Bio-diesel waste as tailored organic fertilizer for improving yields and nutritive values of *Lycopercicum esculatum* (tomato) crop

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Abstract

The present study focused on utilization of the de-oiled *Jatropha* cake, a bio-waste from the bio diesel extraction process, as organic fertilizer. Tomato crop was subjected to different treatments comprising varying composition of the *Jatropha* cake fertilizer, applied at varying age of plant life-cycle. The impact on yield, morphological and nutritional parameters was studied. Results revealed that an optimal (2-3 % by weight) dose applied on the soil at the time of onset of flowering (around 45 days), significantly enhanced the yield along with improvements in morphological parameters. There were significant improvements in key nutrients viz. protein, vitamin C and reducing sugars. High value of key nutrients like TSS (total soluble solid), vitamin C, proteins, reducing sugar and tomato fruit yield were obtained with soil + 3% cake mixed in 60 day transplant.

These results indicate that composted de-oiled *Jatropha* cake has tremendous potential of being utilized in a cost effective way as tailored organic fertilizer, as a replacement of chemical fertilizers.

Keywords: *Jatropha*, *Lycopercicum esculatum*, Lycopene; Organic fertilizers, Tomato.

1. Introduction

Excessive use of chemical fertilizers and pesticides adversely affects the soil environment, leading to declining crop productivity and production of potentially harmful food, unsafe for human consumption. These unintended harmful effects and the energy intensive processes involved in the production of inorganic fertilizers have resulted in several research initiatives for developing organic fertilizer alternatives (Chaturvedi *et al*., 2012). Recently, use of several types of bio-waste have shown potential of being used as fertilizers like *Jatropha*, Neem, Salvadora, *Maduca* and *Pongamia* not only to improve the organic content of soil vis-à-vis acting as effective insecticides and fungicides (Chaturvedi *et al*., 2008a; Chaturvedi *et al*., 2008b).

Jatropha curcas is currently gaining lots of interests for biofuel production and reclaiming marginal lands. *Jatropha curcas* (L) or physic nut is a multipurpose and drought resistant large shrub or small tree and can be grown without much effort (Kumar and Sharma, 2008; Kumar *et al*., 2010; Kumar and Sharma, 2011). Seed production from plants propagated from seeds can be expected within 2-3 years. Seed of the plant is the most valuable product and used for production of biodiesel. The residue oil cake generated during oil extraction biodiesel production can be used as fertilizer (Cano-Asseleih *et al*., 1989; Jayasigh, 2003). *Jatropha* oil cake is rich in nitrogen, phosphorous and potassium and can be used as organic manure (Jayasigh, 2003; Kumar and Sharma, 2008). The cake is rich in nitrogen (3.2%) and phosphorus (1.4%) and potassium (1.3%) and can be used as manures; however, this content varies depending on the source. The large amount of de-oiled seed cakes produced by oil-extraction process is valuable as organic manure due to its high nitrogen content (up to 6.48%) and can be used to maintain fertility of soil (Singh *et al*. 1996; Chaturvedi *et al*., 2008a; Chaturvedi *et al*., 2010). The presence of toxic component, phorbol ester in *Jatropha* cake discourages people from using this nitrogen and phosphorus rich cake as animal feedstock also an important environmental concern, if utilized as compost (Das *et al*., 2011). Low quantity of phorbol ester in *Jatropha* compost also reduces the possibility of accumulation in plants grown on compost treated soil.

Many studies have demonstrated the efficacy of *Jatropha* based organic fertilizers (Sharma *et al*., 2008; Sinha *et al*., 2011), there have been inadequate research efforts to develop tailor made organic fertilizers for specific crops or plants that fulfill the essential nutrient requirements of those plant species at various stages of its growth. The present research has utilized *Jatropha* oilseed cake, a by-product of the *Jatropha* based bio-diesel extraction process in composted form, as organic fertilizer, to study its impact on yield of *Lycopercicum esculatum* (tomato). Presented research aimed to develop organic fertilizer compositions having synergistic mixtures of seed cakes produced from *Jatropha* based oil extraction process, applied at specific time of tomato crop life cycle. This research aimed to develop compositions that are tailor made (including composition of oil seed cake and time of application/transplant into treated soil) organic fertilizer application for the tomato to get improving yields without compromising nutrient quality of fruit *Lycopercicum esculatum*.

2. Materials and Methods

The study was conducted at National Botanical Research Institute (NBRI), Lucknow, India (27.10°N and 81.13°E) and *Jatropha* cake was obtained from a local bio-diesel extraction expeller unit situated at NBRI, Lucknow. Composting process of the deoiled cake involved developing rectangular compost pits (0.67 m x 0.33 m x 1.32 m). *Jatropha* deoiled cake (20 kg) was grounded and placed on a floor of the pit and covered by a layer of straw and then covered by soil and 60% moisture was maintained throughout composting. The process was carried out for 6 months as described by Chaturvedi *et al*. (2009). The composition of composted *Jatropha* seed cake was determined by kjeldahl digestion (Nelson and Sommers, 1972), phosphorus by Spectrophotometrically (Olsen *et al*., 1954). For estimation of micronutrients in soil, digestion was carried out with acids. This digested clear sample was taken through Atomic Absorption Spectrophotometer and Ca, Fe, Cu & Zn contents were estimated.

The experiment was conducted in two seasons. In the first year, trials with wide range of *Jatropha* deoiled cake proportions in soil were undertaken. The seedlings of cultivar *Bezosheetal* were raised on the nursery bed prepared in the normal soil without fertilizer treatment. These were then transplanted at the time of flowering (i.e. around 40 day's age) to pots prepared with varying proportions of composted *Jatropha* de-oiled cake. It was noted that for soil containing composted de-oiled cake at 4-8%, the plants growth was adversely affected and above 10% cake compositions, plants did not survive. This possibly indicates development of toxicity due to presence of higher doses of cake.

Therefore, experiments were conducted with 1% (T1), 2% (T2) and 3% (T3) composted de-oiled cake in normal garden soil and filled in earthen pots of 6 kg capacities.

In Indian climate, a tomato plant reaches flowering stage after about 35 days and fruiting stage after about 60 days. Therefore, transplant from nursery to the treated pots was carried out at three stages of growth i.e. just before flowering (35 days or D1), onset of flowering (40 days or D2) and onset of fruiting (60 days or D3) (Chaturvedi *et al*., 2009).

The pots for control were also prepared under three different ages of transplants and randomized design was used. This way, the total number of treatments were 12 (4 for different doses of organic fertilizers including control applied for three different ages of transplant). Three replicate pots for each treatment were used. For analysis, samples from each treatment were collected at the red ripe stage of the biological cycle.

The morphological parameters like plant height, number of leaves, flowers per plant, number of fruits and yield were recorded after 35^{th}, 40^{th} and 60^{th} day of transplant. The following nutrient parameters were analyzed in tomato fruits like; Total Soluble Solids (TSS), vitamin C, protein, reducing sugars, lycopene and pectin. TSS content of the fruit was measured by Hand Refractometer (Erma, Japan) by putting small quantity of pulp between the prisms. The TSS was expressed in terms of degree Brix. Lycopene was analyzed using Mohr's titration method of Ranganna (1986), vitamin C was estimated by iodine titration method and pectin by 2, 6 dichlorophenol dye titration method as described by Ranganna (1986). The Reducing sugars were quantified by Folin and Wu's method (AOAC, 1975).

The data was subjected to statistical analysis. Two-way ANOVA was carried out to test the null hypothesis that different treatments, in terms of varying concentration of *Jatropha* de-oiled cake in soil and varying days of transplanting, have no significant impact on the yields and nutritional values of fruit. Similarly, one way ANOVA tests was carried out to verify whether the difference in mean values of yield and nutritional parameters under various treatments are statistically significant. 5 % significance levels were used for statistical analyses.

3. Results and Discussion

3.1. Growth parameters

The data on nutrient content of composted de-oiled cake is summarized in Table 1. Result presented in the table 1 showed that, the compost has balanced composition in terms of NPK value. It also contains other micronutrients like Ca, Fe, Zn and Cu. The data on morphological parameters like height of plant, leaf per plant and number of flowers and tomato fruits after application of composted *Jatropha* cake are presented in Table 2. The results revealed that in for treatment D1 (35th day transplant), plant height was increased up to 31 cm for T1 (1% cake), 36.5 cm for T2 (2% cake) and 37.5 cm for T3 (3% cake) as compared to 20 cm for control. In case of D2 (40th day transplant), the increase in height was not that encouraging and it was only 34 cm even at the highest concentration (T3) as compared to 26 cm of control. However, for D3 (60th day transplant), the height of plant was substantially higher under treated soil as compared to control. The best results were achieved for T2 (2% cake) and T3 (3% cake) where the growth was 38 cm and 36 cm respectively compared to 27 cm for control. Similar trend was observed in case of number of leaves, flowers and fruits as well as the yield. A significant increase was observed in almost all the parameters as compared to control.

The yield recorded along with the results of ANOVA tests are displayed in Table 3. The results show that the F values obtained for treatment factor percentage of organic fertilizer and treatment factor days of transplant were higher than the F tabulated value corresponding to 5 % significance, thus rejecting the hypothesis that the treatments do not result in change in values of average yield. For D1 (35th day transplant), highest yield of tomato plant was obtained for T2 (2% cake) whereas it displayed a declining trend at higher concentration of organic fertilizer (3% cake). Almost similar trend was observed for other treatments under transplants D2 (40th day transplant) and D3 (60th day transplant), wherein there was an increasing trend from 1 % (i.e. T1) concentration to 2 % (i.e. T2), but a declining trend at higher concentrations (i.e. T3). This may be the confirmation of toxic effect of the cake that manifests prominently in higher concentrations and affects the yield of the plants. The plant growth parameters were not much affected with higher dose of the cake, which might be due to the specific mode of actions of the toxins present in the cake. Our results are supported by work of Tasosa *et al*. (2001), recorded a significant difference by using increased application rates of *Jatropha* and castor cakes on the growth rate of tomato and total above ground matter.

Table 1: Composition of Composted *Jatropha* cake.

Constituent	Amount
N (%)	2.95
P (%)	0.83
K (%)	1.0
Ca (%)	0.65
Fe (μg g^{-1})	785
Zn (μg g^{-1})	47.5
Cu (μg g^{-1})	26.4

Table 2. Effect of composted *Jatropha* seed cake application on morphological parameters of tomato crop.

Treatments	Nutritional parameters	Plant Height (cm)	No of leaves per plant	No of flowers	No of Fruits
35th day transplant (D1)	**Control**	20 (16-24)	6.5 (5-8)	6 (3-9)	3 (2-4)
	T1, 1 % organic fertilizer	31 (12-50)	10 (6-14)	10 (5-15)	6 (2-10)
	T2, 2 % organic fertilizer	36.5 (21-50)	11.5 (8-15)	12.5 (10-15)	8.5 (6-11)
	T3, 3 % organic fertilizer	37.5 (19-56)	12.5 (8-17)	21.5 (5-38)	9 (6-14)
40th day transplant (D2)	**Control**	26 (20-30)	7.5 (4-13)	4.5 (1-7)	4 (2-5)
	T1, 1 % organic fertilizer	28 (23-34)	11.5 (10-13)	8 (2-14)	4.5 (2-7)
	T2, 2 % organic fertilizer	32 (29-38)	9 (8-10)	8 (2-11)	8 (6-10)
	T3, 3 % organic fertilizer	34 (26-40)	12 (8-15)	3 (1-5)	6 (2-12)
60th day transplant (D3)	**Control**	27 (26-31)	9.5 (4-13)	5 (2-12)	4 (2-9)
	T1, 1 % organic fertilizer	28 (15-50)	11 (9-14)	7.5 (2-15)	7 (2-10)
	T2, 2 % organic fertilizer	36 (27-50)	12 (9-15)	8 (0-14)	8 (6-11)
	T3, 3 % organic fertilizer	38 (19-54)	11 (6-15)	19 (5-38)	7 (2-14)

% denotes proportion by weight, of composted *Jatropha* de-oiled cake in soil. Note: Range is given in parenthesis.

The results revealed that for D1 (35th day transplant), compared to the yield in control soil (91 grams), the yield per plant was highest in T2 (2% cake) appeared 163 grams followed by 126 grams with T3 (3% cake). The yield obtained for T1 (1% cake) was similar to that for control. The yield corresponding to T2 (2% cake) was significantly higher (79%) as compared to control. For D2 (40th day transplant), the highest yield per plant was recorded 145 grams for T2 (2% cake) as compared to control (123 grams), but the difference was not significant. For D3, the highest yield 181 grams was witnessed in T2 (2% cake) followed by 173 grams and 156 grams at T3 (3% cake) & T1 (1% cake) respectively. All were higher than yield corresponding to control. The yield corresponding to T2 (2% cake) and T3 *Jatropha* cake was significantly higher than control and the increase was 46 % and 40% respectively. Earlier researches have also revealed the beneficial effects of nutrients like nitrogen and phosphorus on plant growth and yield of moong and bean (Agarwal *et al*., 2006). Wani and Sreedevi (2005) observed that use of *Pongamia* cake improved the maize grain yield by 87% over the farmers' practice. Chaturvedi *et al*. (2008b) also reported similar improvement in yield of Chrysanthemum flower using *Jatropha* cake. In a study conducted by Page (1966), reported that vegetable plants receiving organic manure were always larger than those receiving inorganic fertilizer. Hence it can be concluded that the best results were obtained by using T2 (2% cake) and T3 (3% cake) treatment and D3 (60^{th} day transplant) transplant.

Table 3. Yield of Tomato fruit under various treatments: along with ANOVA test results Yield (grams per plant).

	Transplant	Control	1%(T1)	2%(T2)	3%(T3)
Transplants	35 days (D1)	91	79.17	163.33*	126.66
	40 Days (D2)	123.75	108.33	145	128.33
	60 Days (D3)	124	156.67	181.67*	173.33*
ANOVA Test Results F Values	Treatment factor: % of organic fertilizer	12.26			
	Treatment factor: Plant age when applied	17.23			
	Interaction	2.56			

F tabulated values for treatments % Organic Fertilizer (d.o.f 3) = 3.01, age of transplant (d.o.f 3) = 3.40 and interaction between the two treatments (d.o.f 6) =2.51. * Indicates that the average value of the nutrient is significantly higher than the control. Significance level is 5 %. [T1: 1% organic fertilizer, T2: 2% organic fertilizer, T3: 3% organic fertilizer].

3.2 Nutrition parameters

The nutrition values observed under various treatments and the results of ANOVA tests are displayed in Table 4. The results of two way ANOVA show that for nutrients like lycopene, pectin, protein, vitamin C, reducing sugar and TSS, the F calculated values are higher than corresponding F tabulated value corresponding to 5 % significance, which rejects the hypothesis that the varying treatments in terms of *Jatropha* cake as well as treatment related to age of transplant do not result in change in nutrient content.

Table 4: Nutritional Values recorded for Tomato fruit under various treatments along with ANOVA test results.

Treatments	Nutritional parameters	Lycopene	Pectin	Protein	Vit. C	Reducing Sugar	TSS
		mg 100 grams^{-1}	mg 100 grams^{-1}	mg 100 grams^{-1}	mg 100 grams^{-1}	%	(^{0}B)
35th day transplant (D1)	**Control**	3.43	252.67	557.01	29.37	1.27	5.2
	T1, 1 % organic fertilizer	5.82*	140.00**	654.26*	36.23*	0.91**	5.77*
	T2, 2 % organic fertilizer	3.48	385.33*	681.3*	28.98	1.11**	5.13**
	T3, 3 % organic fertilizer	1.50**	197.33**	769.48*	43.48*	2.06*	5.40*
40th day transplant (D2)	**Control**	3.45	263	557.04	29.4	1.27	5.18
	T1, 1 % organic fertilizer	4.1*	330.33	664.89*	30.4	1.14**	5.57*
	T2, 2 % organic fertilizer	3.09**	298.66	749.43*	26.1	1.04**	5.6*
	T3, 3 % organic fertilizer	2.24**	255.33	652.38*	21.70**	0.90**	5.9*
60th day transplant (D3)	**Control**	3.43	260	544.7	29.6	1.27	5.2
	T1, 1 % organic fertilizer	3.47	241	757.08*	36.2*	1	5.37
	T2, 2 % organic fertilizer	2.95	229	912.51*	29	1.73	5.6*
	T3, 3 % organic fertilizer	3.48	224.3	679.24*	34.8*	2.57*	5.8*
ANOVA Test Results F Values	**Treatment: % of organic fertilizer**	3320.35	7.43	82.75	28.88	17.36	48.32
	Treatment: age of transplant	3215.92	6.04	15.07	47.45	14.11	11.64
	Interaction	1243.89	14.04	14.65	20.06	9.06	23.26

*Indicates that the average value of the nutrient is significantly higher than the control. **Indicates that the average value of the nutrient is significantly lower than control. Significance level is 5 % F tabulated values for treatments % Organic Fertilizer (d.o.f 3) = 3.01, age of transplant (d.o.f 3) = 3.40 and interaction between the two treatments (d.o.f 6) =2.51.

The analysis of average nutrient values recorded under various treatments reveals that for D1 (35th day transplant), nutrients like vitamin C, reducing sugar and proteins were found to be highest in fruits produced on T3 (3% cake) and they were significantly higher than corresponding values for control. However, under this treatment, values for pectin, lycopene were significantly lower than that for control.

In case of T2 (2% cake), significantly high values were achieved for protein and pectin as compared to control and the differences was insignificant for vitamin C and lycopene. For D2 (40th day transplant), TSS and Protein values were significantly higher for fruits produced in soil with T2 (2% cake) as compared to corresponding control values. However, there was insignificant change recorded for lycopene, pectin and vitamin C. As in case of yield the results for T1 (1% cake) and T3 were not encouraging as far as nutritional parameters were concerned. For D3 (60th day transplant), TSS, vitamin C, protein and reducing sugar values for fruits produced in T3 (3% cake) were significantly higher than their corresponding control values. However, for key nutrients like lycopene and pectin, the values were similar to the corresponding control values and the difference was not significant.

Similarly, for fruits produced in T2 (2% cake), average values for TSS and proteins were significantly higher than corresponding control values. However, for other key nutrients like lycopene, pectin, vitamin C and reducing sugar, the variations as compared to control were insignificant. The improved yield with organic manure over inorganic fertilizer might have been due to improved physical characteristics of the soil (Mbagwu and Ekwealor, 1990) as well as presence of macro and trace elements.

Chaturvedi *et al*. (2009) also found increase in nutritional quality of tomato fruit using organic residue like tobacco waste. Sahrawat and Mukerjee (1976) have demonstrated enhancement in yield of rice crop by 31- 54%, total nitrogen uptake by 36-68% and the grain protein by 2-14% due to the nitrification inhibiting properties of karanjin, the major crystalline principle of karanja seed. Similarly, Wang and Liu (2002) recorded significant increase of tomato crop sugar level and fruit quality by using vermi-compost.

Odedina *et al*. (2007) found that animal dropping and agro-waste enhanced nutrient status of tomato. Thus, we can conclude that for 60 days transplant (D3), significantly high yields obtained on soils mixed with 2 % *Jatropha* cake (T2). However, only TSS and protein content was found to be significantly higher than control, while for other nutrient contents, the difference was insignificant as compared to control. For fruits produced on 60 days transplant (D3) and soil mixed with 3 % cake (T3), substantially high values for key nutrients like TSS, vitamin C, proteins and reducing sugar are being obtained and the yield obtained is significantly higher than control. The possible reason for this is the incorporation of organic materials into soil promotes microbial activity and, in turn improvement of soil fertility as it promotes mineralization of the important organic elements. But main challenge is to optimize the crop needs before adding the organic waste, because it sometimes leads to imbalance of nutrients (Sinha *et al*., 2011). Therefore in order to gain optimum crop yield, it becomes imperative to optimize an organic residue according to the crop requirements.

4. Conclusions

We conclude that, by adding appropriate proportions of composted de-oiled *Jatropha* cake in soil has significant positive influence on yield. Substantially high yields were obtained in soils mixed with 2 to 3% *Jatropha* cake and 60 days of transplant. For 60 days transplant and 2 % composted cake, the yield was highest and nutrient values were unaffected. However, in case of 3 % cake, yield as well as key nutrient levels were enhanced significantly as compared to control. This study proposed that composted de-oiled *Jatropha* cake has good potential of being utilized in a cost effective way as tailored, crop specific organic fertilizer, as replacement of chemical fertilizers.

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