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# Intake, blood metabolites and hormonal profile in sheep fed processed Jatropha (*Jatropha curcas*) meal

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ABSTRACT

The aim of the study was to evaluate the effect of feeding processed Jatropha meal (JM) incorporated diets on intake, digestibility, blood metabolites and hormonal profile in sheep. The fifteen adult male non-descript sheep were randomly divided into three equal groups. Throughout the 90-d experimental period, all the animals were fed oat (*Avena sativa*) straw *ad libitum* and one of three concentrate mixtures: maize 320 g/kg, wheat bran 340 g/kg, rice bran 110 g/kg, soybean meal 200 g/kg, mineral mixture 20 g/kg, and salt 10 g/kg ($T_1$) and sodium chloride (NaCl; 10 g/kg JM; $T_2$) or calcium hydroxide [$Ca(OH)_2$] processed JM (5 g/kg JM; $T_3$) substituting 120 g/kg of crude protein (CP) in $T_1$ ($T_2$ and $T_3$). The metabolism trial was conducted during the last 7 d of the experiment. Blood samples were collected at day 0, 30, 60 and 90 of the experiment to study the blood metabolites. The phorbol ester and haemagglutination (HA) activity were reduced considerably in processed JM. The dry matter, organic matter and CP intake were lower ($P<0.05$) in sheep fed $T_2$ and $T_3$ compared to $T_1$. The packed cell volume, serum albumin, glucose, serum urea, triiodothyronine, thyroxine and testosterone contents decreased ($P<0.05$) in sheep fed $T_2$ and $T_3$ compared to $T_1$. Alkaline phosphatase activity was lower ($P<0.01$) in $T_2$ and $T_3$ compared to $T_1$, whereas lactate dehydrogenase and aspartate amino transferase activities increased ($P<0.01$) in sheep fed $T_2$ and $T_3$ compared to $T_1$. From the observations, it is concluded that, even though phorbol ester content and HA activity was markedly decreased by processing JM with NaCl and $Ca(OH)_2$, this was not sufficient to reach a safe level for feeding sheep. A reduced nutrient intake and unusual blood metabolite levels was evident in the processed JM-fed animals.
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## 1. Introduction

A huge shortage of feeds in developing countries gives every scope to search for alternate feed resources for reducing feeding costs. Jatropha (*Jatropha curcas*) is most widely planted medicinal plant (Openshaw, 2000) and there has been an increasing awareness of growing this species on barren land. In addition to being a source of bio-diesel, Jatropha meal (JM) can also be a source of economic protein supplement in animal feed (Makkar and Becker, 1999). Due to the importance of biodiesel production from the Jatropha seeds, its cultivation is being popularized in many countries.

*Abbreviations:* ALP, alkaline phosphatase; AST, aspartate amino transferase; $Ca(OH)_2$, calcium hydroxide; CP, crude protein; DM, dry matter; HA, haemagglutination; JM, jatropha meal; LDH, lactate dehydrogenase; NaCl, sodium chloride; NaOH, sodium hydroxide; OM, organic matter; PCV, packed cell volume.

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However, Jatropha plant is toxic (Ahmed and Adam, 1979) to livestock, which is widely used as a fence to protect the food crops (Heller, 1996) from the animals. Nevertheless, despite all the advantages, protein rich JM reported to contain anti-nutritional factors such as phorbol ester, curcin, trypsin inhibitor (Makkar and Becker, 1999; Adolf et al., 1984; Rakshit et al., 2008). The above observations have thus limited its use as a food or feed source.

The trypsin inhibitor and curcin present in JM can be reduced by heat treatment (Aderibigbe et al., 1997), whereas it will not be possible to destroy phorbol ester by heat treatment because they are heat stable. However, it is possible to reduce its concentration in JM by chemical treatments (Makkar and Becker, 1999). The curcin is found to have proteolytic and haemagglutination (HA) activity on red blood cells (Bolley and Holmes, 1958), for that reason, HA test can be performed for screening of curcin content in variously processed JM.

More information regarding toxic factor of JM is available. However, little information is available on effect of feeding variously processed JM to livestock. Feeding of JM affects the blood biochemicals (Awasthy et al., 2010), and concentrations of triiodothyronine, thyroxine (Todini et al., 2007) and testosterone are a good indicators of normal body metabolism. Hence, the present study was aimed to evaluate the effect of feeding processed JM incorporated diets on intake, digestibility, blood metabolites and hormonal profile in sheep.

## 2. Materials and methods

### 2.1. Experimental animals

Fifteen non-descript adult sheep were randomly divided into three equal groups. All the animals were housed in a well-ventilated shed having provision for individual feeding. Before the start of the experiment, the animals were dewormed with levamisol hydrochloride (Lemasol-75; Ranbaxy Laboratories, New Delhi, India) 7.5 mg/kg body weight subcutaneous to control internal parasites. Butox (Intervet, India Ltd., Pune) 2 ml/l water was sprayed all over the body to control external parasites.

### 2.2. Chemical and physical processing of JM

The residue left after solvent extraction of Jatropha seeds is referred as JM, which was procured from Ayurvet Ltd, New Delhi, India and subjected for various chemical processing (Anandan et al., 2005) to reduce its toxin content. JM was processed chemically with 5, 10, 20, 25 g/kg dry matter (DM) sodium chloride (NaCl), 2.5, 5, 10, 20, 25 g/kg DM sodium hydroxide (NaOH) or calcium hydroxide [$Ca(OH)_2$] and 10, 20, 30 g/kg DM urea. JM was also subjected for soaking (over night) and roasting (100 °C for 30 min).

### 2.3. Estimation of phorbol ester

Phorbol-12-myristate-13-acetate (Calbiochem Ltd., KGaA, Darmstadt, Germany) standard was used for estimation of phorbol ester in JM using the High Performance Liquid Chromatography (Shimadzu 10A; Kyoto, Japan) method (Makkar et al., 1997). The phorbol ester concentration in raw and processed JM was calculated (g/kg DM) by comparing peak area of each standard and sample as follows:

$$\text{Phorbol ester (mg/kg)} = \frac{\text{Area of sample} \times \text{concentration of standard} \times \text{final volume}}{\text{Area of standard} \times \text{weight of sample}}$$

### 2.4. HA test

HA test was performed to screen the toxins present in JM using the RBCs of chicken, rabbit, guinea pig, sheep and goat. HA activity of raw and variously processed JM extract was expressed as reciprocal of end point dilution and matrix formation (Aregheore et al., 1998).

### 2.5. Feeds and feeding

Based on the results of HA test and phorbol ester content, processing with 10 g/kg DM NaCl and 5 g/kg DM $Ca(OH)_2$ were selected for further feeding trial with sheep. Three different concentrate mixtures were computed for the present experiment. Throughout the 90-d experimental period, all the animals were fed oat (*Avena sativa*) straw *ad libitum* and one of three concentrate mixtures: maize 320 g/kg, wheat bran 340 g/kg, rice bran 110 g/kg, soybean meal 200 g/kg, mineral mixture 20 g/kg, and salt 10 g/kg ($T_1$) and NaCl (10 g/kg JM; $T_2$) or $Ca(OH)_2$ processed JM (5 g/kg; $T_3$) substituting 120 g/kg of crude protein (CP) in $T_1$ ($T_2$ and $T_3$). Vitablend (AD3) containing vitamin A – 50,000 IU and D3 – 5000 IU/g was added at 6 g/kg concentrate mixtures. The concentrate mixtures were made isonitrogenous and offered to the respective groups at 10:00 h to meet the CP requirement as per the NRC (1985). The rest of the nutrient requirements were met by *ad libitum* feeding of oats (*A. sativa*) straw. Green fodder was provided once in a week to meet vitamin A requirement. Leftover feed

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