

Transformation of toxic potential of *Jatropha curcas* (Ratanjyot) into protein source: A mini-review

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ABSTRACT

The production of animal largely depends on supplying of quality feed and proteinaceous supplement to the animals. *Jatropha* plant can grow in the barren lands, and are used as a source of biodiesel. Besides, the plant may act as a rich proteinaceous source. However, the antinutritional factors present in the seed and seed oil of the plant may hamper the availability and beneficial use of the plant. Curcin and phorbol esters are the major toxic compounds present in the plant; these toxic compounds cause to produce liver and kidney diseases. Detoxification of these toxic compounds by physical and chemical means converting to less toxic seed cake may serve the purpose of using this plant in future as a replacement of costly protein supplement for animals. Therefore, in modern world, it is recommended to utilize the protein source by neutralizing the antinutritional factors. This mini-review describes the updates on how *J. curcas* can be utilized as a supplementary source of protein for animals by decreasing its toxicity.

Keywords

Biodiesel, Curcin, *Jatropha*, Phorbol esters

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INTRODUCTION

India, a developing country, has rich biodiversity of flora and fauna. About 70% population of the country depends on agriculture and livestock for their

livelihood. The animal production level depends on supplying of quality feed and other proteinaceous supplement to the animal. In India, farmers pay high cost for supplying protein source to their animals. The use of barren land for increasing production of *Jatropha curcas* make this floral entity special. *J. curcas* Linn (Greek:-iatros-doctor, trophe-food), also known as physic nut, purging nut, Ratanjyot, Barbados nut, Black vomit nut and *Curcas* bean, belongs to the floral Euphorbiaceae family having origin in Central and South American countries and was introduced into Asia and Africa as an oilseed bearing tree by Portuguese seafarers.

The major energy carriers for *Jatropha* are raw oil and esters, and its use as replacement of energy sources is well documented (Kywe et al., 2009). Several studies have been carried out on the holistic approach to utilize *Jatropha* as a source of multiple energy carriers (Gunaseelan, 2009). The plant is not showing the alleopathic reactions and not secreting any of the harmful chemicals neither organic nor inorganic to check the growth of the neighbouring vegetation. The plant is hardy in nature and can sustain the high range of temperature, and can survive economically for 30 years. The plant is highly susceptible to frost but can sustain fertility and growth even in poor soil profile and in drought prone areas as of Laddakh and Maharashtra (Munch et al., 1989). The generation of de-oiled *Jatropha* waste (DJW) increased tremendously in recent years due to the high demand of biodiesel as an alternative biofuel (Kumar et al., 2013). A *Jatropha*-based biodiesel plant produces 2.5-3.0 tons of solid waste (DJW) per one ton of biodiesel (Srividhya et al., 2010). It has been reported that the reducing sugars

recovered from lignocellulosic waste product could be a low cost source of solubilising protein (Liang et al., 2010; Kootstra et al., 2011). In addition to that, it can also be used in enzyme production such as xylanase (Das et al., 2011; Joshi and Khare, 2011).

Jatropha is a genus of approximately 175 succulent plants, shrubs and trees. Taxonomically, *J. curcas* is classified under Kingdom- *Plantae*, Order- *Malpighiales* and Family - *Euphorbiaceae*.

Characteristics and description of the plant

J. curcas is a large coarse annual shrub which can grow up to 3.5 to 4.5 meters (8-15 feet) in height with an annual seed yield of 5 tones per hectare (Makkar et al., 2009).

Roots: Normally, five roots are formed from seedlings, one central and four peripheral. A tap root is not usually formed by vegetative propagated plants (Solsoloy et al., 1997).

Leaves: The leaves are large, green to pale green in color with five to seven lobes, hypostomatic and paracytic (Rubiaceous) type stomata (Gupta, 1985).

Seed: The seeds are mature when the capsule changes from green to yellow 3-4 months post flowering. The seeds are black and the seed weight per 1000 seeds is about 727 g, there are 1375 seeds/kg in the average. The physic nut is a diploid species with $2n=22$ chromosomes (Li et al., 2010).

Habitat: *J. curcas* Lin (physic nut, purging nut), a tropical plant originated from Central America but has been naturalized in most tropical and subtropical countries from South-America to Africa and Asia. Its tolerance of various soil and climatic conditions allows its vast distribution within the so called "*Jatropha* belt" stretching between 30° N and 35° S (Jongschaap et al., 2007).

Current applicability: It is cultivated mainly as a hedge in many Latin American, Asian and African countries. The use of the seed oil for cooking purposes and the press cake as animal feed is not possible due to the presence of toxic compounds (Gubitz et al., 1998). Therefore the experiments are going in order to detoxify the plant cake and seeds and use them efficiently as a source of protein to the animals. Nowadays, *Jatropha* has been drawn attention of reserchers as different parts of this plant has potentials to be used as animal feed, biofuel, and herbal medicine.

The seed kernel of *Jatropha* contains about 60% oil; this oil convertible into biodiesel, and can be used as a substitute for diesel fuel. The seed cake produced after extraction of oil is considered as a good source of plant nutrients (Foial et al., 2001). The seeds contain up to 30-45% oil with a fatty acid pattern similar to that of edible oil, the percentage of essential amino acids and mineral content can be compared to those of other seeds.

All parts of *J. curcas* have been used in traditional human medicine and for veterinary purposes for a long time (Duke, 1985). For example, it has been used to cure diseases like hemorrhoids, gonorrhoea, dysentery, coated tongue, small pox, skin infections, and infertility. It is also used for making candles and soap. But the use of the seed oil for cooking purposes is not possible due to the content of toxic compounds. Phorbol esters (Figure 1) (phorbol-12-myristate 13-acetate) have been identified as the major toxic compound in this plant (Makkar et al., 2009).

Toxic principles

The following toxic principles (compounds) have been detected in *J. curcas*:

1. Curcin: The seeds of *Jatropha* contain a toxalbumin curcin belonging to ribosome-inactivating proteins (RIP), and causes the specific modification of the larger rRNA to cause inhibition of ribosomal synthesis that in turn inhibit the protein synthesis (Endo et al., 1987).

Based on physical properties, RIPs are classified into (a) type 1 RIP, which are single-chain (approximately 30 kD) proteins having enzymatic activity and inhibiting cell-free protein synthesis *in vitro* relatively nontoxic to cells and animals; and (b) type 2 RIP, which are heterodimeric proteins (approximately 60 kD) consisting of A chain (similar to type 1 RIP) attached to sugar-binding B chain (lectin) by a disulfide bond. Type 2 RIP is highly toxic as compared to type 1 RIP which are relatively nontoxic *in vivo* due to the absence of the sugar-binding chain. Curcin, a type1 RIP (28.2 kD, isoelectric point 8.54), was isolated from the seeds of *J. curcas*. Curcin has protein translation inhibitory activity or *N*-glycosidase activity like other type 1 RIP (Lin et al., 2010).

2. Diterpenes (Phorbol esters): Diterpenes are believed to be the most potent compounds synthesized by *Jatropha* species. There are at least 20 diterpenes reported from *Jatropha* species. Among the diterpenes, a group of compounds having tigliane skeleton called phorbol esters are the most toxic molecules in a *Jatropha* species. Six phorbol esters have been identified in *J.*

curcas (Haas et al., 2002). Seed and seed oil of the plant having the diterpenes in the form of Phorbol esters. Phorbol esters are natural compounds and highly toxic, cathartic and skin irritant (Makkar et al., 2009).

The placement of -OH group makes the phorbol an active (β) or inactive (α) type, which results in spatial arrangement of ring D as in the **Figure 1** in tigliane and precludes activation of PKC. The inactive α phorbol esters have similar physicochemical properties and lipophilicity as the active β phorbols are unable to activate PKC due to conformational changes (Silinsky et al., 2003).

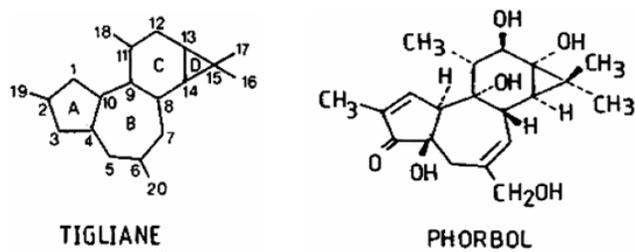


Figure 1: Structure of Tigliane and Phorbol ester.

Phenolic compounds

Phenolic compounds are widely distributed in plants. Tannins are polyphenolic compounds constitute insoluble complexes with proteins and essential nutrients (Sosulki, 1979). Excess consumption of tannins can damage the mucosal lining of the gastrointestinal tract, alter the excretion of certain cations and increase the excretion of proteins and essential amino acids (Reddy et al., 1994).

Phytates

Phytic acid, the hexaphosphate of myoinositol, functions as the chief storage form of phosphate in mature seeds leading to a reduction in bioavailability of many essential minerals like iron, calcium and magnesium (Erdman, 1979).

Trypsin inhibitors

The trypsin inhibitors may cause an increase in the digestive enzymes secretion by inducing hypertrophy and hyperplasia of the pancreas. This was explained by the hypothesis that the growth depression caused by the inhibitors was the consequence of an endogenous loss of important aminoacids which serve as basis for enzyme secreted by a hyperactive pancreas (Liener, 1994; Siddhuraju et al., 2002).

Pharmacological uses of *J. curcas*

Antioxidant activity: Hydro-alcoholic extract of the leaves, stem and root of *J. curcas* had showed significant antioxidant activity using in vitro antioxidant models like DPPH radical scavenging activity, nitric oxide radical scavenging activity, hydroxyl radical scavenging activity, reducing power method and hydrogen peroxide radical scavenging activity (Diwani et al., 2009).

Hepatoprotective activity: Methanolic fraction of *J. curcas* L. showed hepatoprotective activity on aflatoxin B1 induced hepatic carcinoma in animals (Balaji et al., 2009).

Wound healing activity: It was reported by Shetty et al. (2006) that the herbal ointment of *J. curcas* leaves and barks accelerates the healing process by increasing the skin wound contraction, breaking strength, granulation tissue breaking strength, and dry granulation tissue weight and hydroxyproline levels in rats.

Antimetastatic and antiproliferative activity: Methanolic fraction of *J. curcas* L. was studied for its anti-metastatic activity using B16F10 melanoma cells in C57BL/6 mice. It was studied using MTT (3- [4, 5-dimethylthiazol-2-yl] -2,5-diphenyltetrazolium bromide; Thiazolyl blue) assay. The IC50 was found to be 24.8 $\mu\text{g} / \text{mL}$ (Balaji et al., 2009).

Antimicrobial activity: Methanolic, ethanolic and water extracts of stem bark from *J. curcas* L. revealed in vitro antimicrobial activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, and other microbes (Igbinsosa et al., 2009).

Antidiabetic activity: Antihyperglycemic effect of 50% ethanolic extract of leaves of *J. curcas* Lin was studied in normal and alloxan induced diabetic rats (Mishra et al., 2010).

Anti-inflammatory activity: The methanolic extract of this plant exhibits anti-inflammatory activities in acute carrageenan-induced rat paw edema. Besides, it speculated activity against formalin-induced rat paw edema, cotton pellet-induced granular tissue formation, and turpentine-induced exudative changes in rats and mice (Mujumdar et al., 2004).

Pregnancy terminating effect: Fetal resorption was observed with methanol, petroleum ether and dichloromethane extracts indicating the abortifacient properties of the fruit in rats. Besides, it might interrupt

pregnancy occurred at an early stage after implantation (Goonasekera et al., 1995).

Antiulcer activity: Methanolic extract of *J. curcas* Lin showed the antiulcer activity using aspirin induced gastric lesions in Wister rats (Kannappan et al., 2008).

Anthelmintic activity: Aqueous extract of leaves have anthelmintic activity against nematodes (Ahirrao et al., 2008)

Antifungal activities: The ethanolic extract of *J. curcas* L. seed cake exhibited antifungal activities against several fungi, for example, *Pythium aphanidermatum*, *Fusarium oxysporum*, *Colletotrichum gloeosporioides*, *Curvularia lunata*, *Lasiodiplodia theobromae*, *Colletotrichum capsici*, and *F. semitectum*. The extract contained phorbol esters, which is responsible for showing antifungal activities (Donlaporn et al., 2010).

METHODS OF DETOXIFICATION OF THE PLANT PRODUCTS

There are several physical and chemical means by which detoxification of seeds and seed oil of the plant can be achieved to destroy phorbol esters in feed. Solvent extraction of phorbol esters followed by heat treatment to inactivate lectin in *Jatropha* seed meal was reported to convert the nontoxic meal to a high-quality protein source for livestock.

Toxicopathological findings of poisoning of *J. curcas* in animals and human beings: Various studies have been performed on the toxic potential of the plant in several species model as mice, rats, pigs, goats etc. Gandhi et al. (1995) have investigated the toxicological effects of *J. curcas* (ratanjyot oil) in rats, reporting that an acute oral LD₅₀ of the oil to be 6 mL/kg bwt. The oil was reported to cause toxicological manifestations like diarrhea, gastrointestinal inflammations and skin irritation followed by necrosis. Single oral dose of 50 mg/kg bwt of acetonitrile extract of *J. curcas* caused slight alterations in liver, kidney and spleen in rats (Abd-Elhamid et al., 2004). Feeding of *J. curcas* seed kernel meal containing phorbol (dosed at 0.23 mg/gm) for 10 days was found to be highly toxic in rats. The mortality occurred within 8-10 days. All rat showed poor appetite and low diet intake, loss of body weight, difficulty in motor functions and severe diarrhea before death (Devappa et al., 2008). Kumar et al. (2014) reported that in rats seed oil (dosed at 1 mL/kg bwt) produced the tubular degeneration of kidney and disorientation of the hepatic portal system.

It has been reported that powdered *J. curcas* seeds produced toxicity to young Nubian goats (5-8 months old; 8-22 kg bwt) at dose ranging from 0.25 to 10 g/kg/day. Dosed at 5-10 g/kg/day was found to be toxic with fatal consequences, and goats died between 2 and 4 days. The clinical signs observed were lack of appetite, reduced water consumption, diarrhea, dehydration and sunken eyes. Post mortem studies revealed hemorrhage in the rumen, reticulum, kidneys, spleen, and heart, catarrhal or hemorrhagic abomasitis and enteritis, congestion and edema of the lung, and excessive fluid in serous cavities (Adam et al., 1975).

Shukla et al. (2013) reported that dosing of seed oil dosed at 1 mL/4 kg bwt for 28 days in goats resulting into the moderate weakness, lethargy and diarrhoea. Acute toxic study conducted by Shukla et al. (2012) reported that seed and seed oil dosed at 4 seed/kg and 4 mL/kg bwt respectively, produced decline in the haematological parameters as haemoglobin and PCV.

The calves force-fed with *J. curcas* seeds at single doses of 0.25, 1, or 2.5 g/kg bwt showed toxic symptoms and death occurred within 19 h of administration. Calves force-fed 0.025 g/kg daily up to 14 d showed signs of poisoning and death between 10 and 14 day (Ahmed et al., 1979).

The accidental ingestion of *J. curcas* seeds in children produced abdominal pain, burning sensation in the throat, vomiting and diarrhea between 30 min to 2 h after consumption with a recovery time of 24 h. Depression and circulatory collapse were also reported in few cases of poisoning in children (Singh et al., 2010).

Diagnosis and treatment of poisoning: Diagnosis can only be done by means of case history and symptoms manifested by the intoxicated animals. Confirmatory diagnosis relies mainly on history of ingestion of seed or any part of *Jatropha* and detection of toxic principles in the stomach content.

The management of *Jatropha* poisoning is similar to that for the castor bean (*Ricinis*). There are no specific antidote, and only symptomatic and supportive therapy are given for the treatment of *Jatropha curcas* toxicity in man and animals. Administration of activated charcoal and a cathartic hasten the elimination of the toxicant. Intravenous fluid administration should be given in order to dilute and eliminate toxicant rapidly to counteract fluid loss due to diarrhea and vomition. Supplementation of activated charcoal (0.1%) and gentian violet (0.05%) resulted in significant improvement in the performance of the birds

with improvement in gross and histopathological changes from 4th week onwards after *J. curcas* seed toxicity in broilers (Sirisha et al., 2008). Activated charcoal (0.1%) was observed as a good ameliorating agent against entero-hepato-nephrotoxicities whereas gentian violet produced better ameliorative action on entero and hepatotoxicities than nephro-toxicity (Sirisha et al., 2008).

CONCLUSION

Jatropha curcas may serve the society as a medicinal plant, fuel source in the form of biodiesel, and source of protein supplement to the animal. The detoxification procedures have to be adapted efficiently and should have been more refined to reduce the incidences of toxicity of the plant. In future, one can potentially use the plant as a good source of protein supplement to animals as replacer of Ground nut cake and soya cake. It can cut loss the cost of feeding of animals; thus farmers can be benefitted both directly and indirectly.

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