



RESEARCH ARTICLE

Benefits of *Jatropha gossypifolia* in Nigeria

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ABSTRACT

The genus *Jatropha* is distributed throughout the tropics and sub-tropics growing in marginal lands and is a potential biodiesel crop worldwide. The plants can prevent soil erosion, grown as a live fence and used as an alternate commercial crop. The seed oil can be used as a feed stock for biodiesel. Alternatively *Jatropha* oil is used in soap, glue or dye industry. The seed cake is rich in nitrogen and phosphorus, and can be used as manure. All parts of the plant including seeds have medicinal properties. However, the toxic components of the seed need immediate attention and efforts are needed to genetically modify *Jatropha* seed toxins to useful and non-toxic components through genetic transformation. This review focuses some basic aspect of the distribution, diversity, biology, cultivation, tissue culture and genetic transformation of *Jatropha*.

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INTRODUCTION

The genus *Jatropha* has 175 known species of the plant belonging to the family Euphorbiaceae (Dehgan, 1984). Originating in the Caribbean, *Jatropha* was spread as a valuable hedge plant to Africa and Asia by Portuguese traders. The plants are drought-resistant succulent shrubs or trees, and are recognized as potential biofuel crop (Jones and Miller, 1991; Openshaw, 2000). *J. curcas*, *J. integerrima*, and *J. glandulifera* are native to South America. Among the various *Jatropha* species *Jatropha curcas*, *J. glandulifera*, *J. gossypifolia*, *J. integerrima*, *J. multifida*, *J. nana*, *J. podagrica*, and *J. tanjorensis* are widely cultivated naturalised and distributed in India. The term *Jatropha* is usually and commonly used to refer to the species *J. curcas*. This plant is a deciduous perennial shrub distributed all over the world. The name is derived from Greek words "iatros" meaning physician and trophē meaning nutrition. So the plant is commonly known as physic nut. *J. cuneata* stems are often used for basket making in Mexico. *J. nana* is a small bushy ephemeral and is endemic to Western India. It shows absence of glandular hairs. Seed oil from *J. nana* is used for energy. In traditional medicine it is used as anti-irritant in ophthalmia (Ambasta 1992; Das and Venkataiah 2000). *J. Gossypifolia* (Linn) is a bushy, gregarious shrub with reddish leaves and flowers and glandular hairs throughout of about 1.8 meters in height,

which grows widely in Nigeria. The leaves are 3-5 lobed, palmately, 20cm glandular hairs. The flowers are red-crimson or purple in corymbs, with greenish seeds in capsule. Different parts of *J. gossypifolia* are used in different countries in many ways. The leaves of *J. gossypifolia* are used for intermittent fevers, carbuncles, eczema, itches and sores on the tongues of babies, swollen mammae, stomachache, and venereal disease. The leaf decoction is used for bathing wounds. The bark contains the alkaloid jatrophine and a lignan (jatroiden) is found in its stem. Seeds are emetic, purgative, and used for body pain. Ethno botanical uses of *J. gossypifolia* reported for cancer, diarrhoea, dysentery, skin diseases (leprosy), arthritis, ulcer, gum infections and wound healing (Hussain *et al.*, 1992; Dash and Padhy 2006; Rajesh *et al.*, 2007). Seed oil of *J. gossypifolia* is used in rheumatism and paralytic affections (Annon 1965). *J. glandulifera* is a small bushy shrub found all over India and has greenish yellow flowers and green leaves. It has glandular hairs only in the axils of leaves. In ayurvedic literature, seeds, leaves, bark and roots were reported for its analgesic properties, inflammation, asthma and bronchitis (Ambasta, 1992; Das and Venkataiah, 2000). Biological activity of shoots reported against cancer (Hussain *et al.*, 1992). Out of these species, *J. curcas* is commonly referred to as *Jatropha* and is studied for its oil quality. Reviews are available on the literature covering mostly *J. curcas*. However, literature shows extensive studies on

other species also and is covered in this review along with that of *J. curcas*.

Occurrence, distribution and adaptation

The centre of origin of *J. curcas* is still debatable, but it is believed to be native of Mexico and Central American region. It has been introduced to Africa and Asia in the new world. It is cultivated in many parts of the tropics and subtropics as a hedge crop and for traditional use (Heller, 1996; Kumar and Sharma, 2008). *Jatropha* occurs mainly at lower altitudes (0-500 m) in areas with average annual temperatures well above 20°C but can grow at higher altitudes and tolerates slight frost. It grows on well-drained soils with good aeration and is well adapted to marginal soils with low nutrient content. The current distribution shows that introduction has been most successful in the drier regions of the tropics and can grow under a wide range of rainfall regimes from 250 to over 1200 mm per annum (Katwal and Soni, 2003). Water stress at lower intensity reduced the net photosynthesis without significantly affecting the photosynthetic structure. However, at higher stress level WUE, net photosynthesis and photochemical efficiency decreased significantly suggesting damage to the alteration in the photosynthetic structure, also (Dou et al. 2008). The photosynthetic structure and function recovered and the plants survived after the stress was removed. It was concluded that *Jatropha curcas* has a high drought tolerance capacity (Dou et al., 2008). However, Tezara et al. (2005) studied the seasonal changes in water relations, photosynthetic parameters of the C3 deciduous shrub *J. gossypifolia*, and reported a decrease in leaf water potential from, total Chl content of the leaf, net photosynthesis and the carboxylation efficiency and quantum yield of PS2, during drought. Photoinhibition during drought in *J. gossypifolia* was observed in the field which might impose a limitation on carbon assimilation during drought (Tezara et al., 2005). Taking this into account and the probability of the plant survival at stake under stress conditions in the field, a recent study field study was done by Das et al. (2010). This study showed that the plant can recover rapidly from mid-day depression of photosynthesis or the photo-inhibition without any significant change in leaf pigment contents, suggesting that *Jatropha* adapt to changing climatic conditions like high temperature, high light and low soil moisture levels of the mid-day in the tropics by efficient adaptation of photosynthesis in the field (Das et al., 2010).

Salt stress affects almost all parts of the globe (Misra et al., 2001, 2006). Although this species had shown satisfactory yield under arid and semiarid regions, the physiological mechanisms controlling its salt stress tolerance are poorly understood. As *Jatropha* is grown in marginal and problem soils, it should have better adaptability to salinity. Gao et al. (2008) reported salinity induced decrease in the biomass, and enzyme activities of superoxide dismutase, peroxidase, catalase, and phenylalanine ammonia-lyase in cotyledons, hypocotyls and radicles of *Jatropha curcas* L. seedlings. Photosynthetic machinery is the most sensitive system to stress (Misra et al., 2001, 2006). Da Silva et al. (2010) studied the changes in photosynthetic adaptability of *J.*

curcas in liquid culture medium to increased salinity. These authors showed that photosynthesis was reduced with both salinity and period of saline stress.

Fujimaki and Kikuchi (2010) evaluated the tolerances of *Jatropha* to drought and salinity stresses and concluded that the stress response functions indicate that *Jatropha* is not more tolerant either to drought or to salinity compared to other major crop such as soybean or wheat (Fujimaki and Kikuchi, 2010). Temperature is an important aspect of climate. *Jatropha* can be grown in wide range of soils in tropical and subtropical parts of the globe with a temperature average of more than 20°C. It can be grown well on degraded soils having low fertility and moisture and also on stony, gravelly or shallow and even on calcareous soils. For economic returns, a soil with moderate fertility is preferred. The emergence of seed required hot and humid climate. It can be cultivated successfully in the regions having scanty to heavy rainfall. Marginal areas of semiarid regions frequently subjected to dry and hot conditions, where most other crops are not able to survive (Francis et al., 2005). However, *Jatropha* grows well under this climate. This species may have potential as an excellent model for the physiological and molecular mechanisms involved with plant resistance to combined abiotic stresses. Silva et al. (2010) evaluated the photosynthetic and antioxidative response of *J. curcas* leaves subjected to isolated and combined drought and heat stresses. Drought significantly retarded photosynthesis and accelerated oxidative metabolism and these negative effects are augmented by heat stress. A combination of heat and drought stress triggers a complex response involving antagonistic and synergistic interactions (Silva et al., 2010). This study showed that *J. curcas* plants did not show protection against drought-induced oxidative stress, especially at high temperatures (Silva et al., 2010). *J. curcas* is relatively vulnerable to low temperatures (chilling stress) especially at the seedling stage (Liang et al., 2007), limiting its cultivation in cold areas such as high mountains. Chilling tolerance varies among plant populations. Zheng et al. (2009) studied the effect of sub lethal chilling on 12 Chinese populations of *J. curcas* to evaluate the effects of chilling on photosynthesis of *J. curcas* and intraspecific differences in chilling tolerance.

Night chilling treatment decreased light-saturated photosynthetic rate significantly for all populations. Conducting the recovery studies for few days, they reported that *J. curcas* was vulnerable to chilling. Chilling tolerance was significantly different among populations. Populations originating from high altitudes had greater chilling-tolerant abilities than populations originating from low altitude. These give variability in the possibility to breed chilling tolerant genotypes of *J. curcas* for growth and cultivation in higher altitudes and in hill tracts (Zheng et al., 2009).

Jatropha cultivation and yield

Jatropha can be grown in marginal wastelands due to its ability to adapt to adverse agroclimatic conditions. It has an estimated annual production potential of 200,000 metric tonnes in India (Tiwari et al., 2007). *Jatropha curcas* can be established from seed, seedlings and cuttings. However, conventional propagation of *Jatropha*

is limited by poor seed germination, scanty and delayed rooting of seedlings and vegetative cuttings (Heller, 1996; Openshaw, 2000; Purkayastha *et al.*, 2010). The best time for planting is in the warm season before or at the onset of the rains. In the former case, watering of the plants is required. The recommended spacing for plantation is 2-3 m by 1.5-3 m for plantations (Jones and Miller, 1992). The number of trees per hectare at planting may range from 1100 to 3300. Growth of the plants is dependent on soil fertility and rainfall, especially the later. Flower and seed production respond to rainfall and nutrients. A poor nutrient level will lead to increased failure of seed development (Gubitz *et al.*, 1999). Like all perennial plants, it displays vigorous growth in youth; this will tail off gradually towards maturity.

Management of jatropha requires the addition of manure and NPK to the planting hole at 2kg compost, 20g urea, 120g SSP and 16 g MOP and urea should be applied in two splits (one and two months after transplanting) at 10 g per plant (Singh *et al.*, 1996). Yearly top dressings of fertilizers including the seed cake should be done. *Jatropha curcas* begins bearing seeds within nine months, reaches commercial productivity in three years and lives for up to 50 years. The potential seed yield of jatropha after five years is 6 tons to as high as 12 tons per hectare depending on the site, climate and management of the plants. The seed yields reported for different countries and regions range from 0.1 to 15 t/ha/y (Heller 1996; Jones and Miller, 1993). Henning (2004) reported that some 18-month-old trees were producing approximately 2.5 kg of seed per tree. In the literature, the reports of yields vary greatly and are confusing. This can be attributed to one or a combination of the following factors including: yields are sometimes given in terms of fruits, seeds, nuts, or kernels; variance in germplasm; unstipulated spacing between plants; no specific data on soils (ranging from marginal to fertile, and if fertilizer was applied); no information on rainfall and other climatic conditions, and if irrigation is being used. Seeds can usually be harvested one year after planting. It is best to harvest the fruits when these have turned yellow to dark brown. Approximately two to three months after flowering, seeds should be collected when the capsules have split open. The seed yield of dried fruits is reported to be approximately 60% w/w basis. There is a wide variation in the seed size of *Jatropha* species. The seed size *Jatropha curcas* is measured as length x breadth x thickness (L x B x T) are 1.69 x 1.4 x 0.84 – 1.84 x 1.31 x 0.85 mm with black color at maturity. The seed size of *J. nana* is 0.87 x 0.47 x 0.35 mm with yellowish caurancle and brown coat colour. *J. gossypifolia* has small brown seeds with stripes; size of seed is 0.74 x 0.44 x 0.35 mm. Seeds of *J. glandulifera* are small, cream-yellow with black dots throughout. Size of seeds is 0.74 x 0.54 x 0.34 mm (Hussain *et al.* 1992).

One might assume that the fruit to seed ratio may be higher in areas of higher rainfall. One kilogram of jatropha seeds consists of 600 to 1,600 pieces of seeds. Seeds are de-hulled by using wooden plank and then winnowed to separate the hulls from the seeds. Before storing, the seeds must be air dried to 5% - 7% moisture content and stored in air-tight containers. Seeds can be stored up to one year at room temperature. Seeds for replanting can be gathered when fruits are already yellow

to dark brown. Seeds should not be dried in direct sunlight because it will affect its germination. Dry, black seeds can be used for oil extraction (Makkar *et al.*, 1998).

Commercial exploitation of *Jatropha curcas* plant and plant parts

Jatropha curcas is a promising species because many products from the plant can be made useful and profitable. The chemical composition of various parts of *Jatropha curcas* plant is as follows:

- i. Bark contains β -Amyrin, β -sitosterol and taraxerol.
- ii. Aerial parts contain o and p-coumaric acid, p-OH-benzoic acid, protocatechuic acid, resorsilic acid, saponins and tannins, β -Amyrin, β -sitosterol and taraxerol.
- iii. Leaves contain flavanoids such as apigenin, vitexin, isovitexin, atriterpene alcohol (C₆₃H₁₁₇O₉) and flavonoidal glycosides.
- iv. Seeds contain Curcin, lectin, phorbolsters, esterases and lipase.
- v. Roots contain β -sitosterol and its β -D-glucoside, marmesin, propacin, the curculathyrans A and B and the curcusones A-D, diterpenoids jatrophenol and jatrophenolone A and B, coumarin-tomentin, coumarino-lignan-jatrophin, taraxerol.
- vi. The seed kernel is rich in phytates, saponins and trypsin inhibitor, which also remain as a major component of the seed cake.

The bark, leaf and tender stems of *Jatropha curcas* yields a dark blue to blackish brown dye which is reported to be used for coloring cloth, fishing nets and lines. The plants and fruit hulls could be used for firewood. *Jatropha* wood is a very light wood and is not popular as a fuel wood source because it burns rapidly. Seed cake results very high-quality charcoal that has the potential to be used in high value markets. But press cake is much more valuable to use as a organic manure and increase crop production in marginal land (Kumar and Sharma 2008). The seed cake can be converted into biogas by digestion in biogas tanks together with other input materials, such as cow dung, leaves or night soil. The biogas can be used for cooking and lighting. The residue can still be used as organic fertilizer, as it retains all of its minerals and nutrients ((Makkar *et al.*, 1998; Kumar and Sharma 2008). The bark of *Jatropha* contains tannin. It can also have the honey production potential as the flowers can attract bees. *Jatropha* seeds contain toxic Phorbol esters and is toxic when consumed (Makkar *et al.*, 1998). However, a non-toxic variety of *Jatropha* exists in some parts of Mexico and Central America which can be used for human consumption after roasting the seeds/nuts (Delgado and Parado, 1989). The young leaves may be safely eaten, steamed or stewed. When grown from seeds, the plants are edible for the first 3 months, since the toxic material has not been developed yet. Levingston and Zamora (1983) report that *Jatropha* seeds are edible once the embryo has been removed; nevertheless on principal, consumption of seeds should be avoided. Non-toxic variety of *Jatropha* could be a potential source of oil for human consumption, and the seed cake can be a good protein source for humans as well as for livestock (Becker, 1996; Makkar and Becker, 1999; Aregheore, 2003).

Medicinal and insecticidal properties

Jatropha is known for its use as purgative/laxative and other medicinal uses. All parts of the plant, including seeds, leaves and bark, fresh or as a decoction, are used in traditional and folk medicine and veterinary purposes (Duke 1988). The methanol extract of *Jatropha* leaves are reported to have Beta blockers which has potential cardiovascular action in humans. The sap from the stem is used to cure the bleeding wounds. The latex of *Jatropha* contains several alkaloids viz Jatrophine, Jatropham and curcain with anti-cancer properties (Van den Berg et al. 1995; Thomas et al., 2008). The roots are reported as an antidote for snake-bites. The anti-inflammatory activity of *Jatropha curcas* L. root is reported by Mujumdar and Misra (2004). The oil is a strong purgative and is used in skin ailments and in rheumatism (Heller 1996; Marroquin et al. 1997). The 36% linoleic acid content in *Jatropha* kernel oil is of possible interest for skincare. Sporadic report on the possible use of *Jatropha* against HIV is also available. All parts of the plant show insecticidal properties (Grainge and Ahmed 1988;

Consoli et al., 1989; Jain and Trivedi 1997; Meshram et al., 1994) for example against insect/pests like cotton bollworm, and on pests of pulses, potato and corn (Kaushik and Kumar 2004). The stem latex of *Jatropha gossypifolia* is routinely used in Africa and Indian sub-continent by herbalists, rural dwellers and some people in urban centers to stop bleeding from nose, gum and skin. Coagulant activity of the stem latex of *Jatropha gossypifolia* was demonstrated by Oduola et al. (2005), who provided scientific basis for its use as a haemostatic agent.

Eco-restoration and controlling soil erosion

Jatropha curcas is capable of growing on marginal land, and helps to reclaim problematic lands and prevent soil erosion. The cultivation of *Jatropha* leads to the conservation of degraded lands, soil (eco-)restoration and management by preventing soil erosion, protects plants against wind erosion and the roots also form a protection against water erosion (Haller 1996). The plant also serves as protective fence around agricultural fields against live stocks influx and grazing. This is a low cost bio fence compared to wire fence and has the advantage of being easily be propagated by cuttings, densely planted or spaced and not fed by cattle.

Use of *Jatropha* oil as a biodiesel, cooking and soap industry

Jatropha curcas L. (Euphorbiaceae), a deciduous perennial shrub with Central America origin, is now widely cultivated in tropics and subtropics worldwide (Deore and Johnson, 2008). Seed oil content of this plant is about 40%, higher than the typical oil crops such as soybean and rape (Gubitz et al., 1999; Deore and Johnson, 2008). The oil can be used in diesel engines after simple processing because it is similar to diesel oil in characteristics, being a potential substitute for fossil fuel and a renewable energy (Berchmans and Hirata, 2008; Deore and Johnson, 2008). Thus, *J. curcas* has been considered as a strategic plant resource in many countries (Carvalho et al., 2008). The oil content in the whole seed is around 27-30%, de-coated seeds ranges from 35-40%

and the kernels 55-60% (www.jatropha.org). The *Jatropha* seed oil has a fatty acid composition similar to that of edible oils (Gubitz et al., 1999). Also, the seed oil can be used as a diesel engine fuel as it has characteristics close to those of fossil fuel, diesel. The composition and content of the *Jatropha* seeds. The plant and its seeds are toxic and so is not edible either by animals or humans. *Jatropha* plants contain several toxic compounds, including lectin, saponin, carcinogenic phorbol, and a trypsin inhibitor. As a result, in 2005 Western Australia banned *Jatropha gossypifolia* as invasive and highly toxic to people and animals. Despite this, the seeds are occasionally eaten in some parts of the world after roasting which reduces some of the toxicity. The sap is a skin irritant, and ingesting as few as three untreated seeds can be fatal to humans. As a result *Jatropha* is commonly used as hedges or fences worldwide to protect agricultural fields. In spite of all these facts, the nonedible oil of *Jatropha* seeds and its derivatives are also used for manufacturing a number of useful products, including candles, high quality soaps, cosmetics, biopesticide and fertilizer as well as for healing several skin disorders (Duke 1988). Variations in the oil content, physico-chemical analysis, FA and OSI contents of various species of *Jatropha* are reported by Bhagat and Kulkarni (2009). The oil content of the seed is within the range obtained for cotton and castor seed oils (Kumar and Sharma, 2008). Seed oil obtained from *J. nana* and *J. glandulifera* is yellow while that of *J. gossypifolia* is cream in colour. *J. nana* and *J. gossypifolia* remains in semi-drying and *J. gossypifolia* in non-drying state at room temperature. Nitrogen content in fatted cake is in range of 2.64% to 3.66% and defatted 4.19% to 6.83% and used as fertilizer. Crude protein is 24.60% in *J. curcas* (Ajam et al., 2005). Crude protein in fatted and defatted cake was in range of 16.51% to 25.67% and 26.16% to 42.66% respectively. OSI of *J. curcas* was 0.75 to 2.1 and is higher than other three species. Among the saturated fatty acids (SFA) palmitic (16:0) and stearic (18:0) acids present in all species at approximately 13%. While unsaturated fatty acid (UFA) like oleic acid (18:1) and linoleic acids were approximately 87%. These values compared with *J. curcas* FA. The FA profile showed similarities with many other oils of plant origin like *Madhuca indica*, *Pongamia pinnata*, *Euphorbia helioscopia* and *Mesua ferrea* (Kirschenbauer, 1965). *Jatropha* seed is usually toxic and so the alternative use of the seed to derive /extract oil is more appropriate and beneficial than simply exploiting the seed as a food crop.

Apart from its use as a biofuel, the *Jatropha* seed oil is reported to be used to produce soap, medicine and pesticides (Shanker and Dhyani, 2006). *Jatropha* oil can be used as a fuel formulation in diesel engines directly and by blending it with methanol (Gubitz et al., 1999). The clear seed oil was used as diesel substitute during the World War II (Agarwal and Agarwal, 2007) and these tests conducted at Thailand during WW II showed satisfactory engine performance (Takeda, 1982). The oil of *Jatropha curcas* is a viable alternative to diesel fuel since it has desirable physico-chemical and performance characteristics as commercial diesel. The oil after transesterification confirms the standard requirements of American and European countries for qualifying as a

biodiesel without much alteration in the engine design (Vinayak and Kanwarjit, 1991; Azam *et al.*, 2005; Tiwari *et al.*, 2007). Currently the oil from *Jatropha curcas* seeds is used for making biodiesel fuel in Philippines and in Brazil. Likewise, jatropha oil is being promoted as an easily grown bio fuel crop in hundreds of projects throughout India and other developing countries. The railway line between Mumbai and Delhi is planted with *Jatropha* and the train itself runs on 15-20% biodiesel. Seed yields under field cultivation can range from 15q/ha to 20q/ha, corresponding to extractable oil yields of 540 L/ha to 680 L/ha. Time Magazine recently cited the potential for as much as 1,600 gallons of diesel fuel per acre per year. *Jatropha* can also be intercropped with other cash crops such as coffee, sugar, fruits and vegetables. On Dec. 30, 2008 of the four Rolls-Royce RB211 engines of a 747 jumbo jet made a two-hour test flight that could mark another promising step for the airline industry to find cheaper as jatropha oil is about 20% the cost of crude oil, and more environmentally friendly alternatives to fossil fuel. Air New Zealand announced plans to use the new fuel blended with jatropha oil for 10% of its needs by 2013 (Berchmans and Hirata, 2008). *Jatropha* oil is also used to soften leather and lubricate machinery (e.g. chain saws). A large quantity of seed cake can theoretically fuel steam turbines to generate electricity. The oil also finds the application in cosmetic industries, for the manufacture of candles and soap. The glycerol has many useful industrial applications as a raw material for the synthesis of 1, 3 propane- diol and other polymeric materials (Vinayak and Kanwarjit, 1991; Berchmans and Hirata, 2008). Biodiesel from *Jatropha* has similar combustion properties as diesel and kerosene. So *Jatropha* oil as a potential domestic fuel for cooking and lighting, with properties similar to kerosene is proposed. But it cannot be used directly in conventional kerosene stoves or lamps. High ignition temperatures and viscosity properties show that *Jatropha* oil will not burn well, and would clog up all the tubes and nozzles in a conventional stove or lamp (Vinayak and Kanwarjit, 1991; Berchmans and Hirata, 2008).

The use of *Jatropha* oil by local soap industry is one of the most economically attractive alternative uses of the oil. The glycerine by-product of the trans-esterification process of *Jatropha* oil can be used to make a high quality soap, or it can be refined and sold at a range of prices, depending on its purity, to be used in an immense range of products, including cosmetics, toothpaste, embalming fluids, pipe joint cement, cough medicine, and tobacco (as a moistening agent). The soap has positive effects on the skin and is therefore marketed for medicinal purposes (Berchmans and Hirata, 2008).

Use of *Jatropha* seed cake

Jatropha seed cake contains curcin, a toxic protein similar to ricin in Castor, making it unsuitable for animal feed. However, it does have potential as good organic manure (Staubmann *et al.*, 1997; Gubitz *et al.*, 1999), replacing chemical fertilizer since it has nitrogen content ranging from 3.2 to 3.8%. Kumar and Sharma (2008) similar to that of neem oil cake and cow dung manure. A fertilizer trial with pearl millet where the effects of on pearl millet were compared (Henning 2004). Pearl millet

yields per ha were maximum in *Jatropha* oil cake treatment (5 t/ha) compared to that with farm yard manure (5t/ha), and NP fertilizer (100 kg ammonium phosphate and 50 kg urea/ha) (Henning, 2004). Press-cake derived from the non-toxic varieties of *Jatropha curcas* may be used as animal feed. The proximate analysis of the seed cake showed 11% oil, has 58-60% crude protein (53-55% true protein content), and the level of essential amino acids except lysine is higher than the FAO reference protein. Seed cake from Mexico and Central American non-toxic varieties may not be toxic. However, nontoxic varieties are not grown in Asia and Africa.

Tissue culture and genetic engineering

The application of molecular techniques in plant diversity conservation becoming increasingly popular, the isolation of impact, high-molecular mass genomic DNA becomes an important pre-requisite (Dhakshanamoorthy and Selvaraj, 2009). However, species of *Jatropha* contain polysaccharides and polyphenols posing a major problem in the isolation of high quality DNA. Although several protocols are used for isolation of genomic DNA in *Jatropha* species, all of them use expensive and toxic chemical liquid nitrogen. Although several successful DNA extraction protocols for plant species containing polyphenolics and polysaccharides compound have been developed, none of these are universally applicable to all plants (Varma *et al.*, 2007) and the published protocols are also limited because of degradation of DNA and other nucleases. Therefore, researchers often modify a protocol or blend two or more different procedures to obtain DNA of the desired quality (Varma *et al.*, 2007). A good isolation protocol should be simple, rapid and efficient, yielding appreciable levels of high quality DNA suitable for molecular analysis. The common procedure is to grind plant tissue in liquid nitrogen and transfer to a preheated extraction buffer (Dellaporta *et al.*, 1983; Mohapatra *et al.*, 1992). Liquid nitrogen can be difficult to procure in remote locations. Thus, a method not requiring use of liquid nitrogen would be helpful to researchers in remote area. Dhakshanamoorthy and Selvaraj (2009) developed a DNA isolation method suited for isolation of genomic DNA in *Jatropha* leaves that can be stored for a longer duration, lasting for several PCR reactions. The method has used no expensive and toxic chemical and is suitable for low-facility laboratories (Dhakshanamoorthy and Selvaraj, 2009). A poor seed germination, scanty and delayed rooting of seedlings and vegetative cuttings (Heller, 1996; Openshaw, 2000; Purkayastha *et al.*, 2010) paved way for the necessity of micro propagation of *Jatropha* through embryo or embryo derived explants cultures with promising method for disease free and in vitro culture manipulations (Purkayastha *et al.*, 2010), a means for conservation of elite germplasm. The other problem of the presence of toxic chemicals in the seeds and seed protein can be mitigated by methods adopted for *Lathyrus sativus* L. (Misra and Misra, 1993; Misra *et al.*, 1994). Genetic engineering appears to be an effective approach to reduce the levels of these toxic substances in seeds (cf. *Lathyrus sativus* L. Misra and Misra, 1993; Misra *et al.*, 1994), and increase resistance to biotic stresses, and furthermore, offers opportunities to modify seed oil for higher engine efficiency (Hossain and Davies,

2010). The development of an efficient regeneration system amenable to genetic transformation is a prerequisite for plant genetic engineering (Misra and Misra, 1993; Misra *et al.*, 1994). Efforts for the last two decades have failed to provide a reliable protocol of in vitro plant regeneration of *J. curcas* (Jha *et al.*, 2007; Deore and Johnson, 2008). However, in recent years, plant regeneration in *J. curcas* has been accomplished through organogenesis from various explants, including mature leaf (Sujatha *et al.*, 2005; Deore and Johnson, 2008), petiole and hypocotyls (Sujatha and Mukta, 1996), axillary node (Sujatha *et al.*, 2005; Shrivastava and Banerjee, 2008) and via somatic embryogenesis from mature leaf explants (Jha *et al.*, 2007). However, amenability of these regeneration systems to genetic transformation methods has not been evaluated. Seed-derived explants are, in general, known to be more responsive to rapid regeneration (Tiwari and Tuli, 2009) and Agrobacterium-mediated transformation (Patnaik *et al.*, 2006; Paz, 2009). Purkayastha *et al.* (2010) recently developed a method for rapid and efficient plant regeneration from shoot apices, and generation of transgenic plants by direct DNA delivery to mature seed-derived shoot apices of *Jatropha*. Mazumdar *et al.* (2010) studied the effects of age and orientation of the explant on callus induction and de novo shoot regeneration from cotyledonary leaf segments of *J. curcas*. Highest regeneration response was reported in the young explants, derived from the cotyledonary leaf of germinating seed compared to the leaves from one- and two-week-old seedlings. This gradient with age of the explants was observed in callus induction (%), shoot organogenesis (%) from callus. The explants cultured with their abaxial side in medium showed significantly higher regeneration response. The youngest explant was found to be most amenable to Agrobacterium-mediated transformation as compared to older explants giving rise to stable transgenic plants in *J. curcas*. Agrobacterium-mediated transformation of *Jatropha* using cotyledonary leaf explants was reported by Li *et al.* (2007). However, this was inefficient and difficult to reproduce. But, in a concerted effort to develop efficient protocol for genetic transformation of *Jatropha*, Mazumdar *et al.* (2010) and Kumar *et al.* (2010) simultaneously reported an efficient Agrobacterium-mediated genetic transformation and plant regeneration protocol for *J. curcas* using leaf explants. Kumar *et al.* (2010) reported highest transformation efficiency of 4-day precultured, non-wounded leaf explants infected with Agrobacterium and co-cultured on Murashige and Skoog (MS) medium supplemented with thidiazuron (TDZ) for regeneration of shoot buds, and proliferation of shoots in MS medium containing kinetin (Kn), 6-benzyl aminopurine (BA), and naphthalene acetic acid (NAA). Subsequent growth of the shoots was promoted in MS medium supplemented with BA and IAA, and rooting in 0.5N MS medium with IBA, IAA, NAA, and activated charcoal. GUS expression analysis, PCR and DNA gel blot hybridization confirm the expression and presence of transgene. A transformation efficiency of 29% was achieved for leaf explants using this protocol Kumar *et al.* (2010). These protocols have the potential to facilitate the genetic modification and subsequent true-to-type in vitro multiplication of *J. curcas*

cultivars. Development of this technology for *Jatropha* can lead to a better understanding and improvement of the bio fuel species (Mazumdar *et al.*, 2010).

Conclusion

Successful application of *Jatropha* is made in soil and water conservation, soil reclamation, erosion control, living fences, green manure, source for bio-diesel, use in soap production, insecticide and as raw material for pharmaceutical and cosmetic industries. However, research on the genetic diversity, adaptability, evaluation and conservation the diverse germplasm of *Jatropha* needs concerted efforts. The crop management and fertilizer requirements of *Jatropha* are not extensive as in other crops. The natural genetic variation available within the species can be exploited to create interspecific hybrids for incorporating a wide range of beneficial traits to be incorporated into *Jatropha* species for improvement of the crop. The toxic HCN content of the bark, fruit, leaf, root, and wood is the main limitation for the use of plant/ and plant parts as such. Also the utilization of seed husk, seed kernel, raw bio-materials in the manufacturing sectors needs to be explored and exploited. The presence of high concentrations of albumin and curcin etc. renders the seeds fatally toxic when consumed. Research needs to be focused in reducing these toxic products, as that done for other crops (Misra and Misra, 1993; Misra *et al.*, 1994).

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