Journal of Arid Environments 77 (2012) 153–156

Contents lists available at SciVerse ScienceDirect

Journal of Arid Environments

journal homepage: www.elsevier.com/locate/jaridenv

Short communication

Evaluation of *Jatropha macrocarpa* as an oil crop for biodiesel production in arid lands of the Dry Chaco, Argentina

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ARTICLE INFO

Article history:
Received 13 October 2010
Received in revised form
20 April 2011
Accepted 24 August 2011
Available online 21 September 2011

Keywords:
New oil crops
Fruit ripening
Plant productivity
Fatty acid
Perennial crops

ABSTRACT

Jatropha macrocarpa grows naturally in arid environments of the Dry Chaco, northwestern Argentina, where freezing winters make impossible the cultivation of other species of the genus *Jatropha*. This article is the first report of *J. macrocarpa* seed productivity aimed to quantify the reproductive performance and seed oil characteristics at different stages of fruit ripening. In addition, *J. macrocarpa* is evaluated as a new oil crop for biodiesel. Fruits from natural populations near La Rioja city were hand-harvested during the 2007/8 reproductive season and classified according to 7 fruit sizes, from the smallest fruits (G1) to the largest (G7 or mature). Total seed production averaged 87 $\pm$ 8 g plant^{-1} throughout the growing season. Explosive dehiscence of mature fruits produced 46% of total seed loss. Mature fruits produced individual seeds of 550 $\pm$ 7 mg with a medium-to-high seed oil concentration ranging from 36 to 40%, and biomass allocation to seeds was 36% of total fruit biomass. Fruits harvested early, at the G6 stage, were lighter and had a significantly less oil concentration. Main fatty acids in mature seeds included linoleic (50%), oleic (32%), palmitic (8.5%) and stearic (7%) acids. Biodiesel quality parameters indicated only slight differences for cetane number and iodine value with respect to European normative requirements. The acceptable oil content of *J. macrocarpa* shows promise to be used for biodiesel production. However, the low productivity per plant and the explosive fruit dehiscence need to be corrected substantially by breeding.

1. Introduction

Since the world biodiesel demand has increased due to governmental decisions aiming to reduce greenhouse gas emissions and dependence on foreign petroleum, the interest for new oil crops able to grow in marginal lands has increased markedly. One of the most promising new oil crops currently considered is *Jatropha curcas* L. (Euphorbiaceae) due to the expected high seed yields, with high oil concentrations and with an oil quality that reaches the international biodiesel standards (Achten et al., 2008). However, *J. curcas* does not seem to be the most appropriate alternative for arid areas with freezing temperatures due to the severe injuries caused by temperatures lower than -4 °C (Andrade et al., 2008) and the low yields obtained with less than 700 mm of annual rainfall (Achten et al., 2008).

For arid lands with freezing winters, *Jatropha macrocarpa* Griseb seems an interesting alternative because its natural area of distribution knows such climatic conditions. *J. macrocarpa* is native to South America, distributed around arid areas of the Chaco Region, called Dry Chaco in Argentina, Chaco Serrano in Bolivia and Chaco Boreal in Paraguay (Fernández Casas and Pizarro Dominguez, 2007). It is a big shrub (up to 2 m tall), with thick and succulent branches with abundant latex. The fruit is a capsule composed of a green exocarp (which remains fresh until fruit maturity), a hard endocarp (1–1.5 mm thick) and seeds (typically three per fruit). Mature fruits have an explosive dehiscence, which causes the fruit split into three parts that separate from the central axis or columella, which is persistent (Lourteig and O'Donell, 1943). Flowering and fructification occur from late spring to summer. Information about *J. macrocarpa* is scarce and mainly refers to botanical aspects, so an evaluation of its productive potential is necessary.

The objectives of the present study were to evaluate the potential of a natural population of *J. macrocarpa* for biodiesel production in arid lands with freezing winters, through: (i) the

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doi:10.1016/j.jaridenv.2011.08.011

determination of seed mass, oil concentration and composition during fruit ripening, (ii) evaluation of the reproductive performance during the growing season and (iii) estimation of the oil aptitude for biodiesel production.

2. Methods

Two experiments were carried out during the growing season of 2007/2008 (November to February) in a wild population of *J. macrocarpa* located 30 km south of La Rioja city, Argentina. The population was composed mainly by *J. macrocarpa* shrubs, scattered across an area of about 1000 m^2 with a naturally established plant density of around 2500 plants ha^{-1}. Exp. 1 was performed to characterize the evolution of seed weight, oil concentration and composition of fruits at different stages of ripening. Fruits harvested on 12/12/07 were grouped in seven categories according to their size and color (from the smallest and green G1 to fully ripe brown fruits G7; Fig. 1 and Appendix 1, electronic version only). Three samples were obtained per category from different shrubs with similar heights ($\sim$1.80 m). The seeds could only be analyzed from the G3 state onward, since the seeds from G1 and G2 fruits were too small and morphologically incomplete. In Exp. 2, the seasonal seed production and the seed losses by dehiscence were determined, as well as the feasibility to harvest in advance at the G6 stage, when the green fruit is nearly mature. A systematic fruit harvest was conducted at three different moments (11/27/07; 01/08/08 and 02/20/08) on ten shrubs of similar heights (1.80 m). Mature fruits (G7) and yellowish green fruits (G6) were harvested at each moment. Due to the fruits' explosive dehiscence, the estimation of the total seed production was carried out by counting the number of columellas present in the racemes, assuming that each dehiscent fruit had three seeds with the same seed dry weight that was found in the harvested mature fruits. The harvested fruits (G7 and G6 stages) were separated into their components (exocarp, endocarp and seeds), oven dried at 70 °C until constant weight, and weighed. Biomass allocation to seeds was determined according to Vilela et al. (2008). Seed oil concentration was determined by the Soxhlet extraction method with hexane for 12 h and fatty acid composition was determined by gas-liquid chromatography according to Rondanini et al. (2003). Iodine value (IV) and cetane number (CN) were estimated from the fatty acid composition as relevant parameters for biodiesel quality. The IV indicates the level of oil unsaturation and was estimated using equations published by Kalayasiri et al. (1996). The CN indicates the ignition speed of biodiesel after injection and was calculated using the equation published by Krisnangkura (1986). Treatments were arranged in a completely randomized design and the data was analyzed by one-way ANOVA and LSD were considered at the 5% significance level.

Meteorological data were obtained from the airport of La Rioja, 30 km away from the experimental site. Seasonal (from November 2007 to February 2008) mean temperature was 25.7 ± 1.4 °C and rainfall was 534 mm, twofold greater than the historical average for this period (260 mm; SMN, 2003, see Appendix 2 and 3, electronic version only).

3. Results

During grain filling, individual seed dry weight was similar throughout fruit stages G3 to G5; the highest weight gains were observed after the G5 stage (Fig. 1). Seed oil concentration increased steadily from stages G3 to G5. After G5, oil concentrations jumped to more than 30% and kept rising to a final 36% at G7 (Fig. 1).

Main fatty acids present in mature seed oil (stage G7) were linoleic (50.3 ± 0.6%) and oleic (32.1 ± 0.5%), followed by a minor proportion of palmitic (8.5 ± 0.2%) and stearic (6.9 ± 0.1%). Altogether, these four fatty acids represented 97.8% of the total, with 83.5% corresponding to unsaturated fatty acids. Few changes in oil composition took place between G3 and G5. During the last filling stages (G5 to G7), an important increase of oleic and linoleic proportions was observed at the expense of a reduction in palmitic acid (Fig. 2). The estimated biodiesel quality parameters changed during fruit filling. The CN value tended to decrease slightly from 49 at G3 to 45 in G6 and, conversely, IV tended to increase from 106 at G3 to 125 at G6. In mature seeds, the CN was 46.1 and the IV was 121.6.

In Exp. 2, fruits were not collected on the first harvest date (11/27/07) because plants were at the beginning of flowering. On the

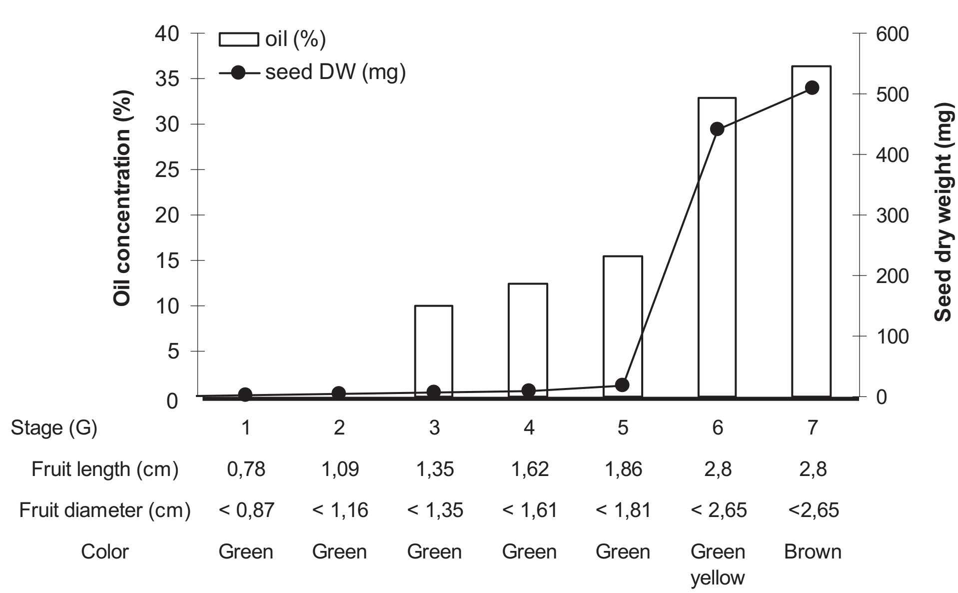

Fig. 1. Individual dry weight and oil concentration (%) in seeds obtained at different fruit ripening stages (Exp. 2) from a natural population of *J. macrocarpa* from the Dry Chaco region.

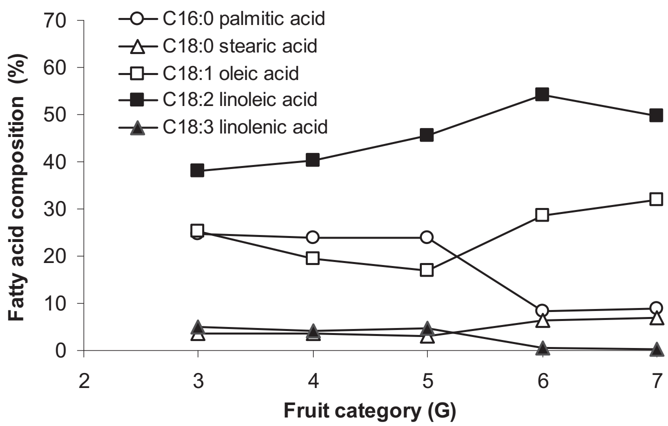

Fig. 2. Fatty acid composition in oil seeds at different ripening stages (Exp. 2) from a natural population *of J. macrocarpa* from the Dry Chaco region.

second harvest date (01/08/08), only fruits at the G6 stage were collected and the seed loss by dehiscence was estimated counting the columella numbers. A new flowering pulse was observed on this date, together with fruits at different maturity stages (G1 to G5) that remained on the shrub, but no mature fruits were found (G7). On the third harvest date (02/20/08), G7 and G6 fruits were collected, and seed loss by dehiscence was estimated. Adding up the two harvests, the total mean seed production per plant was 87.0 ± 7.8 g, with a range from 35 to 150 g (Fig. 3). Seed production on the second harvest date represented 60% of the total seed production per plant, considering G7, G6 fruits and fallen seeds, while seeds on the third harvest date contributed with the remaining 40% (Fig. 3). The seed loss due to dehiscence was very important, representing 47.7% of the total plant production and, conversely, the proportion of mature fruits was very low (5.5% of total seed production, Fig. 3). Biomass allocation in G6 and G7 fruits were compared on the third harvest date. Biomass allocated toward seeds was significantly lower ($p < 0.01$) in G6 fruits (13.6 ± 2.2%) compared with G7 fruits (36.1 ± 0.8%), while allocation toward the exocarp (16.2 ± 0.3% at G6 and 13.0 ± 0.1% at G7) and endocarp

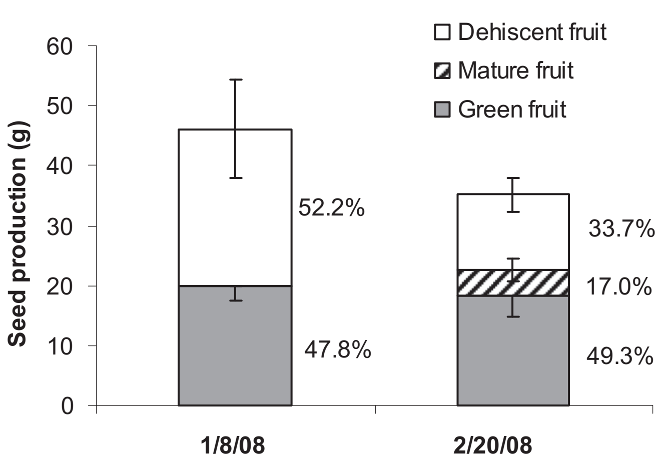

Fig. 3. Seed production of *J. macrocarpa* from a natural population (Exp.1) in the Dry Chaco region for two harvest dates. Mature fruits (G7 stage), nearly ripe fruits (visually at the G6 stage) and seeds lost due to dehiscence were counted separately; percentages adjacent to bars indicate the contribution of each category to the total seed weight. Vertical lines represent standard errors.

(70.1 ± 2.0% at G6 and 50.9 ± 0.7% at G7) were significantly higher ($p < 0.01$) in G6 fruits.

Individual seed dry weight in G7 fruits was similar in both experiments (552 ± 7 and 510 ± 4 mg for Exp. 1 and 2, respectively), providing a range of maximum seed dry weights for *J. macrocarpa* growing in this environment. In contrast, seeds from G6 fruits were different among experiments, with an individual seed weight of 442 ± 35 mg in Exp. 1 and 291 ± 23 mg in Exp. 2. This difference could be explained by the difficulty to differentiate between fruits at the G6 and G5 stages during the manual harvest in Exp. 2. Oil concentrations in seeds from mature fruits were similar in both experiments (40.1 ± 0.5% and 36.4 ± 0.7% in Exp. 1 and 2, respectively). In contrast, seed oil concentration from G6 fruits in Exp. 2 was significantly ($p < 0.01$) lower (20.6 ± 2.5%) than in seeds from G7 fruits, whereas in Exp. 1 little differences in oil concentration were found in seeds from both kind of fruits (Fig. 1).

4. Discussion

Seed production of the natural population of *J. macrocarpa* evaluated in this study was low and variable, with a mean seed production per plant of 87 g and a maximum value of 151 g, representing theoretical yields of 435 and 756 kg ha^{-1}, respectively (considering 5000 pl ha^{-1}, the plant density used in the first experimental plots). Furthermore, since rainfall in summer of 2008 doubled the historical average for La Rioja, lower seed productions could be expected in normal years.

The fruits of *J. macrocarpa* have explosive dehiscence when they mature, which implies a serious disadvantage. Possible alternatives are to breed for indehiscence or to anticipate the fruit harvest. However, our results show that an early harvest implies collecting seeds with half the oil concentration and individual weights compared to mature seeds, mainly due to the difficulties in visualizing the difference between G5 and G6 fruits. Although an early harvest seems the way to reduce seed loss due to dehiscence in the short term, it is necessary to find indehiscent and more productive genotypes through a broad genotype screening program to turn *J. macrocarpa* into an energetic crop for arid lands. The seed proportion in mature fruits of *J. macrocarpa* (36%) was lower than that found in *J. curcas* (78.6%; Gunaseelan, 2009). Therefore, we suggest that variability for this character should be explored because high biomass allocation to the exocarp and endocarp implies higher costs during harvest, transportation and seed processing.

Oil concentration in *J. macrocarpa* mature seeds found in this study were similar than the values reported by Riqué et al. (1963) for a wild *J. macrocarpa* population in La Rioja and similar to the highest oil concentration (39%) reported for *J. curcas* (Ginwal et al., 2004). The fatty acid composition in mature seeds of *J. macrocarpa* was similar to values reported by Riqué et al. (1963), suggesting low natural variability for this character. During seed filling, oil composition in seeds from G6 fruits was similar to that found in mature seeds, indicating that early harvest causes minor effects on oil quality. The estimated iodine value found in this study for mature seeds of *J. macrocarpa* was 121.6, similar to the 120.7 value published by Riqué et al. (1963), and very close to the limit established by the European normative (<120; Committee for Standardization, 2003). Different minimum values have been specified for the CN: 47 in the USA (ASTMD 6751) and 51 in the European Union (Committee for Standardization, 2003). The estimated CN value for *J. macrocarpa* oil was between 45 and 46.3 (seeds from G6 and G7 fruits, respectively), slightly lower than the minimum established by both standards, which represents a quality restriction for its use as a biodiesel feedstock.

The ability of *J. macrocarpa* to survive and produce seeds under arid conditions with freezing winters is the main reason to consider its domestication. The high seed oil concentration observed in this study is acceptable but oil quality should be improved in order to meet the European quality standard requirements for biodiesel. Undoubtedly, the main concerns for the use of *J. macrocarpa* as an energetic crop are related to low plant productivity (756 kg seed ha^{-1} in the most optimistic scenario), and to the explosive dehiscence of fruits. Both aspects should be substantially improved so that *J. macrocarpa* may have a chance to become a commercial crop, and the first logical step is to carry out a broad genotypes screening program to determine the natural variability for both characters.

Acknowledgments

We thank D. Fernádez-Abeijón and Eng. D. Castro for their technical assistance; Eng. L.M. Agost Carreño for carrying out the regional contact in La Rioja and Daniel Cerezo for his farm assistance. Biogreenoil Argentina SRL and ANPCyT (PICT 1617) provided financial support for this study.

Appendix. Supplementary material

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.jaridenv.2011.08.011.

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