Performance of Five Castor (*Ricinus communis* L.) Genotypes in Geographically Diverse Environments of North America

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Abstract

Castor (*Ricinus communis* L.) is an important industrial oilseed crop, which produces the unique fatty acid, ricinoleic acid. Ricinoleic acid is used in the production of nylon, cosmetics, lubricants, and also has medicinal uses. Currently, most North American castor is imported from the world's top producers (India, China, and Brazil). From 2011 to 2013, five castor genotypes were tested for agronomic performance in 12 diverse environments across North America ranging from 29° to 46° latitude. These five genotypes were planted in a Latin square plot design with five replications and were evaluated on harvest index, seed weight, seed yield, oil content, and total oil production.

These results indicated that castor was well adapted to several diverse environments of North America and appears to be adapted to areas with shorter growing season of northern temperate regions where it produced good seed and oil yields. However, castor can be grown competitively across most of North America.

Seed yields of the five genotypes averaged over the 12 environments ranged from 1073 kg ha^{-1} for Brigham to 1291 kg ha^{-1} for Memphis. Average total oil yields of the five genotypes over the 12 environments ranged from 568 kg ha^{-1} for Energia to 480 kg ha^{-1} for Brigham. Harvest index was highest for Ultra Dwarf (22.2 % for seed yield and 13.5 % for oil yield). Analysis of variance and F-tests of genotypes, environments, and the genotype by environment showed that they were highly significant ($p = 0.001$) between all three factors for all indices except for seed yield and oil yield. Most of the genotype by environment interaction seems to be due to the superior performance of castor in the five test environments of latitudes North of 40°. The southern environments

(< 40° N) produced seeds with a higher oil content and had higher harvest indices than the more northern environments. However the northern environments produced almost double the seed yield of the southern environments resulting in more kilograms of oil per hectare.

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Chapter I

Introduction

Castor (*Ricinus communis* L.) is an indeterminate tropical plant native to eastern Africa or western Asia (Moshkin, 1986 Brigham, 1993). It is an important industrial crop, which produces the unique fatty acid, ricinoleic acid. Ricinoleic acid is used in the production of nylon, cosmetics, lubricants, and it also has medicinal uses. Currently most North American castor oil, 15 thousand metric tons, is imported annually from the world's top producers (India, China, and Brazil) (US Census Bureau, 2011). The potential of revitalizing a $15 million dollar commodity to North America after forty years has led to the necessity to identify potential genotypes and growing environments. By evaluating these genotypes in different environments, the agronomic potential can be determined based on the indices of harvest index, seed weight, seed yield, oil content, and total oil production.

Harvest index shows the efficiency of a genotype in using the available resources in an environment to produce the harvested part of the plant. In the case of castor, this represents resources used to produce seed and oil. Seed weight allows a determination on how the plant responds to different environments. Seed size also allows for estimation of seeds that will have good germination and oil content, since more mature seeds will weigh more than immature ones (Severino, 2012). Seed yield estimation determines the potential seed production based on how a genotype performs within an environment. Oil content gives an idea as to the amount of profit that can be made per hectare from a single genotype in each of the given environments.

Castor production, in the United States, which at one time had reached 30,000 ha with yields in excess of 2200 kg ha^{-1} ceased in the 1970s. Brigham (1993) gave four reasons for the cessation of castor production. First, there were low world prices for castor and second higher prices for competing crops in the United States. The third reason was a disagreement of price between producers and the oil mills and the fourth was the end of government subsidized castor crops. There has been renewed interest in castor due to its potential use as a biodiesel feedstock, since castor seed has between 38 and 60 % oil (Atsmon, 1989). Auld et al. (2009) suggested that castor could be grown on marginal lands and yet still produce high levels of oil. This would give castor an advantage over soybeans, sunflowers, and other potential feedstocks. The problem with using castor oil as a biodiesel feedstock is the high viscosity which is caused by the high levels of ricinoleic acid (>80% of methylesters) (Thomas, 2013). Wang et al. (2011) indicated that such a high viscosity would lead to problems during the spray phase of injection and it would lead to incomplete combustion in a diesel engine.

The objective of this study was to determine the best genotype for each of the 12 unique test environments across North America. Genotypes were selected that could be mechanically harvested, while still representing a wide range of plant architecture, among released or soon to be released germplasm.

Chapter II

Literature Review

Environment

Castor (*Ricinus communis* L.) is an indeterminate plant that has been grown for thousands of years. Western Asia or eastern Africa is considered the origin of cultivated castor (Moshkin, 1986 Brigham, 1993). Currently India, China, and Brazil lead world production of the crop with over 2.6 million metric tons of annual production (FAO Statistics, 2012). Castor has also been grown many different countries ranging from Canada to Argentina in the Western Hemisphere and from Russia to South Africa in the Eastern Hemisphere. Specifically in the United States, the growing environments for castor were defined by Domingo and Crooks (1945) as environments with a minimum 180 day growing season, well-drained soils, low humidity, and at least 15 inches (381 mm) of rainfall. Seed yield declines have been observed due to early frosts, drought, or disease pressures. Baldwin and Cossar (2009) suggested that there is a relationship between the latitude that a genotype is developed and how well it performs. They showed that there was a significant difference between the yields obtained in Tennessee compared with Mississippi. The more northern environments outperformed the southern environments regardless of planting date as long as the soil temperatures exceeded the recommended 15.6° C at planting. Koutroubas et al. (1999) showed there was a genotype by environment interaction of castor when it was grown at two distinct Environments in Greece. Laureti (1995) showed that there was a significant difference in plant height, seed weight, and the number of racemes per plant when there was more available water for the plants to use.

Harvest Index

Harvest Index is an estimate of the harvestable material compared to total plant biomass. Donald (1962) defined harvest index as the percent of the plant that could be sold for economic gain. According to Severino and Auld (2013) harvest index for castor should apply to both the seed and the oil percent compared to total biomass, because the oil is the final economic product of the castor plant. Castor is sold by seed weight, the fatty acid composition of the oil and quantity of the oil. Hay (1994) described the process of determining harvest index as cutting the plant at ground level and then determining the yield percent of the total biomass collected. Hay (1994) also cautions against taking harvest index too late or on samples that do not represent the plot. Castor would be a good example of a plant that the harvest index can be taken too late, since when the plant is killed with a frost, the leaves abscess from the plant altering the estimated harvest index. Castor is also an example of a plant in which care must be taken to have representative plants of the plot, because castor is very heterogeneous. Hay (1994) also suggested that a large difference in harvest index measurements would result from the plant being too green and full of water. His reasoning was that water is not evenly distributed in all parts of the plant. Hay (1994) also cautions against comparing harvest indices from different Environments due to differences in row spacing, soil fertility, plant population, and environmental conditions such as rainfall and humidity. However, it is the best factor when determining which variety should be used at a specific environment. Weiss (2000) suggested that harvest index in castor would be low, because of the high levels of protein and oil found in the seed. Severino and Auld (2013) stated that previous castor harvest indices have ranged from 14 % to 71 %. They suggested that harvest index in castor could be improved by having smaller leaves, petioles, and stems. They also suggested thinner capsules, spineless fruit, and many other factors. Hay (1994) stated that the barley cultivar “Golden Promise” had less total biomass than previous genotypes but had higher yields, because less energy went toward biomass production and more of the energy went toward seed production. Harvest index, is based on the concept that a plant should produce more of the economic parts while maintaining the other tissues of the plant at minimal levels. Morrison et al. (1999) showed that in soybeans (*Glycine max* L. Merr.) there has been an improvement in harvest index and a decrease in leaf area over the past seven decades. This shows the potential to use harvest index of castor, to improve profitability.

Seed Weight

Seed weight is an indicator of the quality of the seeds that are harvested. The heavier the individual seed weights; the more likely that the seed will germinate when it is sown the next year. Canvin (1963) showed that castor seeds have reached their maximum size two weeks after fertilization. Moshkin (1986) suggested that seed weight is important for both the germination and yield of the next year's crop. He also suggested that seed density could be used to determine differences in seed sizes of different genotypes. Seed with different density can be separated using a gravity table. Severino and Auld (2013) found that the mother plant had the largest impact on seeds during the first phase of development. During this time, it is competing with all the other seeds on the plant for photosynthates and other nutrients. They also found that in later seed development, environmental forces influenced a seed’s ability to grow and mature rather than that of the mother plant’s allocation of resources. This would lead to large environmental influence at different Environments. Moshkin (1986) determined that seed weights were higher under periods of high temperature than periods of low temperature. This adds to the idea that the environment has one of the largest impacts on final seed yield. Vallejos et al. (2011) showed that there was a significant difference in the final seed dry weight between genotypes. This may be in part due to the difference in seed size between the different genotypes. Vallejos et al. (2011) also showed that there were differences in the rate of seed filling and the length of time that the seeds were filled between the different genotypes. This factor would influence which genotypes are adapted for a particular environment. Since the shorter season environments will benefit from growing genotypes with faster seed filling times. Frey and Huang (1969) showed in oats (*Avena Sativa*) that there was positive relationship between the seed weight and the seed yield produced at the end of the season. However, genotype did affect the magnitude of the correlation between the two variables. Seed weights would be a useful breeding tool in selecting high yielding castor lines. Lakshminarayana et al. (1984) showed that even though there was a difference in seed weight between genotypes, there was not a significant difference in the total oil produced. This suggests that seed weight may be a poor breeding tool for improving the total oil yield of castor.

Seed Yield

Castor has been reported to produce between 2242 and 3363 kg ha^{-1} in the United States (Brigham, 1993). Castor seed yield was shown to be determined by the percent of female flowers on the raceme, the total number of racemes, time of seed filling, and the temperature during filling. Vallejos et al. (2011) determined that castor will flower for approximately three to four months depending on the genotype. They also showed that the genotype greatly influences the hierarchy of the racemes. A castor genotype with fewer number of racemes had a positive impact on the number of seeds produced per raceme. Hooks et al. (1970) determined that there was a significant difference in the number of racemes per plant between reciprocal crosses; it was shown that the maternal parent greatly influenced the total number of racemes per plant. Because the maternal parent influences the total number of racemes, breeders can quickly identify high yielding lines. Laureti (1995) showed that there was a not a significant difference between total yield from one year to the next. Because there was a negative relationship between seeds produced and the number of racemes per plant, a high yielding genotype should do well in multiple environments and years. Moshkin (1986) suggested that yield could be greatly improved by applying nitrogen before the formation of the spike. Severino and Auld (2013) show that each castor seed contributes distinctly to the yield, because it grows and matures under a unique environment. They also showed that the weight of seeds from racemes produced within the same week could differ by over 100 mg. This indicates that because castor is an indeterminate plant, each week is important and that each raceme should be managed to reduce stresses. Soratto (2012) found that by decreasing row and plant spacing could result in significantly higher castor yields.

<u>Oil Content</u>

Castor is an important industrial crop, which produces the unique fatty acid, ricinoleic acid. Ricinoleic acid is used in the production of nylon, cosmetics, lubricants, and it also has medicinal uses. The United States imports approximately 15 thousand metric tons of castor oil each year (U.S. Census, 2011). Castor seed has between 37 and

60 % oil with most of the fatty acids (eighty to ninety %) being ricinoleic acid (Atsmon, 1989). Oil content can be quickly and easily determined from Nuclear Magnetic Resonance (NMR). NMR has been used for fifty years in determining the amount of oil from dry oilseeds as first described by Conway and Earle in 1966. They showed that NMR had a correlation of $r = 0.993$ with oil extraction by petroleum ether extraction of 12 plant species including castor. Koutroubas et al. (1999) determined that oil production was highly dependent on genotype with only a small influence from environment. Hooks et al. (1971) showed that oil content and seed yield were negatively correlated at a significant value of $r = -0.63$ ($P \leq 0.01$). Zimmerman (1958) found there was a large environmental influence on the percent of oil produced by castor. This would indicate that a large number of environments would need to be tested to determine an appropriate genotype for optimum oil production. Hooks et al. (1971) stated that within a single environment the amount of oil did not significantly differ between years. They also found that oil content was also highly effected by maternal influence. This suggests that like seed yield a single genotype may not be best for several environments. Genotypes with high oil content will need to be identified on an environment specific basis. Also, work showed that only one or two years will provide enough data to predict an Environment's oil production. Moshkin (1986) also determined that the central racemes, those produced first, had higher oil content than those produced later. This may be due to the short growing season of the temperate climate that he was working in. Oil content like seed yield would be optimized by plants with a low number of racemes as long as the production on these racemes was maximized. Moshkin (1986) also determined that seeds that were produced under optimal water conditions produced higher levels of oil then

those that were under water stress. This could also indicate an environmental impact on the production of oil. Environments that do not have adequate rainfall or supplemental irrigation may not be suited for castor production.

Chapter III

Materials and Methods

In 2011, five genotypes of castor were selected based on: 1) the ability to be mechanically harvested; and 2) a wide range of plant architecture, and 3) as released or soon to be released germplasm. The environments were chosen based on the interest in castor of individual participants at an Association for the Advancement of Industrial Crops symposium at Fort Collins, CO 2010.

2011 Field Studies

In 2011, five genotypes were chosen for this trial: Brigham developed by Texas Tech University for release in 2015; Energia developed by Embrapa in Brazil 2007; Hale developed by USDA-ARS and Texas Agricultural Experiment Station 1970; Memphis and Ultra Dwarf developed by Mississippi State University for release in 2014. These genotypes were grown in different environments, soil types, and row spacings as seen in Table IV. The trial at Prosper was lost due to excessive rains and a late planting. At all environments, the trial was planted in a 5 x 5 Latin square. In Prosper, the trial was planted with three rows per variety at the other environments the plots were sown with four rows per entry. At maturity, 2 meters were harvested from the center two rows at Citra, Florida, Kinderhook, NY, Lubbock, TX, and Pecos, TX. All above ground biomass was also harvested and weighed from the two meters in all environments except Citra, FL to provide harvest index. The seed was weighed then cleaned. A second weight of the clean seed was then taken. The seed was then evaluated for oil content using a Nuclear Magnetic Resonance (NMR). Three seed samples from each harvested meter plot were analyzed. Harvest index for the oil was also calculated.

2012 Field Studies

In 2012, the same five genotypes were planted under unique growing conditions as seen in Table IV. At all environments the trial was planted in a 5 x 5 Latin square. At Prosper, ND and Citra, FL the trial was planted with three rows per variety at the other environments the plots were sown with four rows per entry. At the end of the season, two meters were harvested from the center two rows at Kinderhook, NY, Lubbock, TX, and Simcoe, ON. At Prosper and Citra 2 meters was harvested from the center row. Also at Citra two harvests were obtained by rattooning the plants, the first was 121 days after planting and the second was 125 days after rattooning. All above ground biomass was also harvested from the two meters except at Prosper, ND and Simcoe, ON. The seed was weighed then cleaned. A second weight of the clean seed was then taken. The total above ground biomass was also weighed to provide a harvest index. The seed was then evaluated for oil content using a Nuclear Magnetic Resonance (NMR). Three samples from each harvested meter plot were examined. Harvest index for the oil was calculated.

2013 Field Studies

In 2013, the same five genotypes were planted at two environments (Table IV). The trial was again planted in a 5x5 Latin square at each environment. At the end of the season, two meters were harvested from the center two rows at both environments. All above ground biomass was also harvested from the two meters at Lubbock, TX. The seed was weighed then cleaned. A second weight of the clean seed was then taken. The total above ground biomass was also weighed to provide a harvest index. The seed was then evaluated for oil content using a Nuclear Magnetic Resonance (NMR). Three samples from each harvested meter plot were examined. Harvest index for the oil was calculated.

Data were analyzed using SAS 9.3 with the GLIMMIX Procedure. Oil content and harvest indices were arcsine square root transformed to normalize the data as described by Zar (2010). The results were derived using the different measured traits to evaluate the differences of performance between the five genotypes in each unique environment.

Table 1. Soil type, latitude, longitude, elevation, row spacing, and test years of six environments where 5 castor genotypes were tested for yield and harvest index.

						Test years		
Test Environments	Soil Type	Latitude	Longitude	Elevation	Row Spacing	2011	2012	2013
				--m--	--m--			
Citra, FL	Sparr Fine Sand	29° 24' N	82° 10' W	21	0.91	X	X	---
Pecos, TX	Hoban silty clay loam	31° 22' N	103° 37' W	789	0.97	X	---	---
Lubbock, TX	Acuff loam	33° 36' N	101° 54' W	990	1.02	X	X	X
Kinderhook, NY	Knickerbocker fine sandy loam	42° 25' N	73° 40' W	61	0.76	X	X	---
Simcoe, ON	Norfolk loamy sand	42° 51' N	80° 16' W	241	0.75	---	X	X
Prosper, ND	Pella clay loam	46° 57' N	97° 01' W	275	0.61	X	X	---

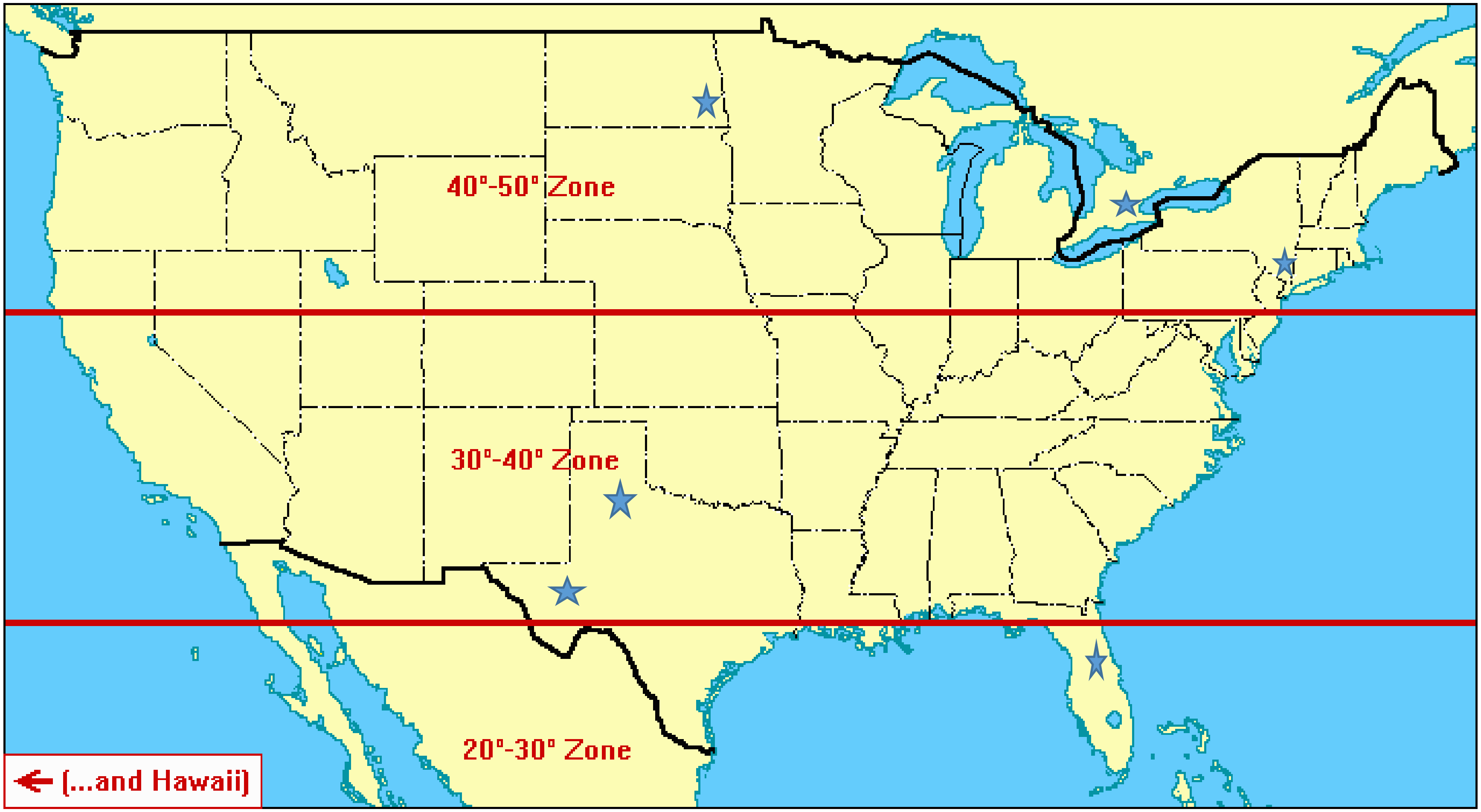

Figure 1. Locations of the 12 environments that five castor cultivars were grown from 2011 to 2013. (Adapted from Chandler, 2014)

Chapter IV

Results and Discussion

This study on the genotype by environment interaction of castor was conducted from 2011 to 2013 to determine the agronomic potential of five genotypes in twelve geographically and climatically unique environments. Genotypes were chosen based on their ability to be mechanically harvested and yet still have a wide range of diversity based on plant architecture. Entries were also chosen to represent currently or soon to be commercially available genotypes.

The aim of this study was to identify the best cultivar for each environment based on the traits of harvest index, seed weight, seed yield, and oil content.

Environments - 2011

In 2011 trials were conducted from south to north at Citra, FL, Pecos, TX, Lubbock, TX, Kinderhook, NY, and Prosper, ND. At Citra, FL the average yields were between 186 and 590 kg ha^{-1} with Energia producing the highest yields and Hale producing the lowest (Table 2). These yields are lower than those obtained by Campbell (2014) though those yields were influenced by plant growth regulators. At Citra, FL the average oil content ranged from 45.8 to 39.8 %, with Ultra Dwarf having the highest level of oil and Memphis producing the lowest (Table 3). The average oil yield per cultivar was highest with Energia and lowest with Hale with a range of 262 to 277 kg ha^{-1} (Table 4). Seed weight ranged from 26.8 to 17.7 grams per 100 seeds with Energia being the best and Ultra Dwarf being the worst (Table 5). Energia had larger seed, which contributed to the total yield and the oil yield. Pecos, TX yields ranged from Hale was the highest yielding variety with 1541 and Energia being the lowest at 857 kg ha^{-1} (Table 2).

These yields were not statistically different from each other. At Pecos, TX the oil content ranged from Ultra Dwarf at 46.1 to 40.4 % for Memphis (Table 3). The estimated oil yield at Pecos, TX ranged from 694 for Hale to 376 kg ha^{-1} for Energia though none of the oil yields were statistically better than the others (Table 4). The seed weight ranged from 23.9 for Hale to 18.6 grams per 100 seeds for Brigham, but these values were not statistically different (Table 5). Ultra Dwarf and Hale both had the highest seed harvest index at 29.3 and Energia performed the worst with 21.3 % (Table 6). Ultra Dwarf had the highest oil harvest index at 24.6 to Energia at 19 % (Table 7). At Lubbock, TX the seed yields ranged from 1484 for Ultra Dwarf to 713 kg ha^{-1} for Energia (Table 2). Energia had the highest oil content at 45.0 and 42.8 % for Memphis (Table 3). The estimated oil yielded from 649 for Ultra Dwarf to 320 kg ha^{-1} for Energia (Table 4). Energia and Hale both had the highest seed weights at 26.4 and Ultra Dwarf had the lowest at 21.5 grams per 100 seeds (Table 5). The seed harvest index ranged from Ultra Dwarf's 22.2 to 11.6 % for Energia (Table 6). Brigham produced the highest oil harvest index at 13.9 and Hale was the lowest at 12.7 % although they were not statistically different (Table 7). Kinderhook, NY had seed yields ranged from 1210 for Memphis to 766 kg ha^{-1} for Hale (Table 2). The oil content at Kinderhook ranged from 43.9 for Ultra Dwarf to 40.3 % for Energia (Table 3). The oil yield was highest with Memphis at 489 to 322 kg ha^{-1} for Hale, but there was no significant difference in the yields (Table 4). The seed weights ranged from 33.5 for Memphis to 25.5 grams per 100 seeds for Ultra Dwarf (Table 5). The seed harvest index ranged from 37.5 for Ultra Dwarf to 17.6 % for Energia (Table 6). The oil harvest index ranged from 23.6 for Ultra Dwarf to 11.2 % for Energia (Table 7).

Environments - 2012

In 2012 trials were conducted from south to north at Citra, FL, Lubbock, TX, Kinderhook, NY, Simcoe, ON, and Prosper, ND. At Citra, FL the crop was rattooned after 121 days resulting in two harvests. The two harvests were analyzed separately due to differences in growing conditions. The first harvest took place 121 days after planting and the second took place 125 days after rattooning. The seed yield ranged from 614 for Energia to 435 kg ha^{-1} for Ultra Dwarf for the first harvest which was not significantly different and 387 for Ultra Dwarf and 207 kg ha^{-1} for Brigham for the second harvest (Table 2). The oil content ranged from 49 for Energia to 37.6 % for Memphis for the first harvest and 40.8 for Ultra Dwarf to 33.9 % for Hale though there was no significant difference between the entries (Table 3). The oil yield for the first harvest ranged from 292 for Energia to 201 kg ha^{-1} for Ultra Dwarf and Memphis, but there was no significant difference between the entries (Table 4). The second harvest resulted in a yield of 164 for Ultra Dwarf to 75 kg ha^{-1} for Brigham though there was no significant difference between the entries (Table 4). The first harvest resulted in seed weights of 24.2 for Energia to 18.7 grams per 100 seeds for Ultra Dwarf (Table 5). The second harvest resulted in seed weights of 30.0 for Brigham to 20.0 grams per 100 seeds (Table 5). The seed harvest index for the first harvest ranged from 12.1 for Ultra Dwarf to 6.8 % for Energia, but there was no significant difference between the entries (Table 6). The second harvest resulted in seed harvest index ranging from 8.3 for Ultra Dwarf to 2.2 % for Hale (Table 6). The oil harvest index for the first harvest ranged from 3.5 for Ultra Dwarf to 0.8 % for Hale (Table 7). At Lubbock the seed yields ranged from 796 for Ultra Dwarf to 547 kg ha^{-1} for Memphis, but the yields were not significantly different (Table 2). The oil content ranged from 48.6 for Brigham to 44.8 % for Memphis (Table 3). The oil yield ranged from 369 for Ultra Dwarf to 210 kg ha^{-1} for Brigham, but the yields were not significantly different (Table 4). The seed weight ranged from 32 for Energia to 29 grams per 100 seeds for Brigham (Table 5). The seed harvest index ranged from 17.4 for Energia to 10.9 % for Brigham, but there was no significant difference in entries (Table 6). The oil harvest index ranged from 22.6 for Brigham to 19.7 % for Memphis (Table 7). Kinderhook seed yields ranged from 2892 for Memphis to 1339 kg ha^{-1} for Hale (Table 2). The oil content ranged from 48.1 for Energia to 43.9 % for Memphis (Table 3). The oil yield ranged from 1345 for Energia to 546 kg ha^{-1} for Hale (Table 4). The seed weight ranged from 35.3 for Energia to 32.1 grams per 100 seeds for Memphis, but there was no significant difference between the entries (Table 5). The seed harvest index ranged from 19.5 for Memphis to 7.3 % for Hale (Table 6). The oil harvest index ranged from 3.7 for Memphis to 1.5 % for Hale (Table 7). Simcoe seed yields ranged from 2023 for Hale to 1588 for Ultra Dwarf kg ha^{-1}, but the yields were not significantly different (Table 2). The oil content ranged from 48.5 for Ultra Dwarf to 45.6 % for Memphis (Table 3). The oil yield ranged from 944 for Hale to 768 kg ha^{-1} for Energia, but there was no significant difference between the entries (Table 4). The seed weights ranged from 35.6 for Memphis to 29.0 grams per 100 seeds for Ultra Dwarf (Table 5). Prosper seed yields ranged from 1895 for Hale to 1277 kg ha^{-1} for Brigham, but there was no significant difference between the entries (Table 2). The oil content ranged from 45.1 for Ultra Dwarf to 41.4 % for Brigham, but there was no significant difference between the entries (Table 3). The oil yields ranged from 831 for Hale to 532 kg ha^{-1} for Brigham, but there was no significant difference between the entries (Table 4). Seed weights ranged from 34.0 for Memphis to 24.8 grams per 100 seeds for Brigham (Table 5).

Environments - 2013

In 2013, trials were conducted from south to north at Lubbock, TX and Simcoe, ON. The seed yields in Lubbock ranged from 1622 for Energia to 1067 kg ha^{-1} for Ultra Dwarf, but there was not a significant difference between the entries (Table 2). The oil content ranged from 51.3 for Energia to 49.5 % for Memphis (Table 3). The oil yield ranged from 827 for Energia to 533 kg ha^{-1} for Ultra Dwarf, but there was not a significant difference between entries (Table 4). The seed weights ranged from 30.2 for Ultra Dwarf to 29.5 grams per 100 seeds for Energia, but there was not a significant difference between the genotypes (Table 5). Seed harvest index ranged from 36.5 for Ultra Dwarf to 17.8 % for Energia (Table 6). Oil index ranged from 15.5 for Energia to 12.1 % for Brigham, but the entries were not significantly different (Table 7). The seed yield in Simcoe ranged from 2445 for Memphis to 1825 kg ha^{-1} for Brigham, but there was no significant difference between the entries (Table 2). The oil content ranged from 51.3 for Energia to 49.5 % for Memphis (Table 3). The oil yield ranged from 1062 for Energia to 742 kg ha^{-1} for Hale, but there was no significant difference between the entries (Table 4). The seed weight ranged from 33.5 for Memphis to 27.2 grams per 100 seeds for Ultra Dwarf (Table 5).

Genotypic Differences

The average seed yield ranged from 2892 for Memphis to 186 kg ha^{-1} for Hale (Table 2). Statistically there was no difference in the average yield of the genotypes as seen in table 2. This indicates that the genotypes chosen had similar yield potential, but the environment elevated or depressed these yield potentials. At Citra, FL the two year average seed yield ranged from 503 for Energia to 342 kg ha^{-1} for Hale (Table 2). The three year average seed yield for Lubbock ranged from 1117 for Memphis to 1034 kg ha^{-1} for Energia (Table 2). The two year average seed yield for Kinderhook was 2051 for Memphis to 1053 kg ha^{-1} for Hale (Table 2). The two year average seed yield for Simcoe ranged from 2044 for Memphis to 1777 kg ha^{-1} for Brigham (Table 2). Unfortunately, there were too many confounding effects to explain the differences between these environments. One of the main driving forces between the differences in yield may be due to row spacing of the different environments as shown in Table 1. Sorrato et al. (2012) showed that a narrower row spacing significantly increased castor yields. The oil content ranged from 51.3 for Energia to 33.9 % for Hale (Table 3). The average oil content between genotypes were significantly different this suggests that there is a large influence of genotype on oil production (Table 3). These findings were similar to those reported by Koutroubas et al. (1999) and Hooks et al. (1971) that genotype and maternal influence contributed to differences in oil content. The two year average oil production at Citra, FL ranged from 44.0 for Energia to 38.1 % for Memphis (Table 3). The three year average oil production for Lubbock, TX ranged from 48.0 for Energia to 45.7 % for Memphis (Table 3). The two year average oil production for Kinderhook, NY ranged from 44.8 for Ultra Dwarf to 42.2 % for Memphis (Table 3). The two year average oil production for Simcoe, ON ranged from 45.9 for Ultra Dwarf to 42.6 % for Hale (Table 3). The oil yield ranged from 1345 for Energia to 77 kg ha^{-1} for Hale (Table 4). The average oil yield between genotypes was not significantly different as shown in Table 4. This suggests that there may be a tradeoff between oil and seed production as Memphis which was often the highest producer in seed yield was usually the lowest in oil content. There may also have been a reduction in the maturity of the seeds as the more southern environments were almost always mathematically higher in oil content then the northern environments. The two year average oil yield for Citra, FL ranged from 224 for Energia to 136 kg ha^{-1} for Memphis (Table 4). The three year average oil yield at Lubbock, TX ranged from 517 for Ultra Dwarf and Hale to 504 kg ha^{-1} for Energia (Table 4). The two year average oil yield for Kinderhook, NY ranged from 902 for Energia to 434 kg ha^{-1} for Hale (Table 4). The two year average oil yield for Simcoe, ON ranged from 915 for Energia to 782 kg ha^{-1} for Brigham (Table 4). The seed weights ranged from 35.6 for Memphis to 17.7 grams per 100 seeds for Ultra Dwarf (Table 5). The average seed weights were significantly different between the different genotypes (Table 5). These results were similar to those of Vallejos et al. (2011), but they were contrary to Moshkin (1986) as there was no decrease in seed weight from south to north. The two year average seed weight at Citra, FL ranged from 24.7 for Brigham to 18.8 grams per 100 seeds for Ultra Dwarf (Table 5). The three year average seed weight for Lubbock, TX ranged from 29.3 for Energia to 27.3 grams per 100 seeds for Ultra Dwarf (Table 5). The two year average seed weight for Kinderhook ranged from 32.8 for Memphis to 30.3 grams per 100 seeds for Brigham and Ultra Dwarf (Table 5). The two year average seed weight for Simcoe, ON ranged from 34.6 for Memphis to 28.1 grams per 100 seeds for Ultra Dwarf and Brigham (Table 5). The seed harvest index ranged from 37.5 for Ultra Dwarf to 2.2 % for Hale (Table 6). The average seed index between genotypes was significantly different between genotypes as shown in Table 6. The average seed harvest index for Citra, FL ranged from 6.3 for Brigham to 10.2 % for Ultra Dwarf (Table 6). The three year average seed harvest index ranged from 15.6 for Energia to 25.3 % for Ultra Dwarf (Table 6). The two year average seed harvest index for Kinderhook, NY ranged from 14.3 for Hale to 27.5 % for Memphis (Table 6). Ultra Dwarf always ranked highest for seed harvest index this suggests that a dwarf type castor would be most desirable as more inputs are going to seed production. This is consistent with the work of Norman Borlaug and others that increased crop production in the green revolution by using dwarf genotypes. The oil harvest index ranged from 24.6 for Ultra Dwarf to 0.9 % for Memphis and Energia (Table 7). The average oil harvest index between genotypes was significantly different between genotypes as shown in Table 7. The average oil harvest index for Citra, FL ranged from 4.2 for Ultra Dwarf to 2.1 % for Energia (Table 7). The three year average oil harvest index for Lubbock, TX ranged from 17.0 for Energia to 16.0 % for Memphis (Table 7). The two year average oil harvest index ranged from 13.4 for Ultra Dwarf to 7.5 % for Hale (Table 7). Ultra Dwarf again had the highest harvest index of all the genotypes tested.

Genotype by Environment Interaction

Memphis, on average, had higher seed yields in the northern environments than in the southern environments. This suggests that the lower rainfall or higher temperatures of the southern environments were detrimental to the overall production of this variety. Energia averaged higher seed yields, oil yield, seed harvest index, and higher seed weights in the northern environment, however it produced higher levels of oil and oil harvest index in the southern environments. Ultra Dwarf on average performed better in the northern environments than the southern, however the oil harvest index for all environments was similar. Hale on average performed better in the northern environments except for oil content and oil harvest index. Brigham also on average performed better in the northern environments than the southern in seed yield, oil yield, and seed weight. However, in the southern environments oil content, seed harvest index, and oil index of Brigham were all higher. On average the southern environments are better for producing higher levels of oil within the seeds, but with the northern environments' ability to produce both higher seed yields and more total oil per hectare made them higher yielding than the southern ones.

Table 2. Seed yield of five castor genotypes grown in 12 environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		kg seed ha^{-1}					
Citra, Fl	2011	230 c†	590 a	399 b	186 c	446 b	**370**
	2012 (H1)	516 a†	614 a	435 a	588 a	534 a	**537**
	2012 (H2)	313 a†	305 a	387 a	251 a	207 a	**293**
Environment Average		**353**	**503**	**407**	**342**	**396**	**400**
Pecos, TX	2011	1524 a†	857 a	1189 a	1541 a	878 a	**1198**
Lubbock, TX	2011	1306 a†	713 b	1484 a	1108 ab	1137 ab	**1150**
	2012	547 a†	768 a	796 a	722 a	431 a	**653**
	2013	1499 a†	1622 a	1067 a	1427 a	1587 a	**1440**
Environment Average		**1117**	**1034**	**1116**	**1086**	**1052**	**1081**
Kinderhook, NY	2011	1210 a†	1137 a	980 a	766 a	840 a	**987**
	2012	2892 a†	2740 a	2152 b	1339 c	1984 b	**2222**
Environment Average		**2051**	**1939**	**1566**	**1053**	**1412**	**1605**
Simcoe, ON	2012	1874 a†	1611 a	1588 a	2023 a	1729 a	**1765**
	2013	2214 a†	2445 a	2035 a	1877 a	1825 a	**2079**
Environment Average		**2044**	**2028**	**1812**	**1950**	**1777**	**1922**
Prosper, ND	2012	1370 a†	1432 a	1806 a	1895 a	1277 a	**1556**
Cultivar Average		**1291 a†**	**1236 a**	**1193 a**	**1144 a**	**1073 a**	**1187**

† Values within a row followed by the same letter are not significantly different at $P \leq 0.05$ by Fisher's Least Significant Difference Test.

Table 3. Percent oil of five castor genotypes grown in 12 environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		---------------	---------------	--% Oil--	---------------	---------------	---------------
Citra, Fl	2011	39.8 d†	44.7 ab	45.8 a	41.3 cd	43.4 bc	**43.0**
	2012 (H1)	37.6 b†	49.0 a	44.4 ab	47.1 a	48.2 a	**45.3**
	2012 (H2)	36.8 a†	38.4 a	40.8 a	33.9 a	36.3 a	**37.3**
Environment Average		**38.1**	**44.0**	**43.7**	**40.8**	**42.6**	**41.9**
Pecos, TX	2011	40.4 c†	43.7 b	46.1 a	44.3 ab	43.2 b	**43.5**
Lubbock, TX	2011	42.8 b†	45.0 a	43.7 ab	44.8 a	44.7 a	**44.2**
	2012	44.8 c†	47.6 ab	46.3 b	47.2 ab	48.6 a	**46.9**
	2013	49.5 b†	51.3 a	49.9 b	50.0 b	50.4 ab	**50.2**
Environment Average		**45.7**	**48.0**	**46.6**	**47.3**	**47.9**	**47.1**
Kinderhook, NY	2011	40.4 c†	40.3 c	43.9 a	42.1 b	42.1 b	**41.7**
	2012	43.9 c†	48.1 a	45.6 b	45.2 b	45.7 b	**45.7**
Environment Average		**42.2**	**44.2**	**44.8**	**43.7**	**43.9**	**43.7**
Simcoe, ON	2012	45.6 c†	47.5 ab	48.5 a	46.5 bc	46.8 bc	**47.0**
	2013	40.2 bc†	43.1 a	43.3 a	38.7 c	41.1 b	**41.3**
Environment Average		**42.9**	**45.3**	**45.9**	**42.6**	**44.0**	**44.2**
Prosper, ND	2012	43.6 a†	43.7 a	45.1 a	43.9 a	41.4 a	**43.5**
Cultivar Average		**42.1 c†**	**45.2 a**	**45.3 a**	**43.7 b**	**44.3 b**	**44.1**

† Values within a row followed by the same letter are not significantly different at $P \leq 0.05$ by Fisher's Least Significant Difference Test of arcsine square root transformed data.

Table 4. Total oil yield of five castor genotypes grown in 12 environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		---	-----------	kg oil ha^{-1}	-----------	-----------	---
Citra, FL	2011	90 c†	262 a	183 b	77 c	194 b	**161**
	2012 (H1)	201 a†	292 a	201 a	281 a	255 a	**246**
	2012 (H2)	117 a†	119 a	164 a	89 a	75 a	**113**
Environment Average		**136**	**224**	**183**	**149**	**175**	**173**
Pecos, TX	2011	623 a†	376 a	551 a	694 a	385 a	**526**
Lubbock, TX	2011	555 a†	320 b	649 a	496 a	507 a	**505**
	2012	246 a†	366 a	369 a	339 a	210 a	**306**
	2013	739 a†	827 a	533 a	715 a	799 a	**723**
Environment Average		**513**	**504**	**517**	**517**	**505**	**511**
Kinderhook, NY	2011	489 a†	458 a	430 a	322 a	353 a	**410**
	2012	1250 ab†	1345 a	1025 bc	546 d	883 c	**1010**
Environment Average		**870**	**902**	**728**	**434**	**618**	**710**
Simcoe, ON	2012	862 a†	768 a	770 a	944 a	811 a	**831**
	2013	877 a†	1062 a	899 a	742 a	753 a	**867**
Environment Average		870	915	835	843	782	**849**
Prosper, ND	2012	590 a†	622 a	817 a	831 a	532 a	**678**
Cultivar Average		**553 a†**	**568 a**	**549 a**	**506 a**	**480 a**	**531**

† Values within a row followed by the same letter are not significantly different at P≤ 0.05 by Fisher's Least Significant Difference Test.

Table 5. Seed weight of five castor genotypes grown in 12 environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		-- g/100 seeds					--
Citra, Fl	2011	21.2 bc†	26.8 a	17.7 c	23.4 ab	23.5 ab	**22.5**
	2012 (H1)	24.1 a†	24.2 a	18.7 c	22.9 a	20.7 b	**22.1**
	2012(H2)	24.7 a†	20.8 a	20.0 a	27.1 a	30.0 a	**24.5**
Environment Average		**23.3**	**23.9**	**18.8**	**24.5**	**24.7**	**23.0**
Pecos, TX	2011	22.6 a†	20.8 a	19.0 a	23.9 a	18.6 a	**21.0**
Lubbock, TX	2011	26.2 a†	26.4 a	21.5 b	26.4 a	23.8 ab	**24.9**
	2012	30.4 a†	32.0 a	30.1 a	29.3 a	29.0 a	**30.2**
	2013	29.8 a†	29.5 a	30.2 a	29.6 a	30.1 a	**29.8**
Environment Average		**28.8**	**29.3**	**27.3**	**28.4**	**27.6**	**28.3**
Kinderhook, NY	2011	33.5 a†	27.9 c	25.5 d	31.3 b	27.3 c	**29.1**
	2012	32.1 a†	35.3 a	35.1 a	33.8 a	33.2 a	**33.9**
Environment Average		**32.8**	**31.6**	**30.3**	**32.6**	**30.3**	**31.5**
Simcoe, ON	2012	35.6 a†	33.1 ab	29.0 c	34.5 ab	31.2 bc	**32.7**
	2013	33.5 a†	29.3 a	27.2 a	27.8 a	28.5 a	**29.3**
Environment Average		**34.6**	**31.2**	**28.1**	**31.2**	**29.9**	**31.0**
Prosper, ND	2012	34.0 a†	27.5 c	27.1 c	31.6 b	24.8 d	**29.0**
Cultivar Average		**29.1 a†**	**27.8 bc**	**25.1 d**	**28.4 ab**	**26.7 c**	**27.4**

† Values within a row followed by the same letter are not significantly different at $P \leq 0.05$ by Fisher's Least Significant Difference Test.

Table 6. Harvest index (total seed yield) of five castor genotypes grown in eight environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		-------------------------------% Harvest Index (Total Seed Yield)---------------------------------					
Citra, Fl	2012 (H1)	10.5 a†	6.8 a	12.1 a	12.0 a	9.8 a	**10.2**
	2012 (H2)	2.7 b†	2.3 b	8.3 a	2.2 b	2.8 b	**3.7**
Environment Average		**6.6**	**4.6**	**10.2**	**7.1**	**6.3**	**7.0**
Pecos, TX	2011	27.8 a†	21.3 a	29.3 a	29.3 a	26.6 a	**26.8**
Lubbock, TX	2011	17.9 bc†	11.6 d	22.2 a	15.1 c	19.0 ab	**17.2**
	2012	11.4 a†	17.4 a	17.2 a	13.4 a	10.9 a	**14.1**
	2013	28.1 b†	17.8 c	36.5 a	29.7 ab	30.6 ab	**28.5**
Environment Average		**19.1**	**15.6**	**25.3**	**19.4**	**20.2**	**19.9**
Kinderhook, NY	2011	35.4 a†	17.6 b	37.5 a	21.2 b	20.7 b	**26.5**
	2012	19.5 a†	14.1 b	14.6 ab	7.3 c	10.6 bc	**13.2**
Environment Average		**27.5**	**15.9**	**26.1**	**14.3**	**15.7**	**19.9**
Cultivar Average		**19.2 b†**	**13.6 d**	**22.2 a**	**16.3 c**	**16.4 c**	**17.5**

† Values within a row followed by the same letter are not significantly different at P ≤ 0.05 by Fisher's Least Significant Difference Test of arcsine square root transformed data.

Table 7. Harvest index (oil yield) of five castor genotypes grown in eight environments across North America in 2011 to 2013.

Environments	Years	Memphis	Energia	Ultra Dwarf	Hale	Brigham	Environment Average
		------------------------------------% Harvest Index (Oil Yield)--					
Citra, Fl	2012 (H1)	4.0 a†	3.2 a	4.8 a	5.7 a	4.3 a	**4.4**
	2012 (H2)	0.9 b	0.9 b	3.5 a	0.8 b	0.9 b	**1.4**
Environment Average		**2.5**	**2.1**	**4.2**	**3.3**	**2.6**	**2.9**
Pecos, TX	2011	21.6 a†	19.0 a	24.6 a	24.3 a	23.8 a	**22.6**
Lubbock, TX	2011	13.3 a†	13.7 a	13.3 a	12.7 a	13.9 a	**13.4**
	2012	19.7 c†	21.9 ab	21.8 ab	21.5 b	22.6 a	**21.5**
	2013	14.9 a†	15.5 a	13.5 a	14.9 a	12.1 a	**14.2**
Environment Average		**16.0**	**17.0**	**16.2**	**16.4**	**16.2**	**16.4**
Kinderhook, NY	2011	21.1 a†	12.6 b	23.6 a	13.4 b	13.2 b	**16.8**
	2012	3.7 a†	3.0 ab	3.1 a	1.5 c	2.1 bc	**2.7**
Environment Average		12.4	7.8	13.4	7.5	7.7	**9.8**
Average		**12.4 b†**	**11.2 b**	**13.5 a**	**11.9 b**	**11.6 b**	**12.1**

† Values within a row followed by the same letter are not significantly different at P ≤ 0.05 by Fisher's Least Significant Difference Test of arcsine square root transformed data.

Table 8. Analysis of variance and F-test values of five castor genotypes evaluated in 12 environments of North America from 2011-2013.

Source of Variation	df	Seed Yield	Oil Content	Oil Yield	Seed Weight	df	Seed Harvest Index	Oil Harvest Index
		---------------------------F-Value------------------------					---------------------F-Value----------------	
Cultivar	4	2.15 ns	20.01***	1.80 ns	11.81***	4	14.91***	4.76***
Environment	11	52.66***	69.90***	52.64***	36.88***	7	83.02***	255.87***
CultivarXEnvironment	44	1.80***	2.28***	1.90***	2.09***	28	2.51***	2.40***
CV %		65.6	7.2	66.7	20.8		35.6	44.5

F-test shown with n.s. are not significantly different (p = 0.05) and those shown with *** are highly significantly different (p = 0.001).

Table 9. Average seed yield, oil content, total oil yield, seed weight, HI of total seed yield, and HI of oil yield of five castor genotypes grown in five northern (> 40° latitude) and seven southern environments (< 40° latitude) across North America in 2011 to 2013.

Cultivar	Seed Yield	Oil Content	Oil Yield	Seed Weight	Harvest Index Seed	Harvest Index Oil
	kg ha^{-1}	%	kg ha^{-1}	g/100 seeds	% HI	% HI
Memphis						
North (> 40° lat.)	1912	42.7	814	33.7	27.5	12.4
South (< 40° lat.)	716	41.7	367	25.6	16.4	12.4
Energia						
North (> 40° lat.)	1873	44.5	851	30.62	15.9	7.8
South (< 40° lat.)	782	45.7	366	25.8	12.9	12.4
Ultra Dwarf						
North (> 40° lat.)	1712	45.3	788	28.8	26.1	13.4
South (< 40° lat.)	822	42.3	379	22.5	20.9	13.6
Hale						
North (> 40° lat.)	1580	43.3	677	31.8	14.3	7.5
South (< 40° lat.)	832	44.1	299	26.1	12.1	13.3
Brigham						
North (> 40° lat.)	1531	43.4	666	29.0	15.7	7.7
South (< 40° lat.)	746	45.0	346	25.1	16.6	12.9

Chapter V

Conclusions

The objective of this study was to evaluate five castor genotypes in 12 unique test environments across North America. These genotypes were selected because they could be mechanically harvested, while still representing a wide range of plant architecture among released or soon to be released germplasm. There was not one genotype that outperformed the others in every environment or trait tested. The seed yields from this study ranged from 1912 kg ha^{-1} in the northern environments to 716 kg ha^{-1} in the southern environments. The oil yields from this study ranged from 814 kg ha^{-1} in the northern environments to 367 kg ha^{-1} in the southern environments. Northern environments produced higher levels of seed and oil yield per hectare compared to the southern environments. These results agreed with Baldwin and Cossar (2009). Domingo and Crooks (1945) also had consistently good seed yields at Urbana, Illinois which is near 40° North latitude. However, the Florida and Lubbock yields in this trial were mathematically higher than those obtained by Domingo and Crooks (1945). Seed yields were reduced by extraordinarily dry conditions in 2011 in Pecos, TX, Lubbock, TX, and both years at Kinderhook, NY. In 2012 seed yields at Lubbock, TX was reduced by late planting and an early freeze. The Citra, FL traits tested were burdened by foliar diseases in 2011 and 2012. This also agrees with Domingo and Crooks (1945) that castor grown at Florida was often plagued with foliar diseases. Average oil content observed in this study ranged from 42.1 % for Memphis to 45.3 % for Ultra Dwarf. These results were similar to those reported by Moshkin (1986) in which Russian castor genotypes produced significant differences in oil content within most environments. The average oil content between environments ranged from 37.3 % in Citra, FL to 50.2 % in Lubbock, TX. These results of this study were also similar to Zimmerman (1958) in that the environment resulted in a difference of 11% to 13 %. Oil yield ranged from 299 kg ha^{-1} in the southern environments to 851 kg ha^{-1} for the northern environments. Oil yields were much higher in the northern environments due to the higher total amount of seed produced even though the oil content was on average lower. Average seed weights for castor were not a very good indicator of seed yield, oil content, or oil yield. The highest seed yields observed in the northern environment that ranged from 1912 kg ha^{-1} for Memphis to 1531 kg ha^{-1} for Brigham. The highest seed weights were obtained from Memphis (33.7 g/100 seeds) and the lowest were from Ultra Dwarf (28.8 g/100 seeds). The highest seed yield was 832 kg ha^{-1} for Hale to Memphis 716 kg ha^{-1} in the southern environments and the highest seed weight ranged from Hale (26.1 g/100 seeds) to Ultra Dwarf (22.5 g/100 seeds). Oil content in the northern environments ranged from 45.3 % for Ultra Dwarf to 42.7 % for Memphis. In southern environments oil content ranged from 45.7 % for Energia to 41.7 % for Memphis. Total oil yields in northern environments ranged from 851 kg ha^{-1} for Energia to 666 kg ha^{-1} for Brigham. In the southern environments total oil yield ranged from 379 kg ha^{-1} for Ultra Dwarf to 299 kg ha^{-1} for Hale. The seed harvest index in the northern environment ranged from 27.5 % for Memphis to 14.3 % for Hale. In the southern environments, seed harvest index ranged from 20.9 % for Ultra Dwarf to 12.1 % for Hale. Oil harvest index ranged from 13.4 % for Ultra Dwarf to 7.5 % for Hale in the northern environments and in the southern environments it ranged from 13.6 for

Ultra Dwarf to 12.4 % for Memphis and Energia. The confounding effects of differential row spacing probably contributed to the differences between environments. The trials in southern environments were grown on rows approximately 20 cm farther apart than the northern environments. Soratto (2012) showed the importance of row spacing for maximizing the seed yields of semi-dwarf castor genotypes in Brazil. There were mathematical difference between the results observed between the different environments in the ranking of genotypes between the years. This suggests that the environments themselves were very different from one year to the next. Additional testing could give a better picture of the suitability of a genotype for a particular environment. Also new breeding efforts should be aimed at combining two or more of these genotypes to produce a better plant in more environments. Ultra Dwarf which performed very well in the northern environments could be combined with Brigham or Energia to produce a plant with higher oil content and higher seed weights which would produce better in the southern environments.

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Appendix

A separate study was performed in 2012 and 2013 on one genotype (PI 179 729) which was tested for inheritance of increased oleic acid when grown in a greenhouse environment. Gas chromatography (gc) was used to determine fatty acid composition. Breeding lines with reduced ricinoleic acid and higher levels oleic acid would produce methylesters with reduced viscosity and potentially better performance as biodiesel feedstock. This could increase the marketability of castor oil as a renewable fuel under the renewable fuels program if the visocity met the ASTM D445 requirements (ASTM, 2012).

<u>Fatty Acid Composition</u>

Wang et al. (2011) screened seed of 1033 genotypes and determined that castor genotypes vary greatly in fatty acid composition. The main fatty acids found to be in castor oil by Ramos et al. (1984) and Wang et al. (2011) were ricinoleic, oleic, linoleic, stearic, and, palmitic acid. Although ricinoleic acid is needed in many industrial products high levels of oleic acid could lower viscosity and subsequently make castor more appealing as a biodiesel. A natural mutant of castor in (PI 179 729) containing high levels of oleic acid was discovered by Rojas-Barros et al. (1998) during a screening of 191 accessions. After self-pollinating this mutant for three generations a twenty fold increase in oleic acid was achieved. Two epistatic genes (ol and Ml) were determined to be controlling the amount of oleic acid. Hamdan et al. (2009) also identified a gene (ol) as controlling oleic acid levels in safflower. Ma and Zai-yun (2007) showed that in *Brassica napus* L., oleic acid content could be improved by crossing selected inbred lines to make elite hybrids. Auld et al. (1992) showed that oleic acid in rapeseed could be improved through chemical mutagenesis in which he may have mutated an analog to ol. Moshkin (1986) determined that by ten days after anthesis, castor seeds have begun producing oil. He showed that 50 % of the total ricinoleic acid was produced within the first three weeks after anthesis. The large amount of ricinoleic acid produced during early seed maturation may indicate a fundamental need for this fatty acid in seed development and subsequent germination. Canvin (1963) showed that oleic acid had the second highest fatty acid concentration for the first two weeks after anthesis. He also showed that the synthesis of the individual fatty acids appeared to occur independently, while the proportion of each fatty acid changed as the oil concentration increased. Wang et al. (2011) showed that oil yields could be very similar between two different genotypes even though they had very different percentages of each fatty acid. Fatty acid composition may be influenced by the maternal parent just as the total oil content. Wang et al. (2011) showed that there was a significant negative relationship of $r = -0.74$ ($P \leq 0.0001$) between oleic and ricinoleic acid found within individual castor seeds.

Oleic Acid Inheritance

In 2012, one castor accession was selected to measure the inheritance of increased oleic acid content based on previous literature for the characteristic. The seeds evaluated were from greenhouse seed increase of this accession performed at Texas Tech University in 2008.

2012 Laboratory

In 2012 two randomly selected seeds from 21 individual plants increased from an accession with potentially increased oleic acid (PI 179 729) were compared to three high ricinoleic acid genotypes. The seeds were cut in half separating the endosperm and embryo, so that the plants could be grown after their fatty acid content of the distal end of the seed were measured. Endosperm samples were methylated using a modified Sanders Method (1978) at Texas A&M Agrilife Research & Extension Center at Lubbock, TX. Once samples had been methylated, the fatty acid methyl esters (F.A.M.E) were analyzed using a Agilent Technologies 6890N Network GC System gas chromatography (GC) at Texas Tech University to determine fatty acid composition. Three individual plants #4, 18, 19 which showed to have the highest levels of oleic acid then had 10, 5, and 5 seeds selected to reanalyze for levels of oleic acid.

Table 10. Fatty acid composition of 30 single castor plants grown in the greenhouse in the spring of 2012 at Texas Tech University in Lubbock, TX.

Genotype	Palmitic C16.0	Stearic C18.0	Oleic C18.1	Linoleic C18.2	Ricinoleic C18.1-OH
	----------------% Methyl Ester---------------------				
4b	3.31	3.14	27.60	11.98	53.97
4f (2012)	4.25	3.64	17.17	10.29	64.64
4I	2.29	1.95	15.57	6.56	73.62
19Aa	3.10	2.75	15.04	9.49	69.62
18e	2.62	3.60	14.55	8.54	70.69
18b	2.26	2.77	14.52	8.86	71.59
Memphis	2.63	2.62	14.19	11.63	68.94
4c	1.78	1.55	14.12	5.66	76.89
23a	3.74	3.35	13.71	13.42	65.77
Hale	2.64	3.59	13.39	11.17	69.20
18b	2.87	3.24	13.34	8.89	71.67
Brigham	2.30	2.69	13.31	9.38	72.32
6OPa	2.49	3.10	13.23	7.93	73.25
18OPa	4.48	3.41	13.18	12.57	66.37
18d	2.43	3.82	12.17	7.43	74.15
19Ae	3.24	3.34	10.58	8.35	74.49
10-selfed-a	3.52	4.22	10.55	14.14	67.58
4g	3.42	2.49	10.48	10.33	73.28
4e	2.99	1.95	9.96	9.46	75.63
19Aa	2.45	3.32	9.62	6.87	77.74
18c	1.80	1.52	9.32	6.55	80.81
19Ac (2012)	3.02	3.05	9.08	7.59	77.27
25b (2012)	3.87	4.07	9.00	10.05	67.48
18a	2.27	3.52	8.55	7.61	78.05
19Ad	3.95	2.31	8.37	6.30	79.07
19Ab	1.89	1.95	7.42	4.42	84.31
4h	2.76	2.19	7.11	9.20	78.75
4d	2.04	1.58	6.87	5.72	83.79
1b	4.52	3.85	6.78	7.56	69.60
4J	1.71	1.49	6.47	5.31	85.03
Average	**2.89**	**2.87**	**11.84**	**8.78**	**73.19**

2012 Greenhouse Study

The top 30 plants representing 11 parent selections were planted in 12 L pots at the Texas Tech University greenhouse (28±40C with 40% RH) a commercial soil mix (metromix 900) was used in a completely randomized design. On February 20, 2012 the first nine plants were planted. One week later February 27, 2012 another 21 plants were sown. A total of nine plants from both plantings germinated, these plants were self-pollinated using polyethylene selfing-bags. The plants were also allowed to open pollinate to see if there was a difference in the levels of oleic acid produced between the two methods. As individual racemes matured, they were removed from the plant with pruning shears and placed in their own sack. After all racemes had matured, the seeds were harvested by hand.

2013 Laboratory

In 2013, two random seeds were selected from each individual raceme of the nine individual plants grown from half seeds in 2012 to measure their heritability of oleic acid. Two seeds from five genotypes with high ricinoleic acid were used as procedural controls. The seeds were cut in half separating the endosperm and embryo, so that the plants could be grown after their fatty acid content was measured. Samples were methylated using a modified Sanders Method (1978) at Texas Tech University at Lubbock, TX. Once samples had been methylated, the fatty acid methyl esters (F.A.M.E) were analyzed using a gas chromatography (GC) at Texas Tech University to analyze fatty acid composition.

Table 11. Fatty acid composition of six single castor plants grown in the greenhouse in the spring of 2013 at Texas Tech University in Lubbock, TX.

Genotype	Palmitic C16.0	Stearic C18.0	Oleic C18.1	Linoleic C18.2	Ricinoleic C18.1-OH
	-------------------------------% methyl ester--------------------------				
19Ac (2013)	7.88	25.99	27.99	9.38	28.76
4f (2013)	1.63	5.99	18.73	2.62	71.03
25b (2013)	8.50	24.81	17.48	5.87	43.35
4f (2013)	1.71	6.45	15.04	2.70	74.11
4f (2013)	1.91	7.90	12.72	3.37	74.11
4f (2013)	2.36	11.41	8.56	2.89	74.77
Average	**4.00**	**13.76**	**16.75**	**4.47**	**61.02**

2013 Greenhouse Study

On March 20, 2013 the top 6 plants were planted in 12L pots at the Texas Tech University greenhouse (28±40C with 40% RH) a commercial soil mix (metromix 900) was used in a complete randomized design. Two plants were lost due to a severe hailstorm on June 5th, which broke the glass and damaged the plants. The plants that remained were self-pollinated using polyethylene mesh bags. The plants were also allowed to cross pollinate to see if there was a difference in the levels of oleic acid produced between the two methods. As individual racemes matured, they were removed from the plant with pruning shears and placed in their own sack. After all racemes had matured, the capsule was removed from the seed by hand.

Results

Oleic acid ranged from 27.6 to 6.47 % in 2012 and in 2013 oleic acid ranged from 27.99 to 8.56 %. Although there was an increase in the amount of oleic acid from the first year to the second, there was a large problem with germination in the selected genotype. The low percentage of germination suggests that there is a physiological inhibition due to increased oleic acid or a decrease of ricinoleic acid. He et al. (2007) suggested that there was a decrease in the enzyme activity of RcACS2 when there was a lack of ricinoleic acid. They also determined that this enzyme might be necessary for seed germination due to its high level of activity during germination.

The objective of this second study was to determine the inheritance of increased oleic acid content in a selected genotype. Although there was an increase in oleic acid from the previous year, there was a problem with germination for the lines with the highest levels. Rojas-Barros et al. (1998) also had problems with the germination of their seeds that had increased oleic acid. Traditional breeding will not likely lead to the development of a castor that has high levels of oleic acid. Another method such as mutagenesis or biotechnology may provide an alternate approach to increased oleic acid levels with acceptable germination rates though they may have to target a different pathway.