Review Article

Jatropha curcas, a promising crop for the generation of biodiesel and value-added coproducts

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The review highlights the specific features of the *Jatropha curcas* plant and its potential for the production of biofuel, protein concentrates as livestock feed and value-added products that could enhance the economic viability of Jatropha seed oil-based biodiesel production. The roles of the plant in carbon capture, enhancing socio-economic conditions, food production in the tropical regions, and influencing micro-climate, vegetation and soil quality are discussed. The paper also gives a comparative account of the toxic and non-toxic genotypes of *J. curcas* from the point of view of their physical and chemical properties and their potential for biodiesel and livestock feed production. Future areas of research are also presented.

Keywords: Biofuel / Co-products / *Jatropha curcas* / Non-toxic Jatropha

Received: October 22, 2008; accepted: March 27, 2009

DOI 10.1002/ejlt.200800244

1 Introduction

The low energy prices potentially thwarted the efforts to reduce greenhouse gas (GHG) emissions until recently. The oil prices today are fluctuating unpredictably, but are likely to stay higher in the future. In the developed economies, high fuel prices do not prevent wealthy people from driving and energy consumption. Carbon dioxide emission will thus be the rule, with net reductions being the exception in the distant future, unless the governments can motivate people to reduce their use of fossil fuel.

The fact that the fossil energy reserves are sufficient for another 100 years is not a good argument for its uncontrolled exploitation and use, as the current emission of close to 10 billion tons of carbon a year could further negatively impact the world climate change. Liquid fuel demand is projected to increase by more than 2% per annum over the next two decades [1]. To cover this demand, renewable energy resources should cover an appreciable part.

On a global scale, the area of degraded land is much larger than the 1.4 billion ha under agricultural use [2] and increases by approximately 10 million ha every year. Assuming that an average grain yield on that land of 2.5 t only per hectare could be achieved, these 25 million tons of grain would be sufficient to feed an additional 100 million people. Therefore, it is important to prevent the degradation of fertile land and to reclaim already lost cropland.

The use of plants and plant products – among others – as replacement of conventional fuels is an excellent option, but depends on sufficient knowledge regarding the plant's relationship with its environment. Because of the danger that such plants might replace food crops in food-insecure regions, bioenergy production should be restricted to otherwise uncultivable land. Knowledge defining appropriate plants, cultivation systems and general agronomic practices to utilise these abandoned lands is a prerequisite for future success. The objective of this review is to discuss the roles of the Jatropha plant in carbon capture, enhancing socio-economic conditions, food production in the tropical regions, and in influencing micro-climate, vegetation and soil quality. The paper also presents a comparative account of the toxic and non-toxic genotypes of *Jatropha curcas* from the point of view of their physical and chemical properties and their potential for biodiesel and livestock feed production.

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2 The Jatropha plant

In a study by Azan *et al.* [3], a comparison of 75 non-edible oilseed plants revealed *Jatropha curcas* L. to outmatch all the

others as a source of biodiesel. De Chelmicki and De Varnhagen [4], De Avila [5], Levingston and Zamora [6], and Martin and Mayeux [7] pointed out that *J. curcas* L. has excellent adaptation capacity to a large variety of soil conditions. Cultivation on dry, stony and very shallow soils is frequent and plants are found on the Cape Verde Islands, thriving even between bare rocks. Equally, Jatropha appears well adapted to conditions of low to very low soil fertility, and mineral deficiency symptoms are rarely observed. The plant must have an enormous capacity to absorb and utilise nutrients under low-fertility conditions as it grows well even on the poorest, mostly P-deficient and acid soils such as those found on the Cape Verde Islands.

A temperature range of 25–35 °C is optimum for *J. curcas* growth, but in some regions in the tropics it may be found at higher altitudes with the risk of light frost. On the other hand, Jatropha can tolerate elevated temperatures to far above 40 °C, as has been documented in a 150-ha plantation in Upper Egypt, Luxor, where on 260 days of the year the temperatures exceed 40 °C. As the region receives no rain (7 mm), sewage water from the city of Luxor is used for irrigation. Temperatures below 20 °C for a week or even shorter periods initiate leaf shedding.

Jatropha is classified as the climax vegetation of tropical savannas in the dry [8] or semi-dry tropics [9]. Its drought tolerance and adaptation capacity to long, severely dry seasons are well developed, to a degree that established plants have been reported to grow even where there is no rain for 2–3 years [10]. On the other hand, Jatropha appears to tolerate humid conditions equally well, showing good growth with high rainfall. Jatropha is therefore highly adaptable to varying precipitation conditions. Heavy rains at the time of flowering could lead to the complete loss of flowers. Jatropha does not tolerate instantaneous flooding.

The genus *Jatropha* is extremely old and may have already existed 70 million years ago on the ancient continent "Gondwanaland" before it split up to form the individual continents. It is a member of the large Euphorbiaceae family. The genus *Jatropha* in the family of Euphorbiaceae consists of between 165 and 175 species. Dehgan and Webster [11] distinguished two subgenera, ten sections and ten subsections [12]. The Caatingas in Northeast Brazil and the dry areas of Mexico have been identified as the centres of diversity.

The name "Jatropha" is derived from the Greek iatros (doctor) and trophe (food). There are two genotypes of *Jatropha curcas*, a toxic and a non-toxic one. The latter genotype is found in Mexico only. Jatropha is a diploid species with $2n = 22$ chromosomes. Therefore, standard quantitative genetic methods can be applied [13].

The monoecious plant (*i.e.* unisexual reproductive units of both sexes appear on the same plant) is pollinated by insects. Jatropha is self-compatible [14], but cross-pollination is supported by a time gap between anthesis of male and female flowers [12]. The existence of protandry may have a strong impact for cultivation of Jatropha hybrids, and its magnitude has to be investigated in more detail. Considering the reproduction biology of Jatropha, manual crossing is possible using standard emasculation and bagging techniques.

Jatropha can be propagated as cuttings or with seeds. Plants propagated vegetatively do not usually form tap roots [15]. The advantage of generative propagation compared to vegetatively propagated plants needs to be substantiated. Seed propagation – even in the case of transplanting nursery-raised seedlings – is clearly less costly but produces a highly variable stand, whilst vegetative propagation (cuttings) allows establishing uniform stands of selected, high-yielding material.

Jatropha curcas is a shrub or a small- to medium-sized tree. Solitary trees grow large and can reach heights of more than 12 m in Paraguay. For many decades, ecologists have long been studying stability in ecosystems on a global level by looking at the structuring and strengths of interacting parameters. According to Costanza *et al.* [2], out of the 10.2 billion ha of terrestrial area of the globe (forest, grassland, cropland), close to 2 billion ha are already degraded and can no longer contribute to food and feed production. In fact, huge areas of former cropland are lost every year. A way to reflect the value of ecosystem loss is to determine what it would cost to reclaim eroded land.

The current practice of converting intact natural resources to produce biofuels and animal feed, especially in the tropical regions, creates a massive carbon debt, changes the gaseous composition of the global atmosphere and negatively impacts human welfare. The services of natural resources to human welfare can no longer be ignored or undervalued, as this will inevitably lead to irreplaceable ecosystem values.

To produce biofuel or animal feed on converted former Amazon forest, for example, leads to a massive carbon release, which can be 17–420 times more than the average annual GHG reduction that the new crop may provide [16]. On the other hand, *J. curcas* is used to prevent or control erosion, and even to reclaim eroded lands in many countries. It is used also as a living fence since it is not grazed by animals. The plant is native to almost all tropical regions. It is a multipurpose plant and yields oil that is easily converted into biodiesel by conventional, proven processes.

2.1 Carbon capture

Growth in the use of liquid fossil fuels has led to substantial increases of CO_2 emission. The international community has put in place measures to reduce gas emissions. The Kyoto protocol (KP) implies a cap-and-trade system for GHG emission and functions as the main biofuel driver in the EU. Political CO_2 reduction targets for the European Community have been implemented and are enforced by the member states.

The KP allows market players to strive for optimum cost allocation and to reduce emissions where it is most cost efficient. In order to achieve the EU biofuel targets, close to 10 million ha in 2010 and 14.2 million ha in 2020 are neces-

sary for biofuel cropping. Of the 82 million ha of arable lands in the EU, approximately 10% is lying idle [17], and the equivalent amount of biofuel that could be produced from 8.2 million ha are to be imported into the community if land use change (grassland/forest) is to be prevented. Energy cropping on converted land may result in a long biofuel carbon debt repayment time. Through its suitability for wasteland cultivation, *J. curcas* provides two mechanisms for GHG abatement: substitution of fossil fuel and CO_2 sequestration through increasing carbon stocks above and below ground. Countries with developing economies cannot be expected to accept a carbon cap on their emissions, because their per-capita CO_2 release and their per-capita income are only fractions of those of industrialised countries. Here the KP's Clean Development Mechanism (CDM) applies, if it can be demonstrated that the CDM project avoids GHG emissions that would have occurred in the most likely scenario in the absence of the project (the so-called baseline scenario). The avoided emissions are considered as real reductions and can be certified as "certified emission reductions" (CER) and can be used in global emission trading. If CER are used, those emissions saved, *e.g.* in a developing country, will be emitted elsewhere. In other words, the total global GHG balance will not change. Generally reducing emissions in the developing world may be more cost effective than in an industrialised country.

Globally, there are huge areas of degraded former croplands available in the developing world that are suitable for planting *J. curcas*. The establishment of the energy plant on such areas not only reduces GHG emissions but also creates opportunities for impoverished farmers and rural labourers. Contrary to other biofuels, the use of *J. curcas* represents real advantages over conventional biofuel sources such as corn, sugar cane and palm, which to a large extent grow on converted lands. Life cycle studies for *J. curcas* need to be undertaken. Unlike other energy plants, Jatropha, when planted on wastelands and degraded lands, does not interfere with food security in food-insecure countries. Such studies will only enable the realisation of proper accounting on the impact on carbon benefits (soil and plant organic carbon stores).

2.2 Biodiesel/bio-oil resource

If a 5% blending of biodiesel with mineral diesel would be mandatory for the whole of the current OECD (only), the total requirement would be above 21 million tons per year. If the two energy markets of China and India and others are considered, it becomes evident that the market potential of biodiesel is huge.

Within the frame of our DaimlerChrysler research and development project in Bhavnagar, India, first street tests with common rail direct injection (CDI) diesel cars started by the end of 2004. To date, over 80,000 L of neat Jatropha biodiesel (Jatropha methyl ester, JME) have been tested under various climatic conditions using personal as well as small track cars of DaimlerChrysler. These tests have shown that there is only a minimally better efficiency of 1.7% in consumption in favour of fossil diesel. JME meets the EN specification 14214 (Table 1).

Striking differences are found for cetane number and emission parameters for Jatropha biodiesel and mineral diesel; specifically, emissions of hydrocarbons and particulate matters are 80% lower from biodiesel than from mineral diesel. The emission of sulphur dioxide using biodiesel is virtually nil.

The oil of the Jatropha plant is not edible because of its toxin phorbol ester, but can serve as biofuel in adapted diesel engines such as the one of the Elsbett engine. Reddy and Ramesh [18] used neat Jatropha oil in a direct-injection diesel engine and showed lower emissions of hydrocarbons and oxides of nitrogen compared to those from mineral diesel. Pramanik [19] established that 40–50% of Jatropha oil can substitute for diesel without any engine modification and preheating of the blends. From the energetic point of view it can be an advantage not to esterify the oil but to use it directly. The gross energy content of the oil has been found to range from below 37 MJ/kg to 39.5 MJ/kg. Especially in remote and difficult to access regions, the use of plant oil might be recommended.

2.3 Protein concentrate as animal feed

With the increase in purchasing power in the emerging countries, the demand for dietary products of animal origin is increasing exponentially. On the other hand, assurance of the availability of high-quality protein, minerals and vitamins through animal products is the most important way to reduce malnutrition in many areas of the world, especially among the young up to the age of five.

Today's global production of *J. curcas* seeds from plantations is still negligible. However, it is believed that approximately 25–30 million ha are currently being established in different parts of the world, largely with the toxic genotype. One ton of seeds (35% shell mass and 58% oil in kernel) yields 270 kg Jatropha kernel meal containing roughly 60% crude protein in dry matter [20].

Looking at the probable global impact of this protein concentrate, it can be calculated that the potential exists to produce Jatropha kernel meal equivalent to 5.6 Mt of soybean meal on a protein equivalent basis by 2020 in China alone, in a conservative scenario, and between 67 and 190 Mt of soybean meal in an optimistic scenario [20]. The kernel meal, if detoxified, could be a good substitute for soybean meal in diets of farm animals and aquaculture species.

2.4 Chemicals for medicinal, pharmaceutical and biopesticide applications

Phorbol esters, which are responsible for the non-edible nature of Jatropha seeds, could form very potent bio-compounds against many plant diseases, and probably against disease

Table 1. Properties of *Jatropha curcas* oil methyl ester (biodiesel) compared to European standards.

Property	Method	Unit	Jatropha biodiesel	EN 14214 standard
Appearance			clear brown	
Density at 15 °C	ISO 3675	kg/m^3	884.2	860–900
Solid contamination	EN 12662	mg/kg	2	<24
Acid number	EN 14104	mg KOH/g	0.11	<0.5
Copper corrosion	EN ISO 2160	grade	1	1
Oxidation stability	EN 14112	h	6.7	>6
Viscosity at 40 °C	ISO 3104	mm^2/s	4.4	3.5–5.0
Iodine number	EN 14111	g/100 g	93	<120
Sulphated ash	ISO 3987	g/100 g	<0.01	<0.02
Water content	EN ISO 12937	mg/kg	590	<500
Flash point	DIN EN 22719	°C	172	>101
Cetane number	EN ISO 5165	–	58.5	>51
Monoglycerides	EN 14105	g/100 g	0.01	<0.8
Diglycerides	EN 14105	g/100 g	0.02	<0.2
Triglycerides	EN 14105	g/100 g	>0.02	<0.2
Free glycerine	EN 14105	g/100 g	<0.02	<0.2
Total glycerine	EN 14105	g/100 g	0.03	<0.25
Methanol	EN 14110	g/100 g	<0.02	<0.2
Ester content	EN 14103	g/100 g	98.9	>96.5
Sodium (Na)	EN 14108	mg/kg	<0.5	<0.5
Potassium (K)	EN 14109	mg/kg		
Magnesium	EN 14538	mg/kg	<0.5	<0.5
Calcium	EN 14538	mg/kg		
Phosphorus	EN ISO 2624	mg/kg	<1	<10
Higher calorific value		MJ/kg	41.3	not specified
Lower calorific value		MJ/kg	38.9	not specified
Oxygen content		%	11.2	not specified

Source: Cooperation Project DaimlerChrysler, Germany; Central Salt and Marine Chemicals Research Institute, India; and University of Hohenheim, Germany.

vectors in animals as well. Jatropha kernels contain varying concentrations of these esters, ranging from less than 1 mg/g to more than 7 mg/g. On average, 70% of all phorbol esters are retained in the oil and the rest is found in the deoiled kernel meal [21]. The extraction of the esters from the oil is comparatively easy, unlike from the meal, which may relate to the plant matrix.

Jatropha oil or a methanol extract of Jatropha oil containing phorbol esters has been shown to have strong insecticidal effects against *Busseola fusca* and *Sesamia calamistis* larvae [22] and *Lipaphis erysimi* [23], and pesticidal effects against *Sitophilus zeamays* and *Callosobruchus chinesis*, deterring their oviposition on sprayed corn and mungbean seeds [24]. These effects are expected to be due to the presence of phorbol esters. Extracts from *J. curcas* L. were found to be toxic to snails transmitting *Schistosoma mansoni* and *S. haematobium* [25]. Compared to aqueous extracts, methanol extracts showed the highest toxicity against all tested organisms with lethal concentration $(LC)_{100}$ values of 25 ppm for cercariae and the snail *Biomphalaria glabrata* and 1 ppm for the snails *Bulinus truncatus* and *B. natalensis*. Phorbol esters at a level of 1 ppm in water also killed all snails of the *Physa* specie, which are also known to be intermediary hosts of schistosomes responsible for causing the deadly disease schistosomiasis. Phorbol esters, when extracted from oil, could have applications as biopesticides in organic as well as conventional agriculture. In addition, they could also be used to control diseases such as schistosomiasis [26, 27]. Although, no concrete data are available on the fate of phorbol esters in the environment, they are considered to degrade completely in soil within 6 days [25]. In order to exploit phorbol esters for various applications, we have recently developed conditions for the isolation of phorbol esters from Jatropha oil and studied their stability under different temperatures. Phorbol esters change their nature during storage and become inactive. Shelf life enhancement studies are also being undertaken in our laboratory. The extraction of phorbol esters from oil, in addition to providing an invaluable product, will also make the process of biodiesel production and use friendly to the worker and environment.

Antiflammatory compounds such as the flavonoids apigenin and its glycosides vitexin and isovitexin, the sterols stigmasterol, beta-D-sitosterol and its beta-D-glucoside [28] are known to be present in leaves. The Jatropha latex has a

proteolytic enzyme, curcain, which has been demonstrated to have wound-healing properties [29]. A novel cyclic octapeptide, curcacycline (Gly-Leu-Leu-Gly-Thr-Val-Leu-Leu-Gly), present in Jatropha latex has been shown to inhibit the classical pathway activity of human complement and the proliferation of human T cells [30]. Jatropha seeds are good sources of phytate. Several beneficial effects of phytate including cancer prevention, reduction in iron-induced oxidative injury and reversal of initiation of colorectal tumorigenesis, and prevention of lipid peroxidation have been reported [31].

The oil has been used for decades for the production of high-value soap in Mali and other African countries. Antimicrobial activity has been reported in oil [32], leaves [33, 34], and roots [35]. Enzymes such as lipases and proteases have been produced using solid-state fermentation with *Pseudomonas aeruginosa* on seed cake [36]. Seed cake could also be a good substrate for the production of other industrial enzymes. Curcin present in seeds has been shown to have antitumor effects [37] and anti-fungal activity [38]. An enzyme (β-1,3-glucanase) isolated from seeds exhibited antifungal activity against *Rhizoctonia solani* Kuha. and *Gibberelle zeae* Schw. [39]. Recently, the anti-HIV effect of 12-deoxyphorbol-13-phenylacetate, a compound synthesised from Jatropha phorbol esters, has been demonstrated. It inhibits HIV entry into target cells [40].

Several unsubstantiated claims have been made in non-peer-reviewed articles, *e.g.* that Jatropha oil is a good topical ointment for pain relief in rheumatism, for skin diseases and for stimulating hair growth. Jatropha latex is an antiseptic and analgesic for wounds. In parts of Asia, Jatropha root is used as antidote for snakebite, and in parts of Africa, Jatropha kernel is used for the termination of unwanted pregnancies. Jatropha leaves are used against ringworm in Nigeria. These pieces of information could lay the foundation for isolation of the active compounds from Jatropha plant parts and the study of their pharmacological properties. This could lead to knowledge-based rational exploitation of various parts and products of the Jatropha plant. There is a need to provide a sound scientific basis to these claims. Phorbol esters have been shown to have anti-microbial activity, and results from our laboratory show that phorbol esters are present in almost all parts of the Jatropha plant (Table 2). Some of the uses of various parts of the Jatropha plant could be attributed to the presence of phorbol esters in these plant parts. Certainly, practicing some of the claims would not be able to elicit the desired effects when tested using rigorous scientific methods and principles. For example, Jatropha bark has been considered as a good source of blue dye and tannins [41]. We analysed the tannin content of bark and found that the tannin content is very low (outer dark bark: tannins 0.7% and condensed tannins 0.2%; inner green bark: tannins 3.1% and condensed tannins 1.7%; tannins as tannic acid equivalent and condensed tannins as leucocyanidin equivalent), suggesting that bark does not have the potential to be used as a source of tannins.

Table 2. Phorbol esters in different parts of the toxic *Jatropha curcas* plant.

Parts	Phorbol esters [mg/g dry matter][§]
Kernel	2.00–6.00
Leaves	1.83–2.75
Stems	0.78–0.99
Flower	1.39–1.83
Buds	1.18–2.10
Roots	0.55
Latex	not detected
Bark (outer brown skin)	0.39
Bark (inner green skin)	3.08
Wood	0.09

[§] As phorbol-12-myristate 13-acetate equivalent

2.5 Co-products for energy generation

The seed shell of Jatropha has 45–47% lignin and has a high energy value (~19.5 MJ/kg). Related to the gross energy content, 2.1 kg of shells is equivalent to 1 kg of fossil oil. The husk (dried fruit encapsulate) of Jatropha also has a high energy content (15.6 MJ/kg) [42] and, hence, both these materials could be used for generating energy through burning. The portion of the fruit husk is in the range of 37–40%, and the shell portion of the seed is approximately 35% on dry matter basis. It has been found that the highest oil content of seeds is attained when the fruit changes its colour from green to yellow. If optimal agronomic practices are being followed, the high-water-containing fruit husk will not be available as an energy source but will be taken back to the plantation as a soil amendment. In addition, both husks and shells are not suitable as substrates in biogas digesters because of their very low digestibility and thus degradability.

2.6 Jatropha and its possible socio-economic impacts

Most of the 500–600 million small farms in the developing world cultivate 20–30% of their land with high production risks. Failure of harvest on that portion of farmland is the rule. Overuse and inadequate management practices are the reasons for the unfertile nature of the land. It is recommended to plant a perennial, like Jatropha, on this portion of land. The very positive influence of this kind of vegetation will help to reclaim this land in a relatively short period of time and make it again suitable for staple crop production. Besides, an energy crop like Jatropha guarantees a sustained cash flow and seems not to be sensitive to price elasticity. Positive influence is also to be expected with respect to labour engagement in the rural areas. We estimate that a year-round labour force of 30 for 100 ha is required if most of the work on the plantation is by hand labour. A bio-energy plant would also save hard currency of developing countries by reducing the import of fossil fuels.

2.7 Jatropha as an energy crop and its impact on food production in the tropical world

From an ecological point of view, planting diverse species would be preferable over establishing large monoculture stands. However, if a single species, such as *J. curcas* were to be identified as the most productive and best adapted among several oil crops, as has been pointed out in an extensive comparative study by Azan *et al.* [3], a diversification of plantations might also be achieved through planting valuable forest species such as Neem, Pinus, Moringa and other forest species with proven adaptation on the nutrient-poor, degraded lands. This could reduce the ecological risk of insect pest or diseases reaching epidemiological levels, although this risk is considered low in the case of Jatropha due to its genetically heterogeneous condition and generally very robust nature. A diversification with interspaced food crops seems possible as well. Research into new agronomic practices is needed before recommendations can be given.

If staple crops are planted in alleys with energy plants, a much higher yield can be attained because the food crops will profit from the nutrition and the shelter effect of the perennials. Under the umbrella of, *e.g.*, the Jatropha plant, maize, sorghum, millet and other staple crops will profit from the advanced management practices of the energy plant. Experiments conducted by the University of Hohenheim in the Sahel have demonstrated that micro-amounts of 4 kg P/ha applied directly to the plant pit of millet almost doubled the grain harvest from 240 to 400 kg/ha [43].

2.8 Jatropha's influence on micro-climate change

Jatropha curcas is a shrub but can also grow out to the size of a tree as large as 12 m high. It is widely distributed in almost all countries in the tropical regions of Africa, Asia and Latin America. *J. curcas* is an eco-friendly plant. Grasses and other shade-loving vegetables could be grown between the rows of Jatropha and cattle are being observed grazing freely on the grass cover inside the plantation. Its ability to survive and sustain a reasonable production of oil seeds within an acceptable time interval on difficult soils and under harsh climatic conditions are factors that give the plant an advantage over other perennial oil seed candidates. *Jatropha curca*s has also been found to thrive in the desert of Upper Egypt, if appropriate water and nutrient supply is ensured. Up to now, the effects of changing vegetation in desert regions on the local climate parameters have not been investigated. The re-greening of denuded areas will have distinct favourable effects on soil surface temperature, wind velocity and soil moisture. These positive parameters may trigger cloud formation and, as a consequence, precipitation. These changes in soil surface vegetation, *e.g.* with *J. curcas* and other oil plants, and their impact on the atmosphere have not been studied so far because the necessary water resources were not available at an acceptable prize up till now. Efficient and cost-effective methods to provide huge amounts of desalinated seawater are now available that could be used for the establishment of vegetation in the hot deserts around the globe.

2.9 Jatropha's impact on soil quality improvement

Wasteland on a global scale is increasing at an alarming rate. There is not much established knowledge currently on the influence Jatropha could have on land reclamation through soil improvement and additional agro-ecological advantages.

Areas where Jatropha is propagated belong to the most production-insecure regions. Because of the plant's excellent adaptation to a large variety of soil and climatic conditions, it seems to be ideal on land that is otherwise not usable anymore.

If the soil is penetrable, the Jatropha roots provide deep-reaching opening avenues for water infiltration. Besides its intensive root structure, it forms an effective barrier to run-off water after heavy rains. Soil erosion could be effectively reduced or completely prevented and the water-holding capacity of the soil can be utilised to a much greater extent. This is of overriding advantage, especially towards the end of the dry season. The increase in soil fertility would quickly lead to a much better vegetation growth on the former barren land. Organic carbon stores in the soil would build up and support and stabilise agronomic efforts again. There is also hope that the biodiversity will increase through imported seeds (wind, water) or by providing a better growth environment for dormant seeds in the soil. It is anticipated that, after a relatively short period of time, conventional food crops can be grown again as allays between the rows of the perennial Jatropha or perhaps any other energy plant. This system of agronomic practices seems to be especially suited for small-scale farms, which in our global world are the most disadvantaged ones.

3 Comparative evaluation of toxic and non-toxic Jatropha genotypes

3.1 Variation in seed number of fruits

The literature reports that the fruits of *J. curcas* contain three seeds [14]. However, fruits with one, two and four seeds were also observed [21]. Within a genotype, the highest percentage of fruits contained three seeds, followed by two seeds (Table 3), for both of the genotypes.

The fruits containing three and four seeds have been found to be significantly more frequent for the non-toxic genotype ($68.5 \pm 6.14\%$ (SD) *vs.* $59.6 \pm 2.23\%$; $p < 0.05$) and also the variation (as is evident from the SD) was higher for the non-toxic genotype [21]. These variations could be exploited in the breeding programmes to develop a line with the desired traits. A higher percentage of fruits with three and four seeds will render a higher yield. With the distribution as shown in Table 3, on average, 100 fruits of the non-toxic and toxic genotypes would yield 270 and 252 seeds, respectively.

Table 3. Variation in seed number per fruit (% of total fruits) of toxic and non-toxic genotypes.

	Percent fruits containing . . .			
Genotypes	1 seed	2 seeds	3 seeds	4 seeds
Toxic	14.6 ± 1.07^{b}	25.7 ± 2.89^{c}	52.0 ± 5.77^{d}	7.6 ± 3.23^{a}
Non-toxic	8.1 ± 3.80^{a}	23.4 ± 3.83^{c}	58.3 ± 5.77^{d}	10.2 ± 5.61^{e}

Values with different superscripts within a column differ significantly ($p < 0.05$).

3.2 Physical parameters

The seed, shell and kernel masses and the shell/seed ratio (or, in other words, the kernel-to-seed ratio) have been observed to be statistically similar ($p > 0.05$), for both the genotypes, among the four groups of seeds described in Table 3. On the other hand, the seed and shell masses of the toxic genotype were significantly higher ($p < 0.05$) than those of the non-toxic genotype; however, the kernel masses of the toxic and non-toxic genotypes were statistically similar [21], suggesting that the higher seed mass of the toxic genotype is contributed by its higher shell mass. From the same number of seeds from the toxic and non-toxic genotypes, the yield of kernels will be the same; however, from the same weight, the yield of shells will be higher from the toxic genotype. The kernel mass as percentage of the seed mass did not differ significantly between the two genotypes (61.5% for the toxic and 63.9% for the non-toxic genotype) [21]. Similar values have been observed for seeds obtained from India, Nicaragua, Cape Verde and Mexico [42, 44, 45].

3.3 Oil and protein contents

The oil and protein contents of seeds and kernels (Tables 4, 5) have been found to be statistically similar amongst the four seed groups stated in Table 3, both for the toxic and non-toxic genotypes. Also, there was no significant difference between the toxic and non-toxic genotype for these parameters (Tables 4, 5). Our other studies, wherein combined seeds were analysed, also report similar protein and oil contents [42, 45–47].

The above results suggest that 1 t of toxic and non-toxic seeds would yield 615 and 639 kg of kernels, with almost identical oil contents of approx. 57.1 and 56.7%, respectively. The potential oil recovery from 1 t would be 351 and 362 kg for the toxic and non-toxic genotypes using solvent extraction. In other words, to obtain 1 t of oil, 2.85 t of toxic and 2.76 t of non-toxic seeds would be required. When the yield is expressed per unit of seed number (*e.g.* 1000 seeds) the yield of kernels and oil is expected to be similar for the two genotypes. However, from the same weight or same number of seeds, the yield of shells (a good source of energy; see below) will be higher for the toxic genotype. It may be concluded from these findings that the yield of oil from the seeds of the non-toxic genotype is not inferior to that of the toxic genotype which is widely used throughout the world. There is a need to evaluate the seed yield per hectare from both the toxic and non-toxic genotypes in different climatic conditions. At present, it is believed that the non-toxic genotype, since it lacks a kind of

Table 4. Oil content (% of dry matter) in seeds and kernels from toxic and non-toxic genotypes.

	Seeds		Kernels	
Fruits with . . .	Toxic	Non-toxic	Toxic	Non-toxic
1 seed	34.8 ± 1.79	35.3 ± 1.56	56.5 ± 1.03	55.5 ± 0.76
2 seeds	35.8 ± 0.78	37.0 ± 1.09	57.2 ± 0.27	57.6 ± 0.94
3 seeds	35.2 ± 1.81	37.1 ± 0.97	57.4 ± 0.50	57.5 ± 0.69
4 seeds	35.5 ± 1.01	36.0 ± 1.57	57.1 ± 0.42	56.2 ± 0.77

For seeds and kernels separately, the values for toxic and non-toxic genotypes did not differ significantly ($p > 0.05$).

Table 5. Crude protein content (% of dry matter) in seeds and kernels from toxic and non-toxic genotypes.

	Seeds		Kernels	
Fruits with . . .	Toxic	Non-toxic	Toxic	Non-toxic
1 seed	16.6 ± 1.27	18.2 ± 0.74	27.0 ± 1.99	28.5 ± 0.87
2 seeds	16.5 ± 0.63	17.3 ± 1.39	26.3 ± 0.73	26.9 ± 1.84
3 seeds	16.3 ± 1.07	17.3 ± 0.94	26.6 ± 1.12	26.8 ± 1.25
4 seeds	16.7 ± 1.11	18.2 ± 0.63	26.8 ± 1.65	28.4 ± 1.22

For seeds and kernels separately, the values for toxic and non-toxic genotypes did not differ significantly ($p > 0.05$).

toxin (phorbol esters, which are plant defence compounds), would be more prone to various environmental vagaries including pests and diseases. With proper care and management practices, as for any other edible oil seed crop, the non-toxic genotype could give edible oil and seed cake for use in the diets of farm animals and aquaculture species.

3.4 Chemical composition and digestibility of Jatropha kernel meal

The contents of crude protein, ash, gross energy and neutral detergent fibre of kernel meal (residue left after solvent extraction of oil from kernels, with kernels being the inner white material left after removal of the shells) are similar for the two genotypes. Sugar and starch contents (Table 6) and the amino acid composition of the toxic and non-toxic genotypes are almost identical (Table 7). The levels of all essential amino acids, except for lysine, are comparable with the FAO reference protein for a growing child of 2–5 years of age (Table 7). A comparison between the amino acid composition of Jatropha meal and soybean [48] revealed an almost identical pattern for all essential amino acids, except for lysine and the sulphur amino acids; lysine is lower and the sulphur amino acids are higher in the Jatropha meals. The levels of essential amino acids in the Jatropha meals are higher than or similar to those of castor bean meal [42]. The non-protein nitrogen in Jatropha meal formed only 9.0% of the total nitrogen in the Jatropha meals, suggesting the presence of high levels (91%) of true protein [42]. The high protein utilisation efficiency in rats and the rapid growth observed in fish fed non-toxic Jatropha meal [49] suggested that the protein quality of Jatropha kernel meal is very high.

Table 7. Amino acid composition (g/16 g nitrogen) of kernel meal from toxic and non-toxic genotypes.

Amino acids	Toxic	Non-toxic	Soybean meal	FAO (ref. protein)
Essential				
Methionine	1.91	1.76	1.22	2.50[§]
Cystine	2.24	1.58	1.70	
Valine	5.19	5.30	4.59	3.50
Isoleucine	4.53	4.85	4.62	2.80
Leucine	6.94	7.50	7.72	6.60
Phenylalanine	4.34	4.89	4.84	
Tyrosine	2.99	3.78	3.39	6.30
Histidine	3.30	3.08	2.50	1.90
Lysine	4.28	3.40	6.08	5.80
Arginine	11.80	12.90	7.13	
Threonine	3.96	3.59	3.76	3.40
Tryptophan	1.31	ND	1.24	1.10
Non-essential				
Serine	4.80	4.82	5.67	–
Glutamic acid	14.68	15.91	16.90	–
Aspartic acid	9.49	9.92	11.30	–
Proline	4.96	3.80	4.86	–
Glycine	4.92	4.61	4.01	–
Alanine	5.21	4.94	4.23	–

ND, Not determined.
[§] Methionine plus cystine.

Digestibility and metabolisable energy of heat-treated (121 °C, 66% moisture, 30 min) kernel meal, using the *in vitro* gas method [50], were similar for the toxic and non-toxic genotypes, but were lower compared to those for soybean meal by 10% units and by 2.5 MJ/kg dry matter (Table 8). The

Table 6. Chemical composition, sugar and starch contents (%) of kernel meal from toxic and non-toxic genotypes.

	Crude protein	Residual lipid	Ash	Neutral detergent fibre	Total sugar	Starch
Toxic	60.3–62.4	1.5	9.6	18.2	7.7–10.3	9.4–11.2
Non-toxic	60.2–63.8	1.0	9.8	18.0	10.2	10.6

Table 8. Digestible organic matter, metabolisable energy, protease digestibility and rumen-degradable nitrogen of heat-treated (121 °C, 66% moisture, 30 min) kernel meal.

	Toxic	Non-toxic	Toasted soybean meal
Digestible organic matter [%]	78.0	77.3	87.9
Metabolisable energy [MJ/kg]	10.9	10.7	13.3
Pepsin + trypsin digestibility [% of total nitrogen]	89.0	90.1	91.1
24 h *in vitro* rumen-degradable nitrogen [% of total nitrogen]	43.3	28.9	80.9

digestibility of the Jatropha kernel meal protein, determined by treatment with pepsin followed by trypsin, was similar to that of toasted soybean meal, whereas the *in vitro* rumen digestibility of nitrogen was lower by approx. 50% [42], suggesting that Jatropha kernel meal has a high level of rumen-undegradable protein which might be available post-ruminally. These results imply that Jatropha kernel meal from the non-toxic genotype could be an excellent protein source for animals having the potential to give high milk, meat and wool yields. This would also be expected for the Jatropha kernel meal from the toxic genotype once it has been detoxified.

3.5 Anti-nutritional factors in kernel meal from toxic and non-toxic genotypes

The trypsin inhibitor activity has been found to be similar in the toxic and non-toxic genotypes (Table 9), and these values are similar to that in raw soybean meal [42]. The lectin activity of both the toxic and non-toxic meals, as determined by haemagglutination assay, was almost identical. Curcin is a lectin and the similar haemagglutination of the toxic and non-toxic genotypes suggests that curcin is not the principle toxin present in Jatropha seeds. However, it would be interesting to compare the ribosomal inhibition activity of the toxic and non-toxic genotypes since curcin is known to possess this activity. These studies are in progress in our laboratory. The phytate content of both genotypes is almost identical, and it is very high (ca. 9%). Phytate is known to decrease the absorption of minerals, particularly calcium, zinc and iron. Therefore, the addition of phytase enzyme should be considered for feeds containing kernel meal from the non-toxic Jatropha genotype, to mitigate the adverse effects of phytate. Tannins, cyanogens, glucosinolates and amylase inhibitors have not been detected in any of the Jatropha meals [50]. Saponins were present in kernel meal of both genotypes (2.6–3.4%); however, these saponins did not possess haemolytic activity. The level of non-starch polysaccharides was also similar in the two genotypes (Table 10). The corresponding non-starch polysaccharide content of soybean meal, rapeseed meal, cottonseed cake, linseed meal, coconut cake, palm cake and sunflower cake are 15.5, 17.8, 16.4, 19.3, 25.0, 36.8, 39.3 and 19.3% (calculated from the data of [51]). The non-starch polysaccharide level observed in Jatropha meal is similar to that in soybean meal and lower than in other conventional protein-rich feed resources. They do not appear to elicit adverse effects in common carp (*Cyprinus carpio*) and Nile tilapia at 75% replacement of fishmeal protein in the diet by heated meal from the non-toxic genotype of Jatropha, since the growth of both these species of fish was as good as for the fish fed a 100% fishmeal protein diet.

3.6 The case of phorbol esters to be the main toxic principle

Phorbol esters were absent in kernel meal from the non-toxic genotype but were present in high concentrations in the kernel meal from the toxic genotype (Table 9). Phorbol esters, diterpenes of phorbol type, cause severe toxic symptoms in livestock. At least six phorbol esters are present in Jatropha seeds [52]. The phorbol esters are reported to mimic the action of diacylglycerol, an activator of protein kinase C which regulates different signal transduction pathways. Interference with the activity of protein kinase C affects a number of processes including phospholipid and protein synthesis, enzyme activities, DNA synthesis, phosphorylation of proteins, cell differentiation and gene expression. They are also co-carcinogens and have purgative and skin-irritant activities. In humans, accidental poisoning by Jatropha seeds has been reported to elicit giddiness, vomiting and diarrhoea. Mortality has also been reported in a number of animal species, *e.g.* mice, chicks and goats [27, 53].

Phorbol esters are heat stable and, hence, heat treatment is not effective to detoxify kernel meal from the toxic genotype. On the other hand, trypsin inhibitor and lectins are heat labile and can be destroyed by moist heating [44]. The seeds of non-toxic Jatropha are roasted and the kernels are consumed by humans in certain regions of Mexico. Consumption of unroasted seeds of non-toxic Jatropha is known to produce

Table 9. Levels of anti-nutritional and toxic factors in kernel meal of toxic and non-toxic genotype.

Component	Toxic	Non-toxic
Phorbol esters [mg/g kernel][§]	2.79	ND
Total phenols [% tannic acid equivalent]	0.36	0.22
Tannins [% tannic acid equivalent]	0.04	0.02
Phytates [% of dry matter]	9.40	8.90
Saponins [% diosgenin equivalent]	2.60	3.40
Trypsin inhibitor [mg trypsin inhibited/g sample]	21.31	26.54
Lectin activity [1/mg of meal that produced haemagglutination/mL assay medium]	51–102	51–102

ND, Not detected.
[§] As phorbol-12-myristate 13-acetate equivalent.

Table 10. Levels of constituent sugars of non-starch polysaccharides in kernel meal (% of dry matter) of toxic and non-toxic genotypes (using the method of Englyst *et al*. [60]).

	Rha	Fuc	Ara	Xyl	Man	Gal	Glu	GlcA	GalA	Total-NSP
Toxic	0.2	0.1	2.5	1.2	0.3	1.2	4.7	0.9	2.6	12.7
Non-toxic	0.2	0.1	2.7	1.4	0.3	1.2	4.7	0.0	3.0	13.6

Rha, rhamnose; Fuc, fucose; Ara, arabinose; Xyl, xylose; Man, mannose; Gal, galactose, Glu, glucose; GlcA, glucuronic acid; GalA, galacturonic acid; NSP, non-starch polysaccharides.

discomfort in humans, and this could be due to the presence of trypsin inhibitor and lectins. The roasting treatment has been found to reduce the level of trypsin inhibitor completely. The lectin activity decreased by approximately 50% on roasting and the phytate level remains unchanged [46]. Consumption of large amounts of kernels from the roasted seeds might produce discomfort due to the remaining lectin activity.

The protein efficiency ratio, weight growth and intake for rats fed diets containing raw non-toxic Jatropha kernel meal was significantly lower than for a diet containing heated (66% moisture, 121 °C, 30 min) Jatropha kernel meal. The heat treatment inactivated the trypsin inhibitor and the lectins and also increased the protein digestibility, leading to better performance of rats on heat-treated kernel meal. However, when raw and heat-treated Jatropha kernel meal from the non-toxic genotype was fed to carp (*C. carpio*), both groups grew to an almost identical extent [49, 53]. On the other hand, a diet containing kernel meal from the toxic Jatropha, when fed to carp 3–5 g of body weight, decreased the body mass in 4 days. The fish refused the diet on the second day of feeding and abundant mucus was seen in the aquarium, but no fish died. Similar effects were observed when purified phorbol esters from Jatropha were mixed in standard fish feed at a level of 2 mg/g [53].

From the above results, it is evident that the main toxic principle present in Jatropha seed meal is phorbol esters. Trypsin inhibitor, lectins and phytate might aggravate the adverse effects, but are not responsible for acute toxicity.

3.7 Heated kernel meal from non-toxic Jatropha as fish feed

The heat-treated (121 °C, 66% moisture, 30 min) kernel meal was incorporated into carp (*C. carpio*) diet (crude protein level: 38%) at a level of 75% replacement of fishmeal protein and fed to carp. The diet also contained 500 IU of phytase. The extent of growth of fish fed the heated kernel meal was similar to that of the fishmeal-fed fish. In 50 days, both groups grew by 220% from an initial weight of 5 g (our unpublished results). These results suggest that the kernel meal obtained from the non-toxic genotype is an excellent fish feed, and is also expected to be an excellent protein source for other high-yielding farm animal species. These results also show that non-starch polysaccharides, even though present in the Jatropha meal at a high level (12–13%), do not appear to produce any adverse effects and hence could be ascribed as benign in nature.

3.8 Detoxification of kernel meal from the toxic genotype and use as fish feed

Carp (*C. carpio*) is highly sensitive to toxins and can detect phorbol esters at a level of 15 ppm [54]. Therefore, we have been using carp as an animal model for the development of detoxification conditions. Recently, we have detoxified kernel meal from the toxic genotype (patent application submitted) and fed to carp. Fingerlings (250; average weight 3.2 g) were randomly distributed into five treatment groups with four replicates and fed iso-nitrogenous diets (crude protein 38%): T_1 (standard diet, fishmeal-based protein), T_2 and T_3 (50 and 75% of fishmeal protein replaced by soybean meal), and T_4 and T_5 (50 and 75% fishmeal protein replaced by detoxified Jatropha kernel meal). The body weight gain and specific growth rate were statistically similar for T_1 and T_4 and significantly higher compared to other groups. The performance of the 75% detoxified Jatropha kernel meal group was comparable to the 75% soybean meal group, but was lower than that of the fishmeal group; however, the performance of the 50% detoxified Jatropha kernel meal-fed group was better than that of the 50 and 75% soybean meal-fed groups and similar to that of the fishmeal group. None of the blood parameters examined showed signs of toxicity [55].

3.9 Preparation and characterisation of protein concentrate

Large-scale plantation of the toxic genotype of Jatropha has taken place in many developing countries, with the aim of using the oil as biodiesel. In these countries, the oil is produced from whole seeds using a screw press. The seed cake left as a by-product after oil extraction by screw press can contain as much as 500 g/kg of shells as the indigestible material. Therefore, there is a need to separate the high-quality protein from the shells. Using the principle of isoelectric precipitation, the protein concentrate prepared from

the screw-pressed cake obtained from the toxic genotype contained a substantial amount of phorbol esters (0.86–1.48 mg/g), trypsin inhibitor, lectins and phytate. The amino acid composition of the protein concentrate mirrored that of the kernel meal and the available lysine was unaffected by the treatment of producing the protein concentrate [56]. As for the kernel meal, the *in vitro* rumen protein digestibility of the protein concentrate was low and the protein digestibility using pepsin and pancreatin was high [57], suggesting a high-value protein concentrate for high-yielding animals. To make the protein concentrates suitable for use as an ingredient in livestock feed, the phorbol esters must be removed and the trypsin inhibior and lectins inactivated by heat treatment. The adverse effects of phytate could be mitigated by addition of phytase in the diet. For detoxification of the protein concentrate, we are evaluating the efficacy of the detoxification treatment developed for the kernel meal. The procedure optimised [56] for the toxic genotype could also be used for the non-toxic genotype. The protein concentrate will be free of phorbol esters, but would contain other antinutritional factors such as trypsin inhibitor, lectins and phytate. This material, after heat treatment, would form an excellent protein supplement for farm animal species.

3.10 Fatty acid composition and physical properties of oil

The fatty acid composition of the solvent-extracted oil from both the toxic and non-toxic genotypes obtained from Mexico is given in Table 11. There are considerable differences in the levels of oleic and linoleic acids between the toxic and non-toxic oils. Oleic acid is higher in the toxic oil, and linoleic acid in the non-toxic oil. These differences appear to be genetically controlled. The non-toxic oil has a potential to be used as edible oil, and the higher level of linoleic acid (polyunsaturated fatty acid) could be considered advantageous for human health. On the other hand, the higher level of this fatty acid in the non-toxic oil is expected to decrease the oxidative stability of the biodiesel produced from this oil, although the oxidative stability indices did not differ substantially for the biodiesel samples produced from the toxic and non-toxic oils (see below).

3.11 Quality parameters of biodiesel

Water content, acid value, element content (P, Ca + Mg and Na + K) and free and total glycerol of all biodiesel samples produced from oil obtained from the toxic and non-toxic genotype of *J. curcas* were within the European EN 14214 specification (Table 12). The cloud point of biodiesel from the non-toxic oil was lower (0.6 °C) compared to that from the toxic oil (1.9 °C). The difference in cloud point can be directly related to the lower stearic acid content of the non-toxic oil [58]. The values for other parameters such as density, viscosity, flash point, cetane number and oxygen content for the

Table 11. General composition of oil from toxic and non-toxic Mexican genotypes of *Jatropha curcas*.

Fatty acid composition [%]	Non-toxic	Toxic
Myristic, 14:0	0.2	0.1
Palmitic, 16:0	13.4	15.3
Heptadecanoic, 17:0	0.1	0.1
Stearic, 18:0	6.4	6.6
Arachidic, 20:0	0.2	0.2
Behenic, 22:0	traces	traces
Lignoceric, 24:0	traces	0.1
Total saturated	*20.3*	*22.3*
Palmitoleic, 16:1*n*-7	0.8	0.9
Oleic, 18:1*n*-9	36.5	41.0
Eicosenoic, 20:1*n*-9	0.1	0.1
Total monounsaturated	*37.3*	*42.0*
Linoleic, 18:2*n*-6	42.1	35.3
α-Linolenic, 18:3*n*-3	0.2	0.3
Total polyunsaturated	*42.3*	*35.7*
Elements [ppm]	**Non-toxic**	**Toxic**
P	54.90	87.90
Ca	32.80	51.10
Mg	ND	23.90
Na	1.48	13.30
K	6.57	15.30
Fe	0.07	8.31
Iodine value	108.1	96.5
Calorific value	37.8–38.1 MJ/kg	37.5–38.0 MJ/kg
Appearance	light yellow liquid	light yellow liquid

ND, not detected.

Jatropha biodiesel could be obtained from [17, 59]. These parameters also met the EN 14214 standards. Unblended 100% Jatropha biodiesel was tested extensively on the road in India with modern CDI Mercedes cars. A total of 80,000 L of biodiesel was used in these tests. The overall results were highly satisfactory. Differences were found for emission parameters, specifically sulphur and particulate matter, which are 80% lower than in mineral diesel. These tests have also shown that there is only a marginally better efficiency of 1.7% in favour of mineral diesel in fuel consumption.

3.12 Phorbol esters in Jatropha oil, biodiesel and glycerol

A potential major constraint in the widespread acceptance of *Jatropha* as a source of biodiesel could be the presence of phorbol esters, which, when consumed by man and animal, are toxic and are also co-carcinogens [27]. This makes the oil unsuitable for food and feed applications. In view of the cur-

Table 12. General quality parameters of the produced Jatropha biodiesel samples.

Parameters	EN 14214	Non-toxic	Toxic
Water [ppm]	maximum 500	216	290
Acid value [mg KOH/g]	maximum 0.50	0.08	0.16
Cloud point [°C]	–	0.6	1.9
Oxidative stability at 110 °c [h]	minimum 6	7.9	5.9
Elements [ppm]			
P	maximum 10	0.03	1.00
Ca + Mg	maximum 5	ND	ND
Na + K	maximum 5	ND	0.05
Free and total glycerol [%]			
Free glycerol	maximum 0.02	0.005	0.005
Monoglycerides	maximum 0.80	0.73	0.72
Diglycerides	maximum 0.20	0.17	0.21
Triglycerides	maximum 0.20	0.06	0.06

ND, not detected.
Source: Makkar *et al.* [58].

Table 13. Phorbol ester content[§] (mg/g) of the different fractions obtained during pre-treatment and transesterification of different Jatropha oil samples.

Parameters	Toxic	Non-toxic
Crude oil	3.10	ND
Degummed oil	2.68	ND
Acid gums	2.01	ND
Wash water	1.72	ND
Silica-treated oil	2.82	ND
Stripped oil	ND	ND
Fatty acid distillate	ND	ND
Biodiesel	ND	ND
Crude glycerine	ND	ND
Biodiesel wash water	ND	ND

[§] As phorbol-12-myristate 13-acetate equivalent
ND, Not detected.
Source: Makkar *et al.* [58].

rent debate of 'food *versus* fuel', however, this toxicity is a potential advantage for *Jatropha*. *Jatropha* oil can be seen as a 'technical oil' and therefore does not compete directly with the food markets. At the same time, this can also be a disadvantage. Due to the toxicity of the oil, special precautions might need to be exercised during the processing of *Jatropha curcas* seeds and oils. By-products of the vegetable oil pre-treatment and biodiesel production process, such as fatty acid distillate (FAD), gums and glycerine have several applications in the food and feed industry, and the presence of phorbol esters could render them unfit for edible purposes. In one of our studies, we follow the flow of phorbol esters during various stages of pre-treatment and biodiesel production from *Jatropha* oil. The results are presented in Table 13.

During degumming, some phorbol esters were removed in the acid gums and wash water. This implies that the use of these acid gums in animal feed is not possible and care should be taken while disposing of the wash water into the environment. Silica treatment did not decrease the phorbol esters, while stripping/deodorisation at 260 °C at 3 mbar pressure with 1% steam injection completely degraded the phorbol esters. Phorbol esters were not detected in stripped oil, FAD, transesterified oil (biodiesel) and glycerine. However, the presence of possibly toxic phorbol ester degradation products in these fractions could not be ruled out [58]. At present, no information is available on the nature or toxicity of the possible degradation products.

Although phorbol esters were not detected in the biodiesel and glycerine samples in the present study, phorbol esters were detected in biodiesel (Company 1: 0.46–1.20 mg/g; Company 2: 1.16 mg/g) and glycerine (Company 1: 0.67–0.97 mg/g; Company 2: 0.13 mg/g) samples obtained from other industrial plants. These results suggest that different oil pre-treatment conditions could affect the presence of phorbol esters in biodiesel or glycerine produced from toxic *Jatropha* oil. Evaluation of possible phorbol ester degradation products should be conducted in the exhaust of engines using phorbol ester-containing biodiesel.

4 Future studies

The yield and quality of the oil from seeds of the non-toxic genotype of Jatropha are similar to those of the toxic genotype. Studies for a comparative evaluation of the two genotypes for their seed yield and disease susceptibility should be conducted. Selection, breeding and agronomic studies for both genotypes need to be undertaken.

Various bioactive moieties and their pharmaceutical and biological effects appear to have been reported using the toxic genotype of Jatropha. It would be interesting to examine the presence of activities in various parts of the non-toxic Jatropha plant.

The oil from the toxic genotype could be freed of phorbol esters using the deodorisation or stripping process, an oil pre-treatment process, during the process of biodiesel production. The deodorisation/stripping process could be optimised to obtain oil free of phorbol esters. Phorbol ester degradation products could possibly be present in the treated oil so obtained. The toxicity of the stripped oil free of phorbol esters should be investigated using rat and fish as experimental models. Should it be found innocuous in the feeding studies, the toxic Jatropha oil could be turned into an edible oil. The fatty acid composition of Jatropha oil is close to olive oil and the Jatropha oil free of phorbol ester and its degraded products therefore would be a high-value product.

Using the non-toxic genotype, normally, the oil would be used as edible oil; the spare oil, if available, could be turned into biodiesel, with the generation of glycerol as a by-product

for edible uses. The seed meal obtained from the non-toxic genotype would find application as livestock feed. On the other hand, for the toxic genotype, the oil would be turned into biodiesel with the production of glycerol and other co-products for various industrial applications. The use of stripped oil obtained from the toxic oil as an edible oil is also a possibility. The detoxified seed meal would be used as fish or livestock feed. The seed cake from the toxic genotype could also be used as a fertiliser or as a substrate for the production of industrial enzymes through fermentative processes. Another plausible scenario would be the extraction of phorbol esters from the toxic oil for use as a high-value biopesticide, and then use of the oil, now containing low levels of phorbol esters, as a feedstock for biodiesel production. A comparative life cycle analysis taking into account the various uses/possibilities listed here would help to assess the environmental impact of producing and using the toxic and non-toxic oils. The comparative life cycle analysis coupled with a socio-economic analysis would lead to the identification of sustainable approaches for exploitation of the Jatropha plant.

Acknowledgment

We thank Mrs. Sabine Nugent for excellent support in formatting the article to meet the requirements of the journal.

Conflict of interest statement

The authors have declared no conflict of interest.

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