing, the finished tires were found to have met performance specifications.

How could some GR tires meet performance specifications when specific tire stocks did not? Proprietary cure systems are designed to permit some batch-to-batch optimization. Most importantly, tires are multi-component systems. It is the performance of the tire system that determines utility.

How can information such as this be used to determine the best standards for GR? Production and process development can provide GR which meets certain standards of certain physical and chemical properties. Subsequent testing - end-user evaluation in specific rubber compositions - can establish the limits of this material's utility. Rubber quality could then be re-optimized by appropriate modifications in production and processing operations. After an appropriate number of iterations, the end result could well be a commercial polymer with its own set of specifications.

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UTILIZATION OF PLOIDY ANALYSES IN A GUAYULE BREEDING PROGRAM

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The domestication and cultivation of guayule (*Parthenium argentatum* Gray) as a perennial natural rubber crop has been intermittent in the southwestern United States, thus its continued genetic improvement through modern plant breeding is vitally needed to realize yield potential and suitability for commercialization. Natural differences in ploidy levels among individuals are one of several factors that have slowed the rate of genetic gain in guayule breeding programs, thus knowledge of ploidy levels in guayule germplasm would help to accelerate the development of stable,

high yielding cultivars. The objectives of our studies were to develop flow cytometry as a rapid and simple alternative to laborious chromosome counting in guayule and to evaluate the ploidy level in various guayule populations. Utilizing this technique we have evaluated two sets of greenhouse grown guayule accessions available from the National Plant Germplasm System for ploidy level. We have also evaluated three sets of field grown populations. The first was a set of lines developed by genetic modification of AZ-2, the second was an open pollinated breeding population developed from a series of diploid crosses, and the third was another set of guayule accessions from the National Plant Germplasm System. In the greenhouse and field materials obtained from the national Plant Germplasm System, a natural polyploid series that ranged from diploids ($2n=2x=36$) to pentaploids ($2n=5x=90$) was detected, with $4x$ as the predominant ploidy level. In addition, accessions with multiple ploidy levels (i.e., mixed ploidy) were observed, which suggests that the ploidy level of a guayule plant should not be taken for granted. There is also some evidence that utilizing seed from open pollinated sources results in mixed ploidy levels in the progeny. All of the plants tested following genetic modification of AZ-2 were tetraploid as is the AZ-2 parent. Plants tested from the open pollinated diploid population exhibited similar results to those of the guayule accessions from the National Plant Germplasm System with a range of ploidy levels from diploid to pentaploid identified. Notably, linkage of our ploidy level data to that of pedigrees uncovered complex ploidy level variation in guayule breeding programs, which was found to be perfectly concordant with existing ploidy level data supported by chromosome counting. Importantly, this work serves as the basis for future breeding efforts as well as QTL analysis, association mapping studies in guayule, and genome size determination.

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AGRONOMIC EVALUATION OF GUAYULE CULTIVATION IN TWO NORTHERN MEDITERRANEAN AREAS

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The Northern Mediterranean areas of Europe have an annual total rainfall and annual mean temperatures equivalent to those recorded in the Southern U.S. and Northern Mexico, but the distribution of rainfall in the Northern Mediterranean region is different. In Europe, rainfalls are more abundant in winter, while in the region of origin, they are more abundant in summer.

The objective of this study, done within the European EU-PEARLS project, was to determine the feasibility of guayule (*Parthenium argentatum*, Gray) cultivation in these regions.

A germplasm and a fertilization and irrigation trial were planted in 2008 and 2009 in two sites: one in a northern part of the Mediterranean area (Montpellier, South of France) and one further south (Cartagena, South of Spain).

In Montpellier (France), the best cultivars for biomass production were AZ 5, AZ 6, AZ 101, AZ 3, 11619, giving an average of 100 kg of dry weight biomass per plant and per year for stems and branches. Nevertheless, best percentages of rubber in the stems and branches were found for other cultivars; 11693 (7.4%), N 565 (7.0%), and N 566 (5.8%) one year after planting; other cultivars had less than 5% of rubber content. Finally, AZ 3, AZ 5, and AZ 1 had the best rubber yields. Irrigation had a significant weakening effect on biomass yields. The high mortality of the plants after the 2009 winter (over 60%) showed that Montpellier is not currently adapted for guayule cultivation, and can be considered as the northernmost limit for guayule growing in northern Mediterranean region of Europe with current germplasm.

In Cartagena (Spain), the % rubber ranged from 4.08 to 9.34 %, with N 396 and 11604 having more than 9% (stems and branches). The best yields were recorded for 11701, N 575, R1100, 11693. More than 15 cultivars produced more than 200 g dry biomass per plant (equivalent to 10 tons per hectare) per year (stems plus branches). Irrigation had a significant positive effect on yields. With a potential production of more than 700 kg of rubber per hectare per year in the fully irrigated plots, we can predict that the South Spain area is fully adapted for guayule cultivation, provided sufficient watering.

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HERBICIDE TOLERANCE OF RUSSIAN DANDELION

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Natural rubber is classified as a strategic material due to its use in manufacturing a wide array of modern industrial products. It is required in high-performance applications such as aircraft tires, yet the United States imports nearly 100% from tropical sources (the Brazilian rubber tree- *Hevea brasiliensis*). Five Asian countries control about 90% of world rubber exports. In the 1930s, Soviet scientists identified the roots of Russian dandelion (*Taraxacum kok-saghyz* or TKS), a native of central Asia, as a promising source of natural rubber that could be grown as an annual crop in temperate climates. Testing in the US during WWII demonstrated wide adaptation of this crop, including a very promising site in Oregon's Klamath Basin. Recent studies have confirmed this potential, yet information on effective weed control methods is lacking. TKS is not initially vigorous after germination, and thus developing weed control strategies including use of herbicides would be desirable.

The objective of these preliminary studies was to screen likely pre-plant incorporated (PPI), pre-emergence (PRE) and post-emergence (POST) herbicides to find compounds that are tolerated well by TKS and thus developing an herbicide-based weed control program.

A series of replicated herbicide applications were made to TKS at various growth stages, in some case at two rates for a given herbicide, in greenhouse pot and field situations, using natural field soil. TKS germination and herbicide damage were rated compared to control (untreated) plots. Root biomass and rubber content were not measured.

In a greenhouse pot study, TKS seeds germinated very well and exhibited good tolerance to ethalfluralin and trifluralin applied PPI, and to s-metolachlor, pendimethalin, pronamide, and sulfentrazone applied PRE. TKS exhibited fairly good germination and moderate damage from EPTC applied PPI, and to flumetsulam and imazethapyr applied PRE, suggesting some potential of these compounds, but with further study needed to confirm their suitability for TKS. Damage ranged from moderately high to essentially 100% fatal for ethofumesate, clomazone, dimethenamid-P, imazamox, mesotrione, norflurazon, and terbacil.

In a field study using some of the same compounds, TKS exhibited good tolerance to pendimethalin, s-metolachlor, and the lower rate of sulfentrazone applied PRE, although TKS injury increased dramatically at 36 days after treatment compared to 18 days after treatment for pendimethalin. TKS damage was moderate to high for ethalfluralin and triflurain applied PPI and for the high rate of sulfentrazone applied PRE.

To test tolerance to common grass-selective herbicides, existing TKS plants (approx. 15 cm height) received POST applications of either quizalofop or clethodim, each applied at two rates. TKS exhibited no damage from these grass-selective herbicides.

A number of PPI, PRE, and POST herbicides appear to have promise in a Russian dandelion weed control program. Further studies are necessary to identify optimum rates and to pursue required labeling before commercial use can occur.

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PROPERTIES OF NATURAL FIBER/SOY-BASED POLYURETHANE BIOCOMPOSITES

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Natural fibers are increasingly used in the construction of composite materials, but are often used in petroleum

based polymer matrices. A more renewable composite can be created by using both natural fibers and a bio-based polymer matrix. One potential bio-based matrix is soy-based polyurethane (PU), which consists of approximately ~30% renewable feedstock. Use of these new materials requires testing to understand the structure-process-property relationships for these biocomposites. The interfacial properties of the biocomposites are the keys for the mechanical performance of the composites. The hydrophilicity of PU matrix can lead to good interfacial bonding between matrix and cellulosic fibers. However, it is difficult to estimate the interfacial bonding between the randomly oriented fiber mats and PU matrix.

The objective of this study was to determine the basic mechanical properties of biocomposites based on foamed and non-foamed PU matrix structures, as well as determine their interfacial properties.

To test the basic mechanical properties of the biocomposites, panels were compression molded using non-woven mats made from flax, hemp, and kenaf fiber. These fibers were placed in both a foamed and non-foamed PU matrices. To examine the interfacial properties, unidirectional Chinese flax fiber is chosen as the reinforcement for the soy-based PU. The unidirectional composite panels were compression molded with untreated (as-received) flax, NaOH-treated flax, and PU matrix.

Tension and flexural properties were evaluated according to ASTM standards. The interlaminar shear strengths of the composites were evaluated by short beam shear tests and the interfacial shear strengths of the composites are measured by fiber bundle pull-out tests.

The unidirectional flax/PU composites exhibited better mechanical performance than the randomly oriented mat composites. The main different in performance is due to the variation in architecture between unidirectional fiber and mat, however, the lower non-cellulose content in unidirectional flax compared to the than fiber mats also seems to play a role.

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PRETREATMENT OF GUAYULE BIOMASS FOR IMPROVED BIOCONVERSION EFFICIENCY

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Parthenium argentatum, guayule, is a woody desert crop under cultivation in the southwest United States as a source of natural rubber, organic resins, and biofuel.

Natural rubber and resin extraction from guayule yields a high energy, high density, finely-ground biomass feedstock suitable for conversion to bioenergy or biofuels. Enzymatic hydrolysis of guayule biomass requires pretreatment for efficient conversion to glucose and xylose sugars. The objective of this work was to evaluate the efficacy of ammonia fiber expansion (AFEX) and organosolv pretreatments for improved conversion efficiency in guayule shrub, before and after rubber and resin extraction. Guayule shrub was ground and extracted with acetone, to remove resins, then cyclohexane, to remove rubber. Latex-extracted bagasse was also evaluated. Compositional analysis, enzymatic hydrolysis, and fermentation to ethanol were carried out using standard protocols.

The glucan and xylan content of the samples varied between ~ 15-30% and ~ 10-18%, respectively. Overall, compositional analysis results were similar to previous reports. Guayule bagasse has significantly higher glucan and xylan content, likely due to the removal of leaves prior to processing. About one-third by weight of the biomass is composed of lignin, as expected for a hardwood plant.

In this study, untreated guayule shrub and bagasse was indeed recalcitrant to enzymatic hydrolysis. AFEX pretreatment substantially improved overall enzymatic digestibility by 4-20 fold for both untreated guayule shrub and rubber-extracted bagasse. Maximum glucan and xylan conversion achieved for AFEX-treated bagasse was 40% and 50%,

respectively. Organosolv pretreatment at high temperatures with sulfuric acid catalyst yielded up to 99% glucose conversion. In either case, yeast was readily able to ferment the glucose and/or xylose to ethanol from the guayule bagasse hydrolyzate. There were no indications of detrimental levels of fermentation inhibitors specific to guayule. Biomass pretreatment options enhance the potential for guayule as a feedstock for lignocellulosic refineries co-producing biobased products and biofuels.

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General Crops and Products

WATER MANAGEMENT TO INCREASE AND STABILIZE CROP PRODUCTION IN NORTH DAKOTA AND NORTHWESTERN MINNESOTA

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Tile drainage or sub-surface drainage is a practice that was introduced to the United States in 1835. Traditionally, clay tiles were buried in the ground to drain the excess water off of farm land, thus the name tiles. Currently, plastic tubing with holes are buried in the ground to achieve the same effect. Even though tile drainage is a common farming method in areas like the Corn Belt, it is a new technology for the Red River Valley and the clay soils in North Dakota and Northwestern Minnesota.

The past 10-15 years have been abnormally wet and waterlogging has been the main factor in yield loss during that time period. Tile drainage can alleviate water stress to crops and can increase the carrying capacity of the soil so that heavy equipment can access the field for crop management in a timely manner.

Prevented planting acreage, due to wet soil conditions in ND in 2011, are estimated to be 6 million acres. The increased rainfall also caused salinity to become a problem due to rising water tables. Salinity in the Red River Valley encompasses over 1.5 million acres. Tile drainage is a potential way to control and reduce salinity in wet soils. Producers have observed significant yield responses to tile drainage using combine yield monitors. University research also indicates benefits of tile drainage.

Farmers report that their tiled fields are now those where field operations take place first, instead of last. However, flat topography, tight soils, economic uncertainty, and a tradition of surface drainage only, still limit its more widespread adoption of tile drainage in the region.

In the flat areas control structures can be installed to control the water table and providing producers with a tool to manage one more production factor. With the option of water management it is expected that yields, including canola, will increase and that year to year fluctuation in yield levels will decrease. As yields increase so will also the total biomass produced, providing options to utilize residues for biofuel production.

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EVALUATION OF FENUGREEK (*TRIGONELLA FOENUM-GRAECUM* L.) AS NEW CROP IN WESTERN NEBRASKA

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NOT PRESENTED

Fenugreek (*Trigonella foenum-graecum* L.) is an annual legume and known to possess a numbers of medicinal properties (anti-diabetic, anti-cholesterol and anti- cancer). This medicinal property is mostly because of several health-enhancing bioactive compounds (diosgenin, galactomannan, and 4-hydroxy isoleucine) present in matured seed. It is reported to produce high yields of bloat-free forage that can increase both beef and milk production in semiarid regions of western Canada. It is reported to grow well under dryland and limited water condition. Therefore, fenugreek may be a suitable new forage and industrially (nutraceutical) important crop to rotate with winter wheat in western Nebraska, which is characterized by limited water, dry climate, and short-growing season. The objectives were (1) to evaluate fenugreek germplasm for regional adaptation and seed as well forage yield potential, and (2) to study genetic diversity at genome level based on molecular markers. Germplasm available at USDA-National Plant Germplasm System and publicly available varieties were grown in the field. Common agronomic characteristics (plant stand, flowering, seed maturity, plant height, seed yield) were measured. These lines were also genotyped for molecular markers (ISSR, and SSR). Here, we will report extent of variation in the morpho-agronomic traits and DNA marker-based genetic relationship among the germplasm set tested. We expect significant genetic variability for biomass production and seed yield among the genotypes. This set of germplasm will serve as base for the development of cultivars suitable for both forage and seed production in the western Nebraska and neighboring region.

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OPTIMAL STAND DENSITY FOR ETHANOL PRODUCTION FROM ARIZONA-GROWN SWEET SORGHUM

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We have previously shown that sweet sorghum (*Sorghum bicolor* (L.) Moench) can be grown in the arid/semi-arid southwest United States with fewer inputs than commonly used for biofuel crops from other parts of the country (e.g., corn grown in the Midwest US). In order to maximize yields of sugar and biomass (previously shown to be the best predictors of ethanol yield), and provide specific information for growers, a two-year study of various stand densities was performed.

In 2009 and 2010, three commercial varieties (Cowley, M81E, and Topper) were each planted at five densities (2.5, 4, 6.7, 10, and 20 plants per meter). All treatments of each line were harvested at maturity; in addition, juice samples were analyzed for sugars (glucose, fructose, and sucrose) on a semi-weekly basis.

In both years the more plants per area, the lighter the weight of the stalks pressed for juice. These plants also had smaller stem diameters, and in 2009 contained less juice. Juice weights in 2010 showed more uniformity, possible due to slightly more rainfall in that year. In 2009, stems of M81E had significantly smaller diameters than the other two varieties, but these stalks had higher sugar concentrations, averaging 652.4 gL⁻¹ and with the highest at 775.7 gL⁻¹. Sugar values for Cowley were generally in the middle, while Topper was consistently on the bottom. Plants in the densest treatment were tallest with smaller stem diameters.

In both years, all three varieties displayed similar trends in sugar accumulation. Very little glucose and fructose are present up until late July (around 80 days after planting); less than 10 gL⁻¹ of each. Sucrose concentration is below 5 gL⁻¹ until the end of August (about 120 days after planting), when a rapid increase up to and in excess of 100gL⁻¹ occurs at the final harvest in November (at physiological maturity, almost 200 days after planting). While there were differences in many measured characteristics at the different plant densities, the plants responded morphologically and physiologically in such a way that there were no significant differences in predicted ethanol yield. However, we felt that the lower plant densities (2.5 and 4 plants per meter) would fit better in the semi-arid agricultural system of the southwestern US because we found that when plants are too close together, competition for resources produces thinner stalks with less juice, and these stalks have a higher frequency of lodging (falling over).

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Medicinal and Nutraceuticals

SCUTELLARIA OCMULGEE SMALL: AN ENDANGERED PLANT WITH LIMITED DISTRIBUTION BUT UNLIMITED POTENTIALS

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Medicinal plants play a central role as traditional medicines used in many cultures and also as trade commodities which meet the demand of often distant markets. Recent research trends in medicinal plants show that while ethnobotanical surveys continue, there is an appreciable increase in research activity in the area of bioactivity of natural products. As many as 84% of pediatric oncology patients, 50% of breast cancer patients and 37% of prostate cancer patients use complementary and alternative medicine (CAM), including predominantly herbal approaches (Richardson, 2001). *Scutellaria* is a perennial, herbaceous genus in the Lamiaceae family with 350 - 400 species. This genus is well adapted to the North American climate being represented by about 90 species. *Scutellaria* species have been used in many biomedical studies and their anticancer, antiviral, anti-inflammatory, antibacterial, anti-allergenic, and antioxidant properties have been summarized in a recent review article (Li and Khan, 2006).

We initiated *Scutellaria* research at Fort Valley State University in the year 2001 and realized that some of the species are extremely rare. Our main objectives are to carry out regular germplasm collection and phytochemical analysis, ex situ conservation and cryopreservation, study of anti – tumor properties using cell lines and animal model, micropropagation and genetic transformation, flowering, ornamental potential and reproductive biology, field studies to study the potential of *Scutellaria* as a short term premium crop, and hairy root culture experiments to study secondary metabolite synthesis.

Results obtained in the last ten years and areas of research that have unfolded in front of us will be presented.

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CONSUMPTION OF GROUND WHOLESEED FLAX AND HUMAN BLOOD TRAITS

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Flaxseed has been used for centuries as a source of food for humans and animals, and its oil has been used for nutraceutical purposes due to its high content of health beneficial components. Anecdotal information suggests that consumption of wholeseed flax as a food supplement could be beneficial for human health. Flaxseed contain about 42 percent oil, approximately 70 percent of this oil is polyunsaturated fat which is considered healthy for human nutrition. Oilseed flax has been recognized as a functional food in recent times. Functional foods deliver a health boost beyond what is expected from their traditional nutrient content. Flax fits this description perfectly because it is rich in omega-3 fatty acid and phytochemicals. Flax also provides dietary fiber and protein. We conducted an experiment in which approximately 45 g of ground wholeseed flax was consumed by 26 faculty and staff volunteers from Virginia State University for 12 weeks during February – April of 2009. A blood sample before start of consumption and one after completion of the experiment was collected by a commercial medical firm and the blood from both samples was analyzed

for various traits. Results indicated that consumption of wholeseed flax significantly increased carbon dioxide (24.23 vs. 23.03 mmol/L), lowered total serum protein (7.17 vs. 7.02 d/dl) and also lowered total Globulin (2.99 vs. 2.78 g/dl) in the blood. Flax consumption did not affect cholesterol content of blood. Further details of these results will be presented and discussed.

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USING CONTROLLED WATER STRESS TO IMPROVE NUTRIENT QUALITY IN QUINOA SEED

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Many crops and native species grow in areas where they are exposed to many types of abiotic stress, especially water deficit at certain times of the year. It has been reported that several environmental stress factors during plant growth can regulate the accumulation of secondary metabolites and other nutraceutical substances. The response to these factors depends, among others, on the species, stage of development, metabolic status of the plant, as well as on the duration of stress. The economic value that these bioactive substances have acquired because of their benefits to human health, as well as the revaluation of ancient crops such as quinoa (*Chenopodium quinoa* Willd.) make it necessary to improve the quality of the raw material, without reducing yield potential.

This study aimed to determine the effect of controlled water restriction on some chemical components and antioxidant content in seeds of three genotypes of quinoa and its effect on the potential yield. The study was conducted in Chillán, Chile (36° 35'43, 2" S, 72 ° 04'39, 9" W and 140 m elevation) under field conditions, outdoors and under controlled conditions in a greenhouse. The experimental design was a randomized complete block with a split-plot arrangement. Main plot treatments were three irrigation treatments where the water applied was 100%, 60%, and 40% of field capacity throughout the growing season. The subplots included three quinoa genotypes with different levels of improvement. The antioxidant characteristics of quinoa seeds, some nutritional parameters, and seed yield were evaluated.

Results indicate that water restriction should be applied once 50% of the grains are in the grain filling stage, that is, from milk to dough stages. In addition, an increase in the antioxidant capacity was observed in seeds exposed to greater restriction (40% of field capacity), without reducing their yield potential strongly. It was concluded that water stress applied at certain phenological stages of water deficit-tolerant crops, can produce food in a controlled manner, with a better nutritional value, and without reducing seed yield.

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NATIVE VASCULAR PLANTS OF AGRONOMICAL IMPORTANCE OF THE AYSÉN REGION, CHILEAN PATAGONIA

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The Cisnes River basin (5,144 km²) is located in the Aysén Region, Chilean Patagonia. The river is approximately 160 km long and the basin is only 35 km wide in the north-south orientation. Different types of climates can be observed: on the upper sector of the basin. There is a gradual transition from cold steppe to rainy temperate without a dry season (on the central basin), and rainy maritime climate near the estuary. The rainfall decreases from west to east, from ~ 2,000 mm to 600 mm per year. The annual temperatures vary around 9°C. During the winter the temperatures descend below 0°C, especially on the upper sector of the basin with snow. This area has been botanically much less studied than the central zone of Chile. Nevertheless, many native species that grow in this location could have agronomical importance as medicinal plants, forage, tinctorial or ornamental.

The objective was to collect plants along the valley of the Cisnes River, from Amengual Village to the border with Argentina in order to document the uses that local communities give to the native flora.

Plants were collected on 4-7 January, 2011 along the valley of the Cisnes River from Amengual Village (44°44'S, 72°12'W, 324 m), passing La Tapera Village (44°37'S, 71°38'W), Río Cisnes Cattle Ranch (44°29'S, 71°18'W, 907 m) to Frías River-Appelg Pass (44°33'S, 71°06'W, 925 m) on the border with Argentina. Locations were annotated and photos of plants were taken. Dried specimens were kept at the Herbarium of the Universidad de Concepción (CONC-CH) and included in the CONC database for a future checklist of Aysen vascular flora. A bibliographic research of potential uses of the collected species was done and local communities were surveyed.

Three hundred and thirty two herbarium specimens were collected. According to the present or potential uses of them, four categories were found: 1. Medicinal plants: *Apium australe*, *Anthoxanthum odoratum*, *Bromus hordeaceus*, *Chusquea culeou*, *Dactylis glomerata*, *Embothrium coccineum*, *Fragaria chiloensis*, *Fuchsia magellanica*, *Gunnera tinctoria*, *Gaultheria phillyreifolia*, *Nothofagus dombeyi*, *N. pumilio*, *Weinmannia trichosperma*; 2. Food plants: *Apium australe*, *C. culeou*, *F. chiloensis*, *F. magellanica*, *G. tinctoria*, *G. phillyreifolia*; 3. Forage plants: *Alopecurus geniculatus* var. *patagonicus*, *Bromus setifolius* var. *brevifolius*, *Festuca pallescens*; 4. Dyeing plants: *F. magellanica*, *G. tinctoria*, *N. dombeyi*.

This is a first effort to record current or potential uses of the native flora in this region. It is possible to deduce other categories of uses as well as the specific way in which the local communities use these species.

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ENHANCING SEED GERMINATION OF *PARIS POLYPHYLLA* VAR. *YUNNANENSIS* (FRANCH.) HAND.-MAZZ

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Paris polyphylla var. *yunnanensis*, naturally grown in the deciduous forest in the temperate and subtropic region of Yunnan Province in China, is the most utilized plant for pharmaceutical purpose. Its rhizome is the main source of several famous Chinese medicines used for controlling bleeding and alleviating stasis, swell, and pain. *P. polyphylla* var. *yunnanensis* is naturally seed-propagated and its seeds have a long period of dormancy (16 months) with a low germination rate. The objective of this research was to find ways to break seed dormancy and increase the germination rate. In this study, seeds were treated with seven fungal suspensions (fungi were isolated from the rhizosphere soil where the plants were grown), four nutritional solutions, two plant growth regulators, and two chemicals. Seed germination was monitored. In addition, the enzyme activity, the content of steroidal saponins and other active compounds in seeds during the germination were detected.

Seed structure: seed is oval. The testa is red or orange, juicy and succulent. At the beginning the germination, the embryo was observed as an eight-cell ellipsoid structure surrounded by the tightly arranged endosperm cells. The embryo has not fully differentiated. That might be the main reason causing the long seed dormancy and low germination rate. One fungal treatment (MM1) significantly enhanced seed germination with an average of 73.33% germination rate and 0.15 germination index at 0.05 (P < 0.05). Seeds treated with "A" nutritional solution (full-strength nutritional solution)

had the highest germination rate (80%) and germination index (0.16). In the PGR and chemical treatments, the seed germination rate was 76.67%, 83.33%, 76.67%, and 70% and the germination index was 0.15, 0.17, 0.15, and 0.14 when treated with GA₃, 6-BA, ZnSO₄, and PEG, respectively. During seed germination, protein, starch, crude fat was declining; however, soluble sugar was increasing in the first 90 days, and then decreasing. Saponin existed in the testa and seed. In the testa, both saponin VII and saponin II were detected, but in the seed, only small amount of saponin VII was found. The testa contained 0.2 % more saponin VII than the seed.

This research showed that undifferentiated embryo is the main cause of seed dormancy. Treatment of certain fungal, nutritional, PGR, or chemical solutions can shorten the dormancy and increase the germination rate.

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EFFECT OF NATURAL WAX NUTRACEUTICAL COVERS WITH ANTIOXIDANTS IN AVOCADO SHELF LIFE

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At the present time, exists an elevate demand of high quality products containing only natural ingredients due to their good sensorial and nutritional properties. Edibles coatings are transparent films that cover foods against humidity and oxygen. The conservation mechanism consist in create a barrier that works as a modified atmosphere around the product and also acts as a physical obstruction against microorganisms.

The objective of the present work was to study the effect of natural wax nutraceutical covers with antioxidant compounds applied in avocado for extended shelf life time.

Edibles coatings of *Euphorbia antisiphilitica* Zucc. (candelilla), were elaborated with natural wax mixed with natural antioxidants compounds. The tested antioxidant compounds were ellagic and gallic acids, obtained by fungal fermentation. The blanks tested were: a) covers without antioxidants, and b) fruit without cover. Quality physicochemical parameters of avocado shelf life were evaluated.

The obtained results showed that candelilla wax covers with ellagic acid improved the shelf life quality in avocado fruit.

Edibles coatings of candelilla with ellagic acid represent an interesting alternative in order to maintain fruits quality and its nutritional values in post-harvest storage conditions.

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JUNEBERRY CULTIVAR EVALUATION IN NORTH DAKOTA

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In our society today, diet is considered an important factor in the prevention and management of many major diseases. Fruits are known to contain antioxidants that reduce the risk of diseases. Juneberry (*Amelanchier alnifolia* Nutt.), is a shrub that bears a sweet edible pome which is dark blue in color. Juneberry fruits have been found to have

high contents of flavonols, phenolics and anthocyanins, among other elements. Compared with blueberries, strawberries, and raspberries, juneberries are known to have the highest nutritional value. The entire plant was used by Plains Indian tribes in concoctions made from the roots and inner bark to cure a number of diseases as well. However, it is hardly known and less cultivated in North Dakota even though it can successfully grow in this climate.

Juneberry has great potential and must be closely looked at for industrialization. In Saskatchewan, Canada, a successfully established fruit processing sector exists where juneberry fruits are processed into jams, jellies, sauces, frozen fruit, dried fruit, and teas. The ND variety trial was to evaluate fruit quality and yield of the cultivated juneberry to aid with commercialization in North Dakota.

Eleven cultivars were planted in Absaraka in a replicated field trial. Data were collected for analysis. Plant height and plant width were taken of the different cultivars. Berries were separately picked from two samples of the same cultivar, if still alive and bearing fruit. All berries were taken from a plant and separated into marketable and non-marketable, and weighed. Berry diameter and sugar content was determined for each cultivar.

The best cultivar was identified through the various parameters that were measured. The size of the plant and berries, together with uniformity berry maturity and sugar content were determined to evaluate productivity among the selected cultivars.

The many benefits of juneberry can be utilized, starting with maximum cultivar potential. Future research will focus on nutritional differences within the cultivars so that, together, the commercial potential of juneberry can be demonstrated.

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POSTER PRESENTATIONS (Fibers and Cellulosics)

COMPOSITE MATERIALS DERIVED FROM BIOMASS

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Replacing petroleum-based materials with biomass derived materials possessing similar properties and functions is an answer to the pressing concern of oil depletion and environmental degradation. To this end, we have developed a range of materials suitable for different applications from biomass. Wood plastic composites, sugar beet pulp composites and foams, soy protein composites, foams and fibers, and cellulose nanowhisker composites are a few examples of the biomass derived materials we have developed. This presentation gives a brief introduction to the processing, properties, and applications of these materials.

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EMPIRICAL MODELING AS AN EXPERIMENTAL APPROACH TO EVALUATE SIMULTANEOUS SACCHARIFICATION AND WHEAT STRAW FERMENTATION FOR BIOETHANOL PRODUCTION

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In simultaneous saccharification and fermentation (SSF) of lignocellulosic materials, a rapid conversion to ethanol of the produced glucose is expected. The principal benefits are to minimize enzyme sugar inhibition, to improve cellulose conversion rates and to reduce cost compared with separated hydrolysis and fermentation, due to the occurring synergy between enzyme-yeast-substrate in one bioreactor. Wheat straw (WS) is an abundant by-product from worldwide wheat production making it an important substrate for bioethanol production.

The aim of this work was to evaluate the effect of temperature, substrate concentration and loading enzyme on bioethanol production by SSF having as substrate WS pretreated by autohydrolysis (AH) and using flocculating *Saccharomyces cerevisiae* CA11.

A 2³ central composite design was applied and the limits of the different parameters were: 30-45°C; 2-3 % of substrate; 5-30 FPU of cellulose enzyme (Celluclast 1.5) per g dry substrate and 30-60 of β-glucosidase (Novozym 188) IU per g substrate. Ethanol production, residual glucose, and cellobiose were analyzed by HPLC. CO₂ was kinetically monitored by weight loss in Erlenmeyer flasks.

Results showed that after 60 h of fermentation the highest ethanol concentration – 14.84 g/l (with a corresponding CO₂ value of 14.27 g/l) was obtained at 45°C, 3% of substrate, 30 FPU and 60 IU. This value, corresponding to an ethanol yield of 84.2%, shows a low enzyme inhibition during SSF process as the glucose produced by enzymatic hydrolysis is rapidly assimilated for yeast cells.

Overall, it may be concluded that WS pretreated by AH is a good substrate for SSF process as high substrate conversion into ethanol can be achieved as a result of the synergy between enzyme-yeast-substrate.

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POSTER PRESENTATIONS (General Crops)

TOMATILLO: IT'S ORGANIC GREENHOUSE CULTIVATION AT NUEVO LEON, MEXICO

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Mexico holds 11,759 hectares of protected agriculture, reaching for 2010 a volume around 3.5 million tons of annual production. The growth has been observed to be 1,200 hectares per year. At the present time, Mexico is placed on the third place worldwide for the number of organic producers and has around 450 thousand hectares dedicated to organic production. The tomatillo is known (in Spanish) as: green tomato, de cascara, de bolsa, and tomate fresadilla. It is of very high economic importance since it is considered the fifth most planted vegetable in Mexico. It has a market as a fresh domestic and export vegetable, as well as industrial processes, mainly as a green sauce, puree, and whole. This research was aimed at evaluating two tomatillo cultivars under organic conditions in a greenhouse during the cycle in August 2010 to January 2011, in the state of Nuevo Leon, Mexico

The evaluation was conducted in a greenhouse type bitunnel GC model 3000 in the facilities of the Center for Applied Agriculture at ITESM, located in Hualahuises, Nuevo Leon. There were two cultivars of tomatillo: Gran Esmeralda and Gigante. Greenhouse transplant was performed August 18, 2010; under plantation were 1.60 x 0.50 m, single row (two plants per square meter, with a density of 12,500 plants per hectare). The “useful parcel” was 6.4 square meters. The organic production package included a biofertilizer called “vermicompost”, product that consists of Red Worm of California humus, used 3 ton/ha; an organic biofertilizer consisting of beneficial bacteria from the soil called

Bacillus subtilis and fungi from the ground called mycorrhizae. Plagues were controlled with a bioinsecticide derived from Neem tree, cinnamon extracts, garlic and soy, and the use of soapy substances. For the prevention of sicknesses, the fungi *Trichoderma* and agricultural iodides that help plants metabolism were used and the sicknesses were controlled with the use of organic fungicides.

The production showed no statistical differences, it was similar for both cultivars: Gran Esmeralda averaged 61.3 ton/ha, while Gigante 60.5 ton/ha. The first survey was done at 74 days after the transplant, both materials were considered early. One of the more traditional varieties that are planted nationwide is “rendidora”, yet its performance is very erratic and low, as in open field production this variety has achieved 8 to 12 ton/ha. Given the wild nature of the tomatillo, there has been a significant change in open field crops to intensive greenhouse conditions and organic packages that ensure high yields and fruit quality. Due to the high productivity of the tomatillo obtained at this time, Nuevo Leon can produce it in controlled environments and organically, which can be a real agribusiness opportunity in the autumn-winter season for export to North America.

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ADAPTATION SCREENING FOR NEW PULSES IN NORTH DAKOTA

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The continually rising world’s population will require increased food, feed, fiber, and fuel to meet mankind’s needs in the future. Diversifying food crops in production regions will help in this regard and could also benefit agricultural sustainability, depending on the crop, crop rotation, and region. Pulses are high in protein and fiber, and low in fat, which makes them an ideal human dietary component. Pulses complement crop rotations by disrupting pest cycles and fix nitrogen: both of which help improve sustainability.

The objective of this study was to identify potential pulses for adaptation in North Dakota and the surrounding region, to expand crop diversity and benefit agricultural sustainability.

The experiment was conducted in North Dakota at two locations: the Carrington Research and Extension Center and the Prosper off-station site. The experimental design was a RCBD with four replicates and pulses as treatments. At Carrington, the study included twelve pulses: adzuki (*Vigna angularis* (Willd.) Ohwi & H. Ohashi), cowpea (*Vigna unguiculata* (L.), dry bean (*Phaseolus vulgaris* L.), faba bean (*Vicia faba* L.), field pea (*Pisum sativum* L.), lupine (*Lupinus albus* L.), mung bean (*Vigna radiata* (L.) R. Wilczek), natto bean (*Glycine max* (L.) Merr.), otebo bean (*Phaseolus vulgaris* L.), peanut (*Arachis hypogaea* L.), pigeonpea (*Cajanus cajan* (L.) Mill sp.), and tepary bean (*Phaseolus acutifolius* A. Gray). The study at Prosper also included fenugreek (*Trigonella Foenum-graecum* L.) and lima bean (*Phaseolus lunatus* L.). Field pea and dry bean are traditional crops in the region and were included as checks for comparison with the new pulses. Plots consisted of six rows, spaced 30 cm apart and 6.1m in length. Traits for evaluation included: days to emergence, plant stand, days to flowering, plant height and lodging, seed yield, seed weight, and days to physiological and harvest maturity.

Results for stand establishment indicated a pulse and pulse by location interaction. Pulses could be grouped into low, medium, and high groups for stand establishment. Stand establishment was low for cowpea and pigeonpea, medium for adzuki, faba bean, field pea, mung bean, natto, otebo, peanut, and tepary, and high for dry bean and lupine. Ranking differences among the pulses for stand establishment were observed at the two locations. Most of the new pulses produced stands comparable to the adapted check crops.

Good stand establishment provides a competitive leaf canopy and is a prerequisite to high crop performance during the growing season. The new pulses, except for cowpea and pigeonpea, produced adequate stands for good crop performance, provided their maturity isn’t limited by the region’s relatively short growing season.

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EVALUATION OF A PHYSICAL MODEL FOR WEATHER SIMULATION ON A GREENHOUSE WITH NATURAL VENTILATION

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In Mexico the use of greenhouses have been increased in the last years; natural ventilation are a useful tool in places with warm weather because allow to decrease damages caused by high temperatures with economical and energetic efficiency. However in order to evaluate and control the ventilation effect over different climatic conditions, greenhouse characteristics and the studied crops it is necessary to apply simulation studies on physics-based models.

The aim of the present work was to validate a physic-based model simulation centered in the study of greenhouse weather using a simplified heat balance over tomato crops.

Validation study was carried out in Galeana, Nuevo León, Mexico in 2009 and 2010. Natural ventilation data was recorded in a 4 hectares greenhouse of tomato crop (*Solanum lycopersicum* L.). The developed equations were based in the prediction of temperature and relative humidity. The produced data were collected using an automatic climate station in the greenhouse exterior (temperature, relative humidity, solar radiation, and wind velocity) and interior (temperature and relative humidity).

The predictive equations showed values of $R^2 = 0.96$ and 0.89 in 2009 and 2010, respectively for temperature; and $R^2 = 0.91$ and 0.75 in 2009 and 2010, respectively for relative humidity, with 95% of confidence interval. Model limitations were observed in cool hours, specifically above 10°C .

Statistical analysis showed that the developed model allowed the prediction of the greenhouse weather without the necessity of calibration.

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EVALUATION AND CHARACTERIZATION OF BROWN SEAWEED SULFATED FUCANS EXTRACTED UNDER ENVIRONMENTAL FRIENDLY TECHNOLOGIES

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Brown seaweeds are the most important source of non animal sulfated polysaccharides, which comprise a complex group of macromolecules with important biological properties such as anticoagulant, antioxidant, antiproliferative, antitumoral, anticomplementary, anti-inflammatory, antiviral, antipeptic, and antiadhesive activities. Fucoidan is one of the main sulfated fucan, being of great interest due to its biological activities specially the potential to inhibit HIV reverse transcriptase and the possible application as active compound in antiretroviral drugs. Usually, most of the processes to recover sulfated polysaccharides from natural sources consist in acid extractions during long reaction times (1-3 h).

The objective of the present work was to evaluate the extraction of sulfated polysaccharides (fucoidan) from brown seaweeds comparing hydrothermal process based on microwave radiation and high pressurized heating.

Fucus vesiculosus brown seaweed from North Portugal was used in the experiments. The extraction reactions

were carried out under a full factorial design varying time, pressure/temperature, and alga/water ratio.

The percentage of fucoidan recovered was highest with 120 psi, 1 min, and 1 g alga/25 ml water under microwave system and with 180°C, 20 min and 1 g alga/25 ml water under high pressurized heating. Sulfate content in the hydrolysates was not affected by the operational conditions, maintaining values higher than 18%. Monosaccharide composition analyses showed mainly the presence of fucose, galactose and xylose

In conclusion, hydrothermal extractions, as a green technology, showed to be an effective method for sulfated polysaccharides recovery from brown seaweeds with shorter times than those reported in the literature.

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POSTER PRESENTATIONS (Oilseeds)

SYNTHESIS AND PHYSICAL PROPERTIES OF PENNYCRESS ESTOLIDE 2-ETHYLHEXYL ESTERS

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Pennycress (*Thlaspi arvense* L.) is a new crop that is currently developed as an off-season rotation crop between annual corn and soybean production in Central Illinois by USDA-NCAUR. This rotation scheme may offer distinct advantages to farmers by providing additional farm income from an otherwise fallow season with little impact on the subsequent soybean production. The seed contains up to 36% oil with the major fatty acid as erucic (38.1%). The fatty acid composition of pennycress has been shown to have physical properties suitable for biofuels like hydro-treated renewable jet fuel (HRJ). Like many developing new crops, many aspects of pennycress growth, development, potential products, and economic success have yet to be fully characterized. The development of a new crop often depends on the synthesis of novel “green” compounds. Estolides are one such derivative of new crop oils which show promise in industrial applications.

Estolides are formed when the carboxylic acid functionality of one fatty acid links to the site of unsaturation of another fatty acid to form esters. Estolides were derived from pennycress fatty acids and various other fatty acids in the presence of an acid catalyst at 60°C for 24 hrs. The free acid estolides were then esterified to the 2-ethylhexyl esters under standard conditions.

These new estolide esters were converted to their corresponding hydroxy fatty acid and the degrees of polymerization were determined by GC analysis. Physical properties (pour points, cloud points and viscosities) of the pennycress estolide esters were compared to previously synthesized homo-estolides and coco-estolides, which have current industrial applications. These new estolides show promise as a cheap alternative bio-based material.

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EVALUATION OF RARE *HELIANTHUS EGGERTII* ACHENES FOR OIL CONCENTRATION AND FATTY ACID COMPOSITION

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Sunflower (*Helianthus annuus* L.) oil has the potential to be improved for nutritional and industrial purposes through selection and breeding. The narrow genetic base of cultivated sunflower has been broadened by the infusion of genes from wild species, resulting in a continuous improvement in agronomic traits. Interest in using wild species in breeding programs has increased, but information about oil concentration and fatty acid composition is lacking for a number of rare and threatened species.

The objective of this study was to evaluate achenes of a rare wild perennial hexaploid, *H. eggertii* (Eggert's sunflower), from the southeastern USA for oil concentration and fatty acid composition of four major fatty acids, palmitic, stearic, oleic, and linoleic acids.

Thirteen populations of Eggert's sunflower were collected throughout the broad distributional range of the species. Fatty acid composition was determined using gas chromatography on oil extracted from two 10-achene samples for each population converted to methyl esters using an organic catalyzed transesterification method. Oil concentration was determined using nuclear magnetic resonance.

Helianthus eggertii had an average oil concentration of 288 g/kg, which was within the range expected for a wild perennial sunflower species. The average linoleic acid concentration was 728 g/kg, ranging from 582 to 772 g/kg. Linoleic acid concentration was higher than expected for populations grown in southern latitudes. The saturated palmitic and stearic fatty acids in *H. eggertii* averaged 84 g/kg, which is about 30% less than typical cultivated sunflower oil with approximately 120 g/kg.

The lower saturated fatty acid profile and the higher linoleic concentration in the oil of *H. eggertii* indicates that this species has the potential to reduce saturated fatty acids and increase linoleic acid concentration in oil of traditional commercial sunflower grown at southern latitudes. Further research will be needed to determine the inheritance of the fatty acids and oil concentration. Other agronomic traits will need to be maintained during the introgression of these traits into cultivated sunflower.

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EFFECTS OF PLANTING DEPTH ON FIELD ESTABLISHMENT OF PENNYCRESS AND LIGHT CONDITIONS ON SEED GERMINATION

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Pennycress (*Thlaspi arvense*), is a promising oilseed (36% oil) with potential for biofuels and another industrial uses. A winter annual, it may be feasible for use in Midwestern double cropping systems. However, agronomic and biological issues should be studied in order to understand and overcome practical limitations. The objectives of this study were to determine:

- i) the effect of planting depth on emergence and establishment of *T. arvense* accessions, and
- ii) light requirements for germination of *T. arvense*.

The experiment to evaluate the effect of planting depth was conducted in Ames, IA, at the North Central Regional Plant Introduction in a replicated split-plot design, with planting depth treatments as main plot effects. Eight *Thlaspi* accessions were evaluated at two planting depths, 0.5 inches, and surface planting. Flowering and maturation dates, number of plants established, oil concentration, and yield were evaluated.

The planting depth experimental results indicate that depth does impact pennycress germination and establishment, and differences were observed for stands counts and yield. However, the magnitude of impact varies between accessions, with a range of responses.

In order to determine whether the accessions have different light requirements for germination, an experiment was

conducted in 20°C germination chambers under light and dark conditions. Three accessions in were evaluated in a replicated design, and germination was scored every three days. The germination experiment indicated accessions are variable in their ability to germinate under dark conditions, where the reduction in germination in dark compared to light germination varies from 30% for the less sensitive accessions to more than 90% for the more sensitive accessions.

In conclusion, germination of some *Thlaspi* accessions are sensitive to dark conditions; probably due to lack of light limiting phytochrome activation that are responsible, necessary for photo-activation of seed germination. Further experiments should be conducted using pre-treatments to activate the phytochrome. While it may be possible to improve establishment rates under shallow planting depth conditions, producers need reliable production guidance for appropriate planting depth, and germplasm that will germinate readily if field conditions at planting result in minor deviations from intended depth.

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SEED OIL DEVELOPMENT OF PENNYCRESS UNDER FIELD CONDITIONS

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Pennycress (*Thlaspi sp*), has been targeted as a potential oilseed for biofuels industry. Its seeds contain ~36% oil, where erucic acid is the mayor fatty acid presented with 38.1 %. Additionally, the physical proprieties of the methyl esters are in the range to satisfy the needs of the biodiesel market. This species has also shown novel biofumigant characteristics from meal of their seeds (volatile compounds). Due the valuable characteristics (spring and winter types, cold tolerance, early harvesting, fatty acids profile, etc.), pennycress could be used as a crop in the Midwest. However, agronomic issues should be studied to overcome practical limitations.

Harvest must occur when seeds are mature and oil content has reached its maximum value. The objective of study was to determine how the rate of oil accumulation during the last stages of seed development of *Thlaspi arvense* accessions may vary, and to apply this knowledge to make wise harvest decisions.

The samples were harvested from a 2010 fall planting experiment at the PI-USDA-ISU in Ames, Iowa. Three accessions were selected due to their different maturity rates. Each accession was sampled six times prior to seed maturity and harvest; an early maturity accession was sampled five times instead of six, at intervals of four days until harvest time.

The results will be analyzed to test the hypothesis that rates of oil development vary between accessions, and to determine whether the maximum concentration rate is obtained immediately prior to seed physiological maturity, and the time course of moisture content change.

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THLASPI ARVENSE (PENNYCRESS) GERMINATION, BOLTING, AND MECHANICAL HARVEST SEED LOSS

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Pennycress is being developed as an off-season rotation crop between annual corn and soybean production. This rotation scheme may offer distinct advantages to farmers by providing additional farm income from an otherwise fallow season with little impact on the subsequent soybean production. The seed contains up to 36% oil with the major fatty acid as erucic (38.1%). The fatty acid composition in *thlaspi* has been shown to have physical properties suitable for biofuels like biodiesel and hydro-treated renewable jet fuel (HRJ). Like many developing new crops, many aspects of pennycress growth and development have yet to be fully characterized.

A standard germination protocol was established for evaluating the germination rates of *thlaspi* germplasm. Ideal conditions for germination were found to be 12 hour photoperiod with a diurnal temperature variation of 27°C daylight and 11.5°C dark. Germination rates were determined at 14 days with initial germination of most of the seeds occurring by 7 days. Using these germination conditions, a long term seed storage study was undertaken. Freshly hand harvested seed from 2009-2010 plots of the Beecher line were placed in a number of storage conditions; sealed containers, breathable containers, near constant temperature and fully exposed to outdoor temperature fluctuations. Germination rates were determined at 1 month intervals up to 6 months then at 3 month intervals thereafter. Seeds stored in the freezer failed to germinate at any time during the study. Seeds exposed to outdoor temperature variations developed the best germination rates early in the study but the rates quickly diminished upon exposure to cold winter temperatures. Seed stored near constant room temperature developed the best germination with the highest rates observed (85%) after one year of storage.

A set of experiments were conducted in the growth chamber to determine the impact of temperature and day-length on bolting within pennycress. The experiments demonstrated that changing day-length from 12 hours to 7 hours then back to 16 hours at constant temperature did not induce bolting. Conversely, under constant 12 h day-length all the plants bolted after exposure to temperatures near 0°C for 14 days followed by several weeks of temperatures at 15°C.

One tenth acre field plots with densities ranging from 14K to 270K plants per acre were established and allowed to reach full maturity. Immediately prior to combine harvest, 10 weigh boats with a total area of 1.74 ft² were placed in the field. Immediately after the combine harvested the standing crop, the weigh boats were collected and seed loss due to mechanical harvest was evaluated. Seed loss ranged from 27 to 97 lbs/acre with the highest densities yielding the highest loss rates. A qualitative evaluation of the plots prior to harvest indicated that more seed was dispersed on the ground than was collected in the weigh boats after mechanical harvesting indicating that weather induced shattering could be a problem.

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WINTER CANOLA IS A POTENTIAL SOURCE TO DEVELOP HIGH PERFORMANCE SPRING CANOLA GERMPLASM

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Canola is a strategic crop of great importance to North Dakota, is the largest producer of canola in the USA. Increasing seed yield in canola is an important breeding objective for the breeders to satisfy the demand of the growers and seed processing industries. On the basis of growing season, rapeseed (*B. napus*) is classified into two groups, such as annual type and biennial type. The annual type is also known as spring type and biennial type is known as winter type. The yield potentiality of winter type rapeseed is over two-fold compared to spring type canola. Because of severe winter hardiness, winter type canola is not possible to grow in North Dakota.

The objective of this study is to develop high seed yield spring canola utilizing winter type rapeseed.

Crosses were made between winter (cv. ARC-97108) and spring (cv. Regent) type canola. The winter parent ARC-97018 was vernalized to get flower in the vernalization chamber of the North Dakota State University. The F₁, F₂, and backcross progenies were grown in the greenhouse of the North Dakota State University. Data on day to flower, plant height, number of branches/plant, number of pods/plant, pod length, root length, dry stem weight, dry root weight, and seed yield/plant were taken to study the correlation of coefficient among the characters using SAS 9.1 software (SAS Institute Inc., Cary, NC, USA).

Vernalization requirement to flower has been identified as a recessive trait. Two genes loci are responsible for the

vernalization requirement of *B. napus*. Significant positive correlations were found between branches vs. number of pod (0.58***), branches vs. dry stem weight (0.38*), branches vs. seed yield (0.49**), number of pods vs. root length (0.41*), number of pods vs. dry stem weight (0.43*), root length vs. dry root weight (0.46*), root length vs. seed yield (0.37*), dry stem weight vs. seed yield (0.54**).

Two recessive genes are responsible for vernalization in *B. napus*. Plants with vigorous root system showed a positive correlation with number of pods and seed yield.

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THE EFFECT OF NITROGEN RATE ON FIELD PENNYCRESS YIELD AND OIL CONTENT

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Field pennycress (*Thlaspi arvense* L.) is a new potential oilseed crop that is currently being evaluated as a domestic source of biodiesel fuel. The research based information about pennycress nitrogen requirements will help producers apply optimal nitrogen rate and minimize production costs.

This study was initiated to evaluate the effect of nitrogen fertilization rate on pennycress seed and biomass yield, yield components, plant height, and oil content.

A growth chamber experiment was conducted on the spring annual pennycress breeding line '32' from Montana, with six nitrogen rates (0, 25, 50, 75, 100 and 125 lbs. nitrogen per acre) using urea fertilizer (46-0-0). Additional field experiments evaluating the influence of nitrogen rates on the seed yield and quality of the pennycress breeding lines 'Beecher' and 'W-12' were established as well.

The greatest seed and biomass yields of 0.32 and 0.48 g per plant respectively, were achieved with 50 lbs. of nitrogen per acre in a growth chamber environment. Greater nitrogen rates did not result in further seed and biomass yields increasing. A rate of 50 lbs. of nitrogen also resulted in the greatest pods number per plant. The greatest seed number per plant was achieved with 75 lbs. of nitrogen per acre. Height of the plants that received the rate of 25 lbs. of nitrogen per acre was higher than that of non-fertilized control but did not differ from height achieved with increased nitrogen rates. Nitrogen fertilization rates had no effect on the oil content in the seed. This information will be further evaluated in the field experiments with pennycress populations from different geographical regions.

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DETERMINING THE ROLE OF DAY LENGTH AND TEMPERATURE ON THE VERNALIZATION OF FIELD PENNYCRESS (*THLASPI ARVENSE* L.)

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The amount of time required from one generation to the next is a critical factor in the ability of a plant breeder to improve the genetics of a plant species in a timely manner. Field Pennycress (*Thlaspi arvense* L.) is a winter annual weed species that is currently being studied as a potential new source of industrial products and biodiesel.

The objective of this study was to investigate the role of day length and temperature in overcoming the biannual nature of field pennycress. The first experiment investigated cold (4°C) and warm (21°C) temperatures with short (10 h) and long day (16 h) lengths for 30 days. The second experiment focused on the amount of time required in cold temperatures to induce flowering. All plants were germinated under long day length and warm conditions before being

moved to either warm or cold temperatures with varying day lengths. Treatments lasting 3 to 27 days indicated cold temperature was the dominant component necessary for vernalization, however day length also played a small role in the time of vernalization to the onset of flowering. Plants responded to the cold treatment as early as the cotyledon stage. Cotyledon stage seedlings needed a minimum of 16 days of exposure to cold (4°C) temperatures in order to successfully initiate flowering once returned to warm temperatures under long day length.

The development of a rapid flowering protocol in growth chambers will greatly improve the advancement of winter annual field pennycress as a potential new crop in the Midwest.

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QUALITY OF FIELD PENNYCRESS OIL OBTAINED BY SCREW PRESSING AND SOLVENT EXTRACTION

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Field pennycress (*Thlaspi arvense* L., *Brassicaceae*) is a winter annual that grows widely in the temperate North America. Its seeds contain up to 36% oil (db) with the major fatty acid being erucic acid (38%). With an estimated seed production of 1,700 – 2,200 kg/ha, pennycress can be a major source of oil for biofuel. The early harvest date of pennycress compared to other winter annual oilseed crops makes it suitable for a two-crop rotation with soybeans in most of the Midwestern United States. This allows for the production of oil for industrial application without displacing commodity crops for food use.

The 2009 harvest of about 1,500 kg of pennycress seeds from a native stand near Peoria, Illinois provided our first opportunity to conduct oil extraction studies. The goal was to evaluate the quality of the oil obtained by full pressing and pre-pressing followed by solvent extraction. The starting seed had 9.5 % moisture content (MC) and 31.4% oil (dry basis, db). Cooked and uncooked seeds (60 kg) were screw pressed using a heavy duty French Laboratory Screw Press (Model L250). Seed cooking was done using a steam-heated 3-deck French Seed Cooker/Conditioner (Model 324). The seeds were heated to 82°C and held at this temperature for 20 min. The press cakes (350 g/batch) from cooked and cold-pressed seeds were extracted with hexane using a soxhlet apparatus. Oil was recovered from the miscella using a rotary evaporator. The press cakes and hexane defatted meals were analyzed for oil and moisture contents. The oils were analyzed for free fatty acid (FFA), phosphatides (measured as phosphorus [P]), and sulfur (S) contents.

Cold pressing pennycress seeds with 9.5% MC produced press cake with 10.7% oil (db), extracting 74% of the oil. Cooking reduced the seed MC to 5.3% and improved the extraction yield by 19%. Hexane-defatted press cakes had residual oil content of $\leq 0.03\%$, increasing the overall oil yield to 99.9%. Oil from cold-pressed seeds had the lowest FFA, P, and S contents at 0.23%, 1 ppm, and 20 ppm, respectively. Cooking the seeds before pressing increased the oil's FFA to 0.39%, P to 67 ppm, and S to 373 ppm. Hexane co-extracted significantly more phosphatides and sulfur compounds with the oil from the press cakes. Compared to hexane-extracted oil from cold-pressed cake, the oil from cooked seed press cake had higher P (740 vs. 506 ppm) and similar S (172 vs 162 ppm). Phosphatides and sulfur compounds are known to poison catalysts. Therefore, the degree of oil refining needed to bring the oil to the specification of an intended application depends on the method of oil extraction. Phosphatides are easily removed by degumming while sulfur compounds are only partially removed in degumming, caustic refining, bleaching, and deodorization.

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CAMELINA (*CAMELINA SATIVA* L.) SEED YIELD RESPONSE TO NITROGEN, SULFUR, AND PHOSPHORUS FERTILIZER IN SOUTH CENTRAL CHILE

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Camelina (*Camelina sativa* L.), Brassicaceae, is a new oilseed crop, with potential as a low cost feedstock for biodiesel for the North Central United States. The objective of this study was to evaluate seed and oil yield response of camelina to nitrogen (N), phosphorus (P), and sulfur (S) nutrition in South Central Chile. Experiments were conducted in Chillan, El Carmen, Los Ángeles, Gorbea, and Osorno, Chile, in 2008 and 2009. The experiment at Chillan, Los Angeles, and Gorbea was a RCBD, with 4 replicates, in a factorial arrangement of 4 rates of N (0, 75, 150 and 300 kg N ha⁻¹) and two rates of S (0 and 40 kg S ha⁻¹). The experiment at El Carmen and Osorno was also a RCBD with 4 replicates in a factorial arrangement where, in addition to the 4 rates of N, and 2 rates of S, three rates of P (0, 50, 100 kg P₂O₅ ha⁻¹) were included. Seed yield was taken from the center-four rows of each experimental unit. Seed yield components evaluated were i.) number of plants m⁻², ii.) number of silicles plant⁻¹, iii.) number of seeds silicles⁻¹, and iv.) 1000-seed weight. Seed oil content was determined on a 40 mL seed sample, from each experimental unit, with a Newport Nuclear Magnetic Resonance (NMR) analyzer. Also the N content in plant tissue and seed, N uptake, N use efficiency, N agronomic efficiency, and N physiologic efficiency were evaluated. Besides, the S content in plant tissue and seeds, and S uptake was evaluated in El Carmen, Los Ángeles, and Osorno. Each location-year combination was defined as an environment and was considered a random effect in the statistical analysis. The combined analysis of variance indicated the N rate by environment interaction was significant for seed yield. A positive seed yield response with N additions was observed at all environments except in Gorbea. Maximum seed yield was obtained with the 300 kg N ha⁻¹ rate in Los Angeles, El Carmen, and Chillan, and was 2390, 1766, and 1104 kg ha⁻¹, respectively. In Osorno, the seed yield increased to 1917 kg ha⁻¹ with the 150 kg N ha⁻¹ rate. The addition of P or S did not increase seed yield at any of the environments. Nitrogen fertilization increased N content in plant tissue and seed, seed protein content, and N uptake. The N uptake was 103.7 kg N ha⁻¹ with a rate of 300 kg N ha⁻¹ in Los Ángeles. The N use efficiency and N agronomic efficiency had significant differences as N rate increased. Both efficiencies decreased with N applications greater than 75 kg N ha⁻¹. The highest N rate applied decreased oil content from 436 to 416 g kg⁻¹ averaged all environments. Rates higher than 75 kg N ha⁻¹ caused serious plant lodging in Osorno. Camelina N requirement is less than 100 kg N ha⁻¹ lower than that of canola grown in the same area. Plant tissue S content was 0.35% with 40 kg S ha⁻¹ and 0.3% without S fertilizer. The S uptake increased significantly as the rate of N increased. Sulfur uptake was 31.6 kg S ha⁻¹ with 300 kg N ha⁻¹ and only 13 kg S ha⁻¹ without N application in Osorno.

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POSTER PRESENTATIONS (Medicinals and Nutraceuticals)

BASIL *OCIMUM BASILICUM*: IT'S ORGANIC GREENHOUSE CULTIVATION AT NUEVO LEON, MEXICO

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Mexico is considered as a powerhouse for the production of vegetables and aromatic plants for export, mainly for markets such as United States, Canada, Japan, and the European Union. Nowadays, Mexico is placed on the third place

worldwide for the number of organic producers and has around 450 thousand hectares dedicated to organic production. Basil is an aromatic plant with medicinal properties, its popularity as a seasoning is because it is widely used in Italian cuisine. In the last years, its cultivation has been increasing due to its new uses in medicine and in alimentation; naturopaths and great chefs prefer it to be grown organically. This investigation has the purpose of evaluating the cultivation of basil under organic conditions in a greenhouse during the cycle of August 2010 to May 2011 in the state of Nuevo Leon, Mexico.

The evaluation was done in a bitunnel greenhouse model GC 3000 in the installations of the Applied Agriculture Center from ITESM, in Hualahuises, Nuevo Leon. A variety of basil was established: Grande Verde (Los Molinos). The transplantation in the greenhouse was done in August 2, 2010; the plantation perimeter was 1.60 x 0.67 m in a double row. The "useful parcel" was 4 square meters. The organic production package included a biofertilizer called "vermicompost", product that consists of Red Worm of California humus, used 3 ton/ha; an organic biofertilizer consisting of beneficial bacteria from the soil called *Bacillus subtilis* and fungi from the ground called mycorrhizae. Plagues were controlled with a bioinsecticide derived from Neem tree, cinnamon extracts, garlic and soy, and the use of soapy substances. For the prevention of sicknesses, the fungi *Trichoderma* and agricultural iodides that help plants metabolism were used and the sicknesses were controlled with the use of organic fungicides.

The production was done in four cuts, which yielded results of 10.6, 37.2, 20.1, 30.3 tons per hectare. The three first cuts were done in the months of October, November and January, in the autumn-winter season, when in this region it would be very difficult to produce basil in the open field. Under the greenhouse conditions this specie had rapid growth, lush foliage and a height that ranged between 90 and 140 cm. Its leaves are opposite, of a very bright green color, elliptical between 5 and 7 cm long, exhaling a very pleasing aroma when crushed. The plant is most efficient when fresh. There is a wide market due to its aroma, taste, and medicinal properties because it contains essences rich in estragol, eugenol, and thymol. Therefore, basil represents a business opportunity for greenhouse producers in Nuevo Leon, which will increase the alternatives for diversification of crops under protected environments.

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