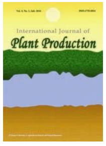

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Seed yield and oil quality of perennial castor bean in a Mediterranean environment

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Abstract

Castor (*Ricinus communis* L.) is an oilseed species, which in southern Italy is cultivated as annual during the spring-summer period under irrigation, but in most temperate areas such as those of eastern coast of Sicily, it could be grown as semi-perennial with no irrigation, by the adoption of autumnal sowings. A field experiment was conducted in southeastern Sicily with the aim of assessing plant surviving, seed yield and oil quality of four castor genotypes originating from different geographical areas (two Sicilian, one Tunisian and one Brazilian). The favorable climatic conditions allowed the plant to survive during the fall-winter period. Seed yield reached 3.45 t ha^{-1} on average of the two years and seed oil content ranged from 45% (Tunisian cultivar) to 48% ('Local RG 2' Sicilian genotype). Oil yield reflected the variation in seed yield. Genetic diversity for fatty acid composition and saponification number, iodine value and cetane number was evidenced. When ricinoleic acid is not taken into account, the oil of all genotypes satisfied the EU standards for biodiesel. The ricinoleic acid was the lowest (79%) in the Sicilian 'Local RG 2' and the highest (89%) in the Tunisian one, revealing a greater suitability of oil of the first genotype for biodiesel. In turn, the oil of Tunisian genotype could be exploited in other bio-based industrial sectors. The study also demonstrated that in the southeastern coast of Sicily autumnal sowings might be advantageous for castor grown as semi-perennial crop, mainly since they allow saving irrigation water.

Keywords: *Ricinus communis* L.; Wild germplasm; Autumnal sowing; Yield; Oil quality; Biodiesel.

Introduction

Castor (*Ricinus communis* L.) is an easily adaptable oilseed plant native of tropical Asia and Africa, that has become naturalized throughout the tropical and sub-tropical regions of the world. In these areas castor is a fast-growing perennial shrub being able to attain up to 10 to 13 m of height, whilst under temperate climates it becomes annual reaching 1.5 to 2.4 m in every single season (Anjani, 2012; Falasca et al., 2012). Castor seed oil represents a natural resource having a wide range of applications, mainly in non-food sectors of industry. Recently, castor oil has been also reported as a promising source of biofuel, although its fatty acid methyl esters (FAMEs) must be used in ratios of 10 or 20% in volume (B_{10} or B_{20}) when blended in petrodiesel, due to the high concentration (over 85%) of ricinoleic acid in triacylglycerols (TAGs), which confers a high viscosity to the oil. However, viscosity can be significantly reduced by transesterification to obtain pure castor FAMEs, useful as biodiesel (B_{100}) (Berman et al., 2011). Therefore, the production of biodiesel from castor oil is technically feasible and has an additional benefit over traditional lubricity additives, due to certain fuel properties as high oxidative stability and low cloud point (Berman et al., 2011; Lavanya et al., 2012). On the other hand, ricinoleic acid, which is a monounsaturated fatty acid with rare chemical features among vegetable oils, is believed to be responsible for certain castor oil healing properties. This fatty acid provides to the oil an extra stability to oxidation, mainly due to the presence of an hydroxyl group (-OH), enhancing its shelf life and making it and its derivatives suitable to prepare a number of products exploited in several industrial fields: medicines, cosmetics, paint, coatings, inks, biopolymers, lubricants, special aviation fuels (Caupin, 1997; Ogunniyi, 2006; Madankara et al., 2013).

Taking into account these potentials, a more extensive cultivation of castor should be encouraged through the use of simple and more advantageous agronomic options such as suitable cultivar, appropriate sowing dates and minimum water input, although irrigation promotes seed yield (Severino and Auld, 2013).

Currently, improved varieties and hybrids of castor with a high yield potential are available (Lavanya et al., 2012), but the wild germplasm of the species with different origin can be a source of new agronomic and oil quality traits for breeding to select more promising genotypes for specific industrial applications or as bioenergy source. In particular, the fatty acids profile of the oil is an important feature to consider for discriminating within the available germplasm of castor.

In semiarid prone environments of southern Italy, castor is usually considered as an annual spring-summer crop that requires irrigation, whose sowings are generally performed in April, when soil temperature reaches a stable level of 16-17 °C, ensuring rapid seed germination and a uniform crop establishment (Weiss, 1983; Falasca et al., 2012). However, under these environmental conditions, as it has been demonstrated for other macrothermal species such as kenaf and sunflower, early sowings could be advantageous also for castor, as they allow the root system to exploit the water stored in the soil during the winter period, reducing irrigation requirements (Anastasi et al., 2000; Patanè and Sortino, 2010).

Preliminary field experiments carried out in southern Sicily and in Tunisia have shown the possibility of exploiting the perennial habit of this species (Sortino et al., 2010a; Sortino et al., 2010b). Differently, in the same environment *Jatropha curcas* plants did not survive after cold winter season (Sortino et al., 2009).

Taking into consideration the ecological flexibility and the perennial habit of the species, early sowings may be beneficial under sub-humid to semiarid climates, since they may extend the growing season and thus allow a greater vegetative development of the plant, enhancing its productivity (Falasca et al., 2012; Ramanjaneyulua et al., 2013).

In this view, the aim of the present study was to explore the feasibility of growing castor as semi-perennial plant in southern Italy under rainfed regime by adopting the autumnal sowing and keeping the crop over a two- year period, through the evaluation of plant surviving, seed yield and oil quality.

Materials and Methods

Experimental site

Field experiment was conducted over the period 2009-2011 at Ispica (36° 47’ N, 14° 54’ E, 46 m a.s.l.), a coastal site of Ragusa province, in southeastern Sicily (South Italy). The soil of the experimental field is classified as typical Xerochrepts-Calcixerollic Xerochrepts-Lithic Xerorthents (USDA, Soil Taxonomy) having the following characteristics: clay 38.0%, sand 37.0%, silt 25.0%, organic matter 2.6%, pH 8.5, total N 1.6‰, available P_2O_5 52.3 mg kg^{-1}, exchangeable K_2O 325.0 mg kg^{-1}.

Genotypes and agronomic management

Four castor (*Ricinus communis* L.) genotypes originating from different geographical areas were compared. The seeds of two genotypes, indicated as 'Local RG 1' and 'Local RG 2', were collected from native plants in two different sites near Ragusa province in southeastern Sicily, whereas the seeds of the other two genotypes were collected from native plants in the areas of Médenine, in southeastern Tunisia and Maranguape, in northeastern Brazilian State of Ceará (reported as 'Tunisia' and 'Brazil', respectively). The experiment was arranged in a completely randomized blocks design with three replicates. Given the big dimension of native plants, exceeding 4 m of height in all genotypes, an equal inter-row and intra-row spacing (2×2 m) was adopted, in plots of 60 m^2 (6×10 m), bordered with an additional strip of plants.

The soil of the experimental area was ploughed during the summer before sowing at a 0.40 m depth and fertilized with 40, 60 and 60 kg ha^{-1} of N (as ammonium sulphate), P_2O_5 (as superphosphate) and K (as potassium sulphate), respectively. Sowing was carried out on 25 September, 2009 and the field area was irrigated with 40 mm of water to promote seed germination and plant establishment. Subsequently, the crop was maintained without irrigation. A further 40 kg ha^{-1} of N (as ammonium nitrate) was supplied as top dressing, when plants were approximately 50-cm tall. Weed control was performed once only, since the crop covered the soil and weeds could no longer grow.

After the last harvest in 2010 (late September), castor plants survived throughout the whole fall-winter season due to the favorable climatic conditions. Therefore, the two-year aged plants, grown with no further input in terms of fertilizer and water, started to flower in late February of the second year (2011) and were assessed again for productivity in the following summer.

Field measurements

Castor plants, whose height, as expected, exceeded 3 m of height in the second year of experiment, produced several lateral branches. Therefore, three harvests per year (from early July to mid August, in 2010, from late July to late August, in 2011) for both Sicilian genotypes and four harvests per year (from early July to late September, in 2010, from late July to late

September, in 2011) for 'Tunisia' and 'Brazil', due to a prolonged flowering period of these two last, were carried out along the growing season. The onset of capsules browning was considered as stage of physiological maturity. In 2010 the first harvest was performed on capsules of primary and secondary racemes (*i.e.* those first flowered) and fruits of racemes of higher orders were collected in later harvests. In 2011, capsules were collected progressively according to the flowering time on racemes of different orders.

At each harvest, five plants per each genotype were sampled and the number of racemes per plant, that of capsules per raceme and seed yield were measured. In addition, five representative racemes per plant were selected as sub-sample and the seeds were extracted manually from capsules for 1000-seed weight.

Seed oil content and fatty acid composition

Seed samples from the first harvest of each growing season were thoroughly cleaned and conditioned in laboratory at 4±1 °C. Oil content on a dry base of the crushed grain was determined according to the standard procedure (ISO 659) by extraction in a conventional Soxhlet apparatus using petroleum ether, bp 40-60 °C. The residual solvent was removed by a rotary evaporator under vacuum at 40 °C, then obtained lipid extracts were dehydrated over Na_2SO_4 and filtered. The fatty acid profile (FAs) of the oil was assessed following the official methodologies (ISO 5508 and 5509) to prepare methyl esters (FAMEs) and to perform gas chromatographic analysis using a HRGC Mega 2 system (Carlo Erba Instruments, Milano, Italy) equipped with flame ionization detector (FID). The stationary phase was a Supelcowax fused silica column (30 m length, 0.25 mm internal diameter, 0.25 µm film thickness). The specific operative conditions were: injector and detector temperature, 250 and 270 °C, respectively; oven temperature programmed from 190 to 250 °C with 2.5 °C/min gradient. He (115 kPa) was used as carrier gas, whereas chromatographic air (310 ml/min) and H (30 ml/min) were the auxiliary gases. N (30 ml/min) was used as make-up gas. One µl of sample was injected. The identification of FAs peaks was performed by comparison with relative retention times of FAMEs standards (Supelco Chem. Co. and Sigma Chem. Co.) and the concentration of each compound was expressed as a percentage of the area under the corresponding peak respect to the total area of all picks. Moreover, FAs categories, such as saturated (SFAs), unsaturated (UFAs), monounsaturated (MUFAs) and polyunsaturated (PUFAs) were calculated and the ratios between the major FAs (oleic/linoleic, oleic/ricinoleic) and between the FAs categories were derived. Other key indexes of the oil also important for biofuel quality, such as saponification number (SN, as mg K_2O g^{-1} oil), iodine value (IV, as I_2 100 g^{-1} oil) and cetane number (CN) were derived as described by Krisnangkura (1986) and Kalayasiri et al. (1996).

Data analysis

Data were subjected to the Bartlett's test for homoscedasticity and then statistically analyzed by a one-way analysis of variance (ANOVA) using CoStat version 6.003 (CoHort Software). Data were analyzed separately by cropping season (Soratto et al., 2012). The Student-Newman-Keuls (SNK) test was applied to evaluate differences between means (Snedecor and Cochran, 1989). Minimum threshold for significance was established at $P \leq 0.05$. In the present study, data of FAs composition of the oil are reported averaged across the two growing seasons, since negligible diferrences were evidenced.

Results and Discussion

Meteorological conditions and plant surviving

Weather conditions during the field experiment reflect those typical of a Mediterranean environment. Minimum temperatures ranged between 4 °C (February 2011) and 21 °C (August 2011), whereas those maximum varied from 15 °C (January 2010) to 33 °C (August 2010) (Figure 1). A total of 490 and 540 mm of rainfall were recorded in the first (from September 2009 to September 2010) and second (from October 2010 to September 2011) growing season, respectively, with a quite regular distribution from autumn to spring. Rainfall was almost absent during summertime.

Castor behaves as perennial under tropical and sub-tropical climatic conditions, whereas it is commonly cultivated as an annual warm-season elsewhere, as in Italy (Laureti et al., 1998). However, in certain areas of southern Italy with mild climate, such as those of southeastern Sicily where the present experiment was carried out, it can exhibit a semi-perennial habit. The species can be grown under a range of 200-4290 mm annual precipitation and tolerates a range of annual temperatures from 7.0 to

27.8 °C (Falasca et al., 2012). Indeed, in this experiment after the first cropping season (2009-2010) the plants survived during the fall-winter period since they experienced a favorable thermal regime and rainfall was adequate to satisfy crop water requirements.

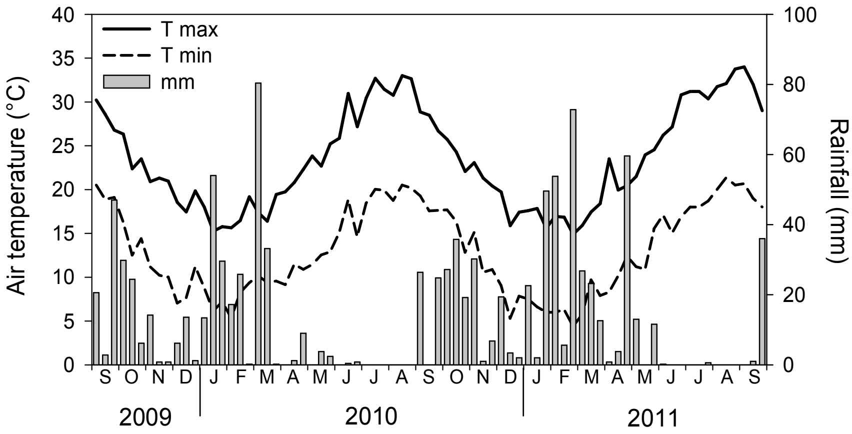

Figure 1. Maximum and minimum air temperatures (ten-day averages) and rainfall (ten-day total) during the experiment.

Yield components

The number of racemes per plant is an important yield component in castor (Severino et al., 2010). On average of genotypes it was equal to a total 18.8 racemes per plant in 2010 (Table 1). In 2011, plants differentiated a greater number of racemes reaching a total 80 racemes, on average of genotypes. A greater plant fertility in the second growing season may be ascribed to the favorable weather conditions occurred during the winter-spring season, especially in terms of rainfall (approx. 500 mm from October 2010 to May 2011). Castor is a tropical C3 plant that, according to its origin, benefits from heavy rainfall and high temperatures conditions. Areas with 450 to 750 mm of rainfall are reported as suitable for castor (Falasca et al., 2012). Favorable thermal conditions, together with the low plant population adopted in this experiment, may have promoted plant growth and following reproductive development. Indeed, as found by Kittock and Williams (1970) and Bizinoto et al. (2010), under widely spaced plants conditions a higher light interception results in a greater number of reproductive structures.

Table 1. Number of racemes per plant (total of harvests), number of capsules per raceme and 1000-seed weight (average of harvests) in four castor genotypes.

Genotype	Number of racemes per plant		Number of capsules per raceme		1000-seed weight (g)	
	2010	2011	2010	2011	2010	2011
Tunisia	26.7[a]	82.7[b]	40.2[d]	27.0[b]	423.8[a]	503.8[a]
Brazil	27.7[a]	105.3[a]	58.3[b]	30.7[a]	315.1[b]	375.8[b]
Local RG 1	11.3[b]	64.7[c]	70.7[a]	30.2[a]	265.1[c]	319.1[c]
Local RG 2	9.3[b]	66.3[c]	56.2[c]	20.9[c]	308.8[b]	384.3[b]
Mean	18.8	79.8	56.3	27.2	328.2	395.8
Significance	***	***	***	***	***	***

*** Significant at P≤0.001, according to S.N.K test. Within each column, different letters indicate significant differences between the means.

Differences in the number of racemes per plant were observed also among genotypes. In the first cropping season, plants of Tunisian and Brazilian genotypes produced more racemes than both Sicilian genotypes, whereas in the second one, Brazilian genotype produced significantly higher number of racemes than the other genotypes. A wide variability for the number of racemes per plant has been reported in literature within different germplasm collections of castor (Woodend, 1993; Anjani, 2012). However, it is not possible to compare data from the second year of experiment with other results, since generally literature refers to annual crops of castor.

The number of fruits per raceme varied appreciably with genotype in the first growing season, with ‘Local RG 1’ and ‘Tunisia’ having, respectively, the highest (70.7) and the lowest (40.2) number of capsules (Table 1). These values considerably declined in the second growing season and variability within genotypes was reduced (from 30.7 capsules in ‘Brazil’ to 20.9 capsules in ‘Local RG 2’). The number of capsules per raceme was much greater than that reported for castor (Soratto et al., 2012). The low plant population adopted in the present experiment may have accounted for the high raceme productivity. Indeed, differently than the number of seeds per fruit, which has a high genetic heritability, the productivity of racemes is strongly influenced by the agronomic factors (Freire et al., 2007; Soratto et al., 2012).

Seed weight, which was on average the 20% higher in the second cropping season, varied with genotype. ‘Tunisia’ produced a significantly greater 1000-seed weight exceeding 500 g in the second cropping season (Table 1).

According to the variation in yield components, castor seed yield changed appreciably with genotype. However, differences were observed between the two cropping seasons. In the first year, cumulative yield was significantly higher (4.05 t ha^{-1}) in 'Tunisia', whilst for both Sicilian cultivars, that differently than 'Tunisia' and 'Brazil' did not produce further after the third harvest, yield did not reach 1.0 t ha^{-1} (Table 2). Average yield (1.8 t ha^{-1}) was within the range reported by Lavanya et al. (2012) for several castor genotypes.

Table 2. Seed yield, oil content and oil yield in four castor genotypes.

Genotype	Seed yield (t ha^{-1})		Seed oil content (%)		Oil yield (t ha^{-1})	
	2010	2011	2010	2011	2010	2011
Tunisia	4.05^{a}	7.35^{a}	44.80^{c}	44.60^{c}	1.81^{a}	3.28^{a}
Brazil	1.50^{b}	5.94^{b}	47.20^{b}	47.90^{ab}	0.71^{b}	2.85^{b}
Local RG 1	0.89^{c}	2.95^{d}	47.50^{b}	47.30^{b}	0.42^{c}	1.40^{d}
Local RG 2	0.72^{c}	3.56^{c}	48.60^{a}	48.40^{a}	0.35^{c}	1.72^{c}
Mean	1.79	4.95	47.02	47.05	0.82	2.31
Significance	***	***	***	***	***	***

*** Significant at P≤0.001, according to S.N.K test. Within each column, different letters indicate significant differences between the means.

In 2011, an overall increase of yield levels was observed. As in 2010, 'Tunisia' was the most productive genotype, with a total yield exceeding 7.0 t ha^{-1}. Less productive were both Sicilian genotypes, whose total yield was anyway close (in 'Local RG 1') or higher (in 'Local RG 2') than 3.0 t ha^{-1}.

Yields obtained in the second year of experiment were within those reported by Koutroubas et al. (1999) with annual cultivar of castor (2.5 to 5.0 t ha^{-1}), although the authors adopted a greater plant population (5 plants m^{-2}). These yield levels are higher compared to those reported for castor. The productive increase in the second year was attributable to both a raised number of racemes per plant (4-fold greater than previous year) and a +20% increased 1000-seed weight that more than fully compensated for the reduced number of fruits per raceme (2-fold lower than previous year). Therefore, the contribution of each component to final seed yield was not stable across years. Higher seed weights greatly accounted for higher yield achieved in both years by the Tunisian genotype. Differently, lower seed yields obtained with the two Sicilian genotypes seem to be attributable to lower number of racemes per plant, which is considered a key trait in the yield of castor (Severino et al., 2010).

Oil content in castor seeds is a trait with high heritability (Soratto et al., 2012). It was 47.0% on average of cultivars and did not change between cropping seasons (Table 2). These results are in agreement with those of Mutlu and Meier (2010) who reported a low variability in oil content of castor across years and locations, but in disagreement with those of Koutroubas et al. (1999), who observed variations of this trait between years. However, oil content was significantly different among genotypes, being the lowest in seeds of Tunisian genotype (44.7% as average of the two years) and the highest in seeds of 'Local RG 2' genotype (>48%). Average seed oil content was close to that observed in cultivars of castor cultivated in central Italy (Laureti et al., 1998), but lower than those attained in a Mediterranean-type area of Greece (49-52%) by Koutroubas et al. (1999).

Oil yield was 0.82 and 2.31 t ha^{-1}, on average, in 2010 and 2011, respectively. It differed significantly with genotype, reflecting seed yield variations. According to Koutroubas et al. (1999), the differences in seed yield, greater than those in seed oil content, accounted for genetic differences in oil yield. The genotypes ranking for oil yield was the same as that for seed yield. Therefore, in both cropping seasons the Tunisian genotype was the most productive in terms of oil yield, although its seeds had low oil content. Contrastingly, both Sicilian genotypes, although with higher seed oil content, provided low oil yields in both years.

Oil quality

The quality of a lipid matter is essentially determined by the FAs composition of TAGs. Meteorological differences between the two growing seasons, although not particularly relevant, did not affect fatty acid composition, which thus seems to be more strictly genetic-dependent than environmental-dependent. The profile of FAs of the oil extracted from the seeds of the four castor genotypes is shown in table 3. Ricinoleic acid (C18:1-OH), typical of castor seeds oil, is quantitatively predominant, although an appreciable variability was found among genotypes. The other prevailing compounds were the five common palmitic (C16:0), stearic (C18:0), oleic (C18:1), linoleic (C18:2) and linolenic (C18:3) FAs. Lignoceric (C24:0) and eicosenoic (C20:1) acids were also found as reported for other cultivars of castor originating from Israel (Berman et al., 2011) (Table 3).

Table 3. Concentration of the major FAs and FAs categories (%) of extracted oil in four castor genotypes. Within each row, different letters indicate significant differences between the means. (U= unsaturated; S= saturated; M= mono; P= poly).

Fatty acid	Genotype				Mean	Significance
	Tunisia	Brazil	Local RG 1	Local RG 2		
Palmitic C16:0	0.90[c]	1.51[b]	1.34[b]	1.85[a]	1.40	**
Stearic C18:0	1.03[b]	1.35[a]	1.36[a]	1.32[a]	1.27	**
Oleic C18:1	2.90[c]	4.50[b]	4.55[ab]	4.82[a]	4.19	**
Ricinoleic C18:1-OH	89.00[a]	79.80[b]	79.60[b]	78.77[c]	81.79	**
Linoleic C18:2	4.50[b]	5.50[a]	5.49[a]	5.72[a]	5.30	**
Linolenic C18:3	0.64[a]	0.40[b]	0.42[b]	0.43[b]	0.47	**
Eicosenoic C20:1	0.00[b]	0.40[a]	0.41[a]	0.38[a]	0.40	**
Lignoceric C24:0	0.00[b]	0.08[a]	0.10[a]	0.09[a]	0.09	**
SFAs	1.93[c]	2.94[b]	2.80[b]	3.26[a]	2.73	**
UFAs	97.04[a]	90.60[b]	90.47[b]	90.12[c]	92.06	**
MUFAs	91.90[a]	84.70[b]	84.56[b]	83.97[c]	86.28	**
MUFAs (no Ricinoleic)	2.90[c]	4.90[b]	4.96[ab]	5.20[a]	4.49	**
PUFAs	5.14[c]	5.90[b]	5.91[b]	6.15[a]	5.78	**

** Significant at $P \leq 0.01$ according to S.N.K test.

The two SFAs palmitic and stearic, together accounted for an average 2.67% of total FAs, but genetic differences were observed, being the Tunisian genotype the poorest (1.93%) and ‘Local RG 2’ the richest (3.17%), owing to the appreciable difference in the concentration of palmitic FA. Both SFAs were found in higher amounts than those reported by Berman et al. (2011) for castor oil. Anyway, the amount of total SFAs reflects also the variation in the concentration of lignoceric acid, which was not detected in ‘Tunisia’ oil.

Ricinoleic acid is a particular MUFA with hydroxyl functionality that generally accounts for almost 90% in castor oil. ‘Tunisia’ oil exhibited significantly higher concentration of ricinoleic acid, which was 12% greater than that of the oil of the other three genotypes. These last had a ricinoleic acid content in the oil (79.4%, on average) much lower than that generally reported for the species in the literature. This result reveals how the differences in FAs composition including ricinoleic acid are mainly due to genetic variability (Berman et al., 2011; Hincapié et al., 2011; Ramanjaneyulua et al., 2013). This hydroxy FA in high concentration increases viscosity and enhances lubricity (Goodrum and Geller, 2005; Scholz and da Silva, 2008; Madankara et al., 2013), making the castor oil suitable for bio lubricants and several industrial applications (Pazir and Muhammad, 1991); however, it limits the use of the oil as source of biofuel

(Berman et al., 2011). Nevertheless, an inverse relationship between the ricinoleic acid concentration and seed oil content was observed ($r = -0.959$, d.f.=2, $P \leq 0.001$).

The level of MUFAs including ricinoleic reflects the differences observed among genotypes for this peculiar compound. When this FA was not considered, the concentration of MUFAs differed as a function of genotype, but the greatest quantity (5.20%) was attained by 'Local RG 2', rather close to that of 'Local RG 1' and 'Brazil' (4.93%, on average). 'Tunisia' oil had a low MUFAs amount when ricinoleic acid is not considered, due to the significantly lower proportion of oleic acid, which was found in concentration much below than that reported in literature for castor oil (Berman et al., 2011; Farias et al., 2011; Hincapié et al., 2011), as well as to the absence of eicosenoic acid. Greater concentration of oleic acid was found in the oil of both Sicilian genotypes.

According to Rojas et al. (2004) and Lavanya et al. (2012), the variations in the concentrations of oleic and ricinoleic FAs across genotypes were complementary as reveals their inverse correlation ($r = -0.998$, d.f.=2, $P \leq 0.001$). It is known that during the castor seed lipidogenesis, oleic acid is synthesized in the plastids and then exported to the cytoplasm following the standard FA pathway. Subsequently, this MUFA is activated to oleoyl-CoA in the cytoplasm and transferred into the endoplasmic reticulum by coenzyme A, where it is hydroxylated by the oleate hydroxylase to form ricinoleic acid (Somerville et al., 2000). Moreover, Velasco et al. (2005) observed that in natural castor mutants for high oleic acid, ricinoleic acid is partially replaced by oleic acid confirming a possible competition between the two MUFAs for the same substrate involved in their biosynthesis.

Castor oil with high oleic acid and low ricinoleic acid concentration (high O/R ratio) could be more suitable as biofuel, as the monounsaturated FAMEs (methyl oleate) (except ricinoleic acid which has a hydroxyl group) enhance certain qualitative indexes of the fuel (iodine value and cetane number). To this regard, due to its lower O/R ratio, the oil of 'Tunisia' is the least suitable to this purpose, but its richness in ricinoleic acid and thus its high viscosity makes it useful as a component to blending lubricants and other industrial applications as textile finishing, coating, inks, cosmetics, anti-fungal and making soaps (Caupin, 1997; Rojas et al., 2004; Ogunniyi, 2006; Madankara et al., 2013). On the other hand, oxidation stability greatly affects the use of biodiesel because of its content in polyunsaturated methyl esters as well (Knothe, 2006). This index decreases with the increase of PUFAs and therefore seed oils having a low proportion of PUFAs are the most appropriated for biodiesel production (Ramos et al., 2009). PUFAs level was lower in 'Tunisia' oil, mainly owing to the low percentage in linoleic acid, which is the prevailing PUFA in castor seed oil. There was a positive correlation between the concentration of oleic acid and that of linoleic acid ($r=0.999$, d.f.=2, $P\leq0.001$), which is not coherent with findings on other oil crops, such as sunflower, where an inverse correlation between oleic and linoleic acids in the oil has been reported (Anastasi et al., 2000). Greater percentage of PUFAs was in oil of 'Local RG 2', although it is partially compensated by the higher proportion of MUFAs not considering ricinoleic acid, resulting in a slight increase of MUFAs/PUFAs ratio, which makes the oil more resistant to oxidation (Curt et al., 2002) (Table 4). Conversely, the oil of the Tunisian accession was poor in PUFAs, but had significantly lower MUFAs (again not considering ricinoleic acid) and thus low MUFAs/PUFAs ratio. The O/L ratio, also known as unsaturation ratio and generally considered as a valuable oil stability index in vegetable oils (Lavanya et al., 2012), was also taken into account. In this experiment, the greatest unsaturation ratio in 'Local RG 2' confirms the potential suitability of this oil for biodiesel, among all castor genotypes evaluated. Similar values of O/L ratio were evidenced in oil of 'Local RG 1' and 'Brazil' genotypes that, therefore, could be appropriate for biofuel as well. These two genotypes exhibited a similar FAs profile, although originating from different geographic areas. Contrastingly, the oil provided by the seeds of Tunisian genotype, with lower oleic acid concentration and O/L ratio, is less appropriate for the use as fuel (Lavanya et al., 2012). According to Lavanya et al. (2012), O/L and O/R ratios were found to be positively correlated across genotypes ($r=0.998$, d.f.=2, $P\leq0.001$).

Table 4. Ratio between FAs and FAs categories, saponification number (mg K_2O g oil^{-1}), iodine value (I_2 100 g oil^{-1}) and cetane number of extracted oil in four castor genotypes (U= unsaturated; S= saturated; M= mono; P= poly).

Ratio/Index	Genotype			
	Tunisia	Brazil	Local RG 1	Local RG 2
Oleic/Linoleic (O/L)	0.64	0.82	0.83	0.84
Oleic/Ricinoleic (O/R)	0.03	0.06	0.06	0.06
UFAs/SFAs	50.28	30.82	32.31	27.64
MUFAs/PUFAs	17.88	14.36	14.31	13.65
MUFAs (no ricinoleic)/PUFAs	0.56	0.83	0.84	0.85
Saponification number (SN)	188.12	177.45	176.87	177.40
Iodine value (IV)	88.25	83.43	83.35	83.32
Cetane number (CN)	55.46	58.29	58.40	58.32

Other critical indexes than types and relative amounts of FAs and their ratios, have been defined to characterize oils, some of which (Saponification number, Iodine value and Cetane number) can be empirically estimated. In particular, the Saponification number (SN) provides information about the nature of FAs in terms of chain length and average molecular weight. The long chain compounds confer low SN due to a relatively fewer number of carboxylic functional groups per unit mass of the oil and consequently high molecular weight. The Iodine Value (IV) is a direct expression of its degree of unsaturation. Typically, drying oils have a high degree of unsaturation, which corresponds to IV ranging from 140 to 180, while non-drying oils containing up to mono-unsaturated Fas, have IV below 100. In the case of solvent-extracted castor oil, Ogunniyi (2006) indicates the range of 177-182 and 80-88 for SN and IV, respectively. Cetane number (CN), similarly to Octane number, is an expression of the autoignition resistance of a fuel. Higher CN means more complete combustion of the fuel and thus greater performance of engine (e.g. good cold start, low formation of white smoke) and vice versa.

The EU, within the standard for biodiesels (UNE-EN 14214 2003), established the thresholds for IV (120 max) and CN (51 min).

The oil of the two Sicilian genotypes as well as that of ‘Brazil’ had similar lower levels of either SN and IV (177.24 and 83.37, respectively) compared to those of ‘Tunisia’ oil (188.12 and 88.25, respectively). CN was lower in the oil of the Tunisian genotype (55.46) in comparison to those observed in the oil of the other genotypes, which were broadly similar (58.34, on average). Anyway, the values of all three oil quality indexes, although with some differences among genotypes which reflect the variation in FAs profile, are in agreement with those reported in the literature for castor oil (Ogunniyi, 2006; Lavanya et al., 2012) and specifically IV and CN satisfy the above-mentioned biodiesel standard imposed by the EU. Ramos et al. (2009) proposed a triangular graph to evaluate the quality of biodiesel obtained from vegetable oils, with the aim of grouping the fuels similar for methyl esters composition (FAMEs). The three angular points of graph correspond to 100% of monounsaturated, polyunsaturated and saturated (FAMEs), respectively. In particular, the green colored area of the triangle corresponding to the intersection of the two areas of high concentration (from 0.8 to 1) of MUFAMEs and low concentration of PUFAMEs (from 0 to 0.3), satisfies the limits imposed by UNE-EN 14214 (2003) for biodiesels having an overall better quality. When oil FAs (not considering ricinoleic acid, which accounts for over 78% of total FAs) are considered, none of the four studied genotypes fall within this optimal area of the triangular graph. However, transesterification decreases the viscosity associated to ricinoleic acid. The oil of all genotypes assessed may be included within the blue area, which satisfies the limit of cold filter plugging point (CFPP), having low SFAMEs (<20%). CFPP indicates the low-temperature filterability of a fuel that may develop operability problems in relation to the plugging of filters, when overnight temperatures approache -10 and -15 °C (Dunn and Bagby, 1996). CFPP depends on the content in FAMEs of long carbon chain SFAs, such as lignoceric acid. The longer the carbon chains of FAMEs in the biodiesel, the worse the low-temperature properties (Wu et al., 2005).

Conclusion

The results of the present study indicate that castor grows well under mild climate conditions of coastal areas of Sicily even with no irrigation input, shifting the growing season in autumn-spring period by bringing forward sowing time in early autumn. Indeed, in this period soil temperatures are still satisfactory for seed germination and seedling establishment and the crop can benefit from rainfall in winter time, escaping the drought conditions occurring in late spring and summer, with advantageous agronomic effects. The yield levels achieved under rainfed regime suggest that castor cultivation could be valuable in certain semiarid prone environments of southern Italy. Among the studied genotypes, 'Tunisia' was found to be the most productive in terms of oil yield, although it produced seeds lower in oil content. This last conformed to the EU standards EN 14214 for the main critical indexes, specifically iodine value and cetane number used to qualify the lipid raw-material for biodiesel production, but its high ricinoleic acid content makes this oil more appropriate for other industrial products as textile finishing, coating, inks, cosmetics, anti-fungal and making soaps. The Brazilian and the two Sicilian genotypes were less productive in terms of seed yield, but provided seeds richer in oil than 'Tunisia'. This oil satisfies the above-mentioned biodiesel EU standards in having a low concentration in ricinoleic acid, being thus more appropriate for biodiesel production.

The existence of genetic diversity in terms of fatty acid composition and the stability of this last to changeable environmental conditions, suggest the possibility to select within the castor germplasm of different origin as a source of quality traits to be used in breeding programs, according to the different industrial applications. Moreover, the existence of a wide germplasm of perennial castor throughout the Mediterranean area allows to exploit the possibility to breed for low temperature tolerance of the plant and for more synchronous seed ripening as well, this last in order to reduce costs of harvest.

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