

ISSN 1819-1878

Asian Journal of  
**Animal**  
Sciences

## Effect of Feeding Castor Seed Cake Based Diets on Growth, Nutrient Utilization, Immune Response and Carcass Traits in Lambs

D. Nagalakshmi and K. Dhanalakshmi

College of Veterinary Science, Sri Venkateswara Veterinary University, Hyderabad, India

*Corresponding Author: D. Nagalakshmi, College of Veterinary Science, Sri Venkateswara Veterinary University, Hyderabad, India*

### ABSTRACT

An experiment was conducted to study the effect of utilizing Castor (*Ricinus communis*) Seed Cake (CSC) (30-45% Crude Protein; CP) as sole protein supplement, replacing groundnut cake (GNC) on growth performance, nutrient utilization, immune response and carcass traits in lambs. Two isonitrogenous and isocaloric complete diets (10.5% CP and 58% TDN) were formulated with 13% GNC and 10% CSC and fed *ad libitum* to 14 *Nellore* male lambs (27.54±1.154 kg), divided at random into two groups of seven animals for a period of 150 days. The lambs in both groups grew linearly with average daily gain of 75.33 and 74.28 g, respectively with no significant difference. Inclusion of CSC did not affect the Dry Matter (DM) and CP intake and was comparable to that of control lambs but the intake of metabolizable energy was lower ( $p < 0.01$ ) in CSC fed lambs. The lambs fed either GNC or CSC utilized DM and nutrients with similar efficiency. Inclusion of CSC in complete diets of lambs did not affect the nutrients digestibility, balances of nitrogen, calcium and phosphorus, hematological (haemoglobin, total erythrocyte and leucocyte counts) and biochemical constituents (total protein, albumin, globulin and creatinine) except for higher ( $p < 0.01$ ) digestibility of crude fibre in CSC fed lambs compared to GNC feeding. While, the antibody titers against heat killed *Brucella abortus* and chicken RBC antigens assessed at 75 day of feeding and the skin indurations against PHA-P mitogen, assayed at 150 day of feeding was lower ( $p < 0.01$ ) with CSC feeding. The carcass characteristics (dressing percentage, proportion of meat, bone and fat, proportion of edible and non-edibles, whole sale cuts, organ weights and chemical composition of *Longissimus dorsi* muscle) were not influenced by inclusion of CSC as sole protein supplement. Histopathological lesions observed were mild to moderate areas of necrosis, congested blood vessels with increased kuffer cell activity in liver, swollen kidney tubules with increase in mononuclear cell infiltration, decreased goblet cell activity and infiltration of neutrophils in intestines. The study suggested no adverse effect on the nutritional performance and carcass traits of lambs with inclusion of 10% CSC in complete diets but the immune response was depressed and lesions of pathological significance was observed in vital organs compared to GNC fed lambs.

**Key words:** Carcass traits, castor seed cake, growth, histopathology, immune response, lambs, nutrient utilization

### INTRODUCTION

The chronic shortage of conventional oil cakes for livestock feeding in most of the developing countries has compelled for the search of alternatives for these costly protein supplements. Groundnut cake is the conventionally used protein supplement in ration of ruminants in most parts

of the country but at times its limited supply and seasonal availability escalates its cost. India ranks first in castor seed (*Ricinus communis*) production with an annual availability of 1.644 million t (FAO., 2014). The residue obtained after extraction of oil from castor seeds i.e., Castor Seed Cake (CSC) is available to the tune of 1.12 million t and has potential to be used as protein supplement in animal diets because of its high crude protein and energy comparable to the conventional ones but limited because of potent antinutritional factors such as ricin, ricinine, allergen and chlorogenic acid (Albretsen *et al.*, 2000; Audi *et al.*, 2005). Many detoxification methods employing various physical (dehulling, soaking, heating, steaming, boiling, autoclaving and extrusion) (Anandan *et al.*, 2005; Ani and Okorie, 2009), chemical (ammonia, formaldehyde, lime, sodium chloride, tannic acid, sodium hydroxide and calcium compounds), (Anandan *et al.*, 2005; Oliveira *et al.*, 2011) and fermentation (Oso *et al.*, 2011; Ulanova and Kravchenko, 2013) methods were tried but with limitations and the results were variable. Moreover, the feasibility of adopting various detoxification methods and the additional cost of detoxification limits the use of unconventional oilseed meals in animal feeds compared to conventionally used protein supplements. Therefore, these unconventional oil seed meals could be used as such in limited amounts considering its nutrient profile and anti-nutritional factors present in them. Gowda *et al.* (2009) and Oliveira *et al.* (2015a) reported no adverse effect on the nutritional performance of adult sheep and male goats due to feeding of raw or lime treated castor seed cake replacing soyabean meal in the rations. Thus, a detailed study was undertaken to explore the possibility of utilizing CSC as sole source of protein in complete diets of lambs reared under intensive system.

## **MATERIALS AND METHODS**

**Animals, housing management, feeds and feeding:** Fourteen Nellore lambs (27.54+1.154 kg) were dewormed before study and randomly allotted to 2 dietary treatments varying in protein sources. All the lambs were housed in cement floored well-ventilated shed with provision of feeding, watering and open paddock for roaming for 2 h in early morning to have sufficient exercise. All lambs were dewormed at regular intervals alternatively with albendazole and fenbendazole throughout the experimental period. Two isonitrogenous and isocaloric sorghum stover based complete diets (inclusive of roughage and concentrate ingredients) in mash form with 10.5% protein were formulated with 13% groundnut cake (GNC) or 10% CSC as sole protein supplements. The lambs were fed the respective diets *ad libitum* to meet the nutrient requirements of ICAR (1998) for maintenance and daily weight gain of 100 g for experimental duration of 150 days. Refusals, if any were measured next day before the offering of feed. The quantity of complete diets to be offered daily was adjusted fortnightly as per body weights recorded at the end of every fortnight. The body weight of individual lambs was recorded at the onset of the experiment and thereafter at fortnightly intervals in the morning before feeding and watering for two consecutive days to assess the changes in body weight and growth rate. Efficiency of nutrient utilization was calculated as unit intake per unit gain.

**Nutrient utilization and balances:** A metabolism trial of 6 days duration following 120 days of experimental feeding was conducted on all lambs in metabolic cages to determine the nutrients digestibility, plane of nutrition and nutrients balance (nitrogen, calcium and phosphorous) in lambs. The metabolism cages had provision for individual feeding, watering and collection of feces and urine separately. Daily feed offered, residues left, feces and urine voided were recorded on a 24 h basis. Appropriate aliquots of feed, refusals and feces were dried at 105°C for 24 h to

determine Dry Matter (DM). The pooled dried samples of feces, feed and feed residues were ground with 1 mm sieve and analyzed for proximate constituents and energy to arrive at nutrients digestibility. Loss of nutrients in urine was taken for determining the balances. Suitable aliquots of urine was fixed with H<sub>2</sub>SO<sub>4</sub> for N estimation and preserved in plastic vials for Ca and P estimation.

**Hematological and biochemical constituents:** Blood was collected from each lamb before feeding at 75th day of experiment and transferred 5 mL into bottle containing ethylene diamine tetra acetate for hematological parameters, then centrifuged at 1,100x g for 10 min to obtain serum and stored at -20°C for biochemical estimations. The hemoglobin (Hb), Total Erythrocytes Count (TEC) and Total Leukocytes Count (TLC) were estimated by cyanomethemoglobin method using Hayems fluid (Coles, 1986) and white blood count diluting fluid (Schalm, 1986), respectively. Serum samples were analyzed for total protein (Reinhold, 1953), albumin (Gustafsson, 1976) and creatinine (Bonsnes and Tauskey, 1945).

**Humoral immune response:** The humoral immune response was assessed after 75 days of experimental feeding using heat-killed *Brucella abortus* S<sub>99</sub> and chicken RBC as immunogens. All lambs were sensitized with heat-killed *B. abortus* S<sub>99</sub> and 20% chicken RBC suspension, administered intramuscularly as per the BW and a booster dose of antigen was given after 15 days. Serum was collected from lambs on 7, 14, 21, 28 and 35 days Post Sensitization (PS) to estimate the antibodies against *B. abortus* by Standards Tube Agglutination Test (STAT) and chicken RBC by direct hemagglutination assay (Wegmann and Smithies, 1966).

**Cell mediated immune response:** After 150 days of feeding, the Cell Mediated Immune response (CMI) was estimated by Delayed Type Hypersensitivity (DTH) against phytohemagglutinin-P, measured as increase in skin thickness in all animals at 0, 24, 48, 72 and 96 h post injection (Quist *et al.*, 1997).

**Carcass studies:** Four representative lambs from each group were slaughtered on 155th day of the experiment for carcass characteristics. The lambs were fasted overnight with free access to water, slaughtered by 'Halal' method after noting the overnight fasted weight. After skinning and evisceration, weight of hot carcass (CW), weight of primal cuts, edible (carcass, liver, heart, kidneys, testes, dressed head and feet) and inedible (blood, empty gastrointestinal tract, skin, spleen and lungs with trachea) weights and organ weights were recorded. Separated bone, meat and fat of carcass of each animal were weighed and expressed as percentage of CW. The *Longissimus dorsi* muscle from one side of the carcass was preserved by deep freezing (-18°C) for analysis of the chemical composition of muscle.

**Histopathology:** The lambs during slaughter were investigated for gross abnormalities of all the organs, if any. The representative tissue pieces of heart, lungs, liver, kidney, testes, spleen and intestines were collected in 10% neutral buffered formal saline for histopathological studies. The fixed tissues were subjected to overnight washing, dehydrated in ascending grades of alcohol, cleared in xylene, embedded in paraffin and further the paraffin blocks were cut into 4-5 µ thickness sections with the help of microtome. Sections were lifted on precoated slides and subjected to routine haematoxylin and eosin stain after drying (Culling, 1963) and were examined for histological changes.

**Analytical procedure:** The feed and fecal samples collected during metabolism trial were analyzed for proximate constituents, P (AOAC., 2000) and Ca (Talapatra *et al.*, 1940). The Gross Energy (GE) in feed, feces and urine samples were estimated by bomb calorimeter to arrive at Digestible Energy (DE) and Metabolizable Energy (ME). Methane production was calculated by an equation suggested by Blaxter and Clapperton (1965) based on digestibility coefficient of energy. Minced meat samples were analyzed for moisture, fat, protein and ash (AOAC., 2000). The ricin content in the CSC was estimated according to the method of Kabat *et al.* (1947) and with modifications suggested by Waller and Negi (1958).

**Statistical analysis:** The data, except for immune studies was subjected to statistical analysis in one-way classification under a completely randomized design using General Linear Model of SPSS 12.0. Two way ANOVA was carried out for immune parameters with diet and days post sensitization as two factors. Comparison between means was done using Duncan multiple range test (Duncan, 1955) at 5 and 1% level.

## RESULTS AND DISCUSSION

The CSC used in the present study contained 44.11% Crude Protein (CP) and 25.66% Crude Fibre (CF), higher than the conventionally used protein supplement GNC. The CP of CSC used in the experiment was higher than the values reported by Gowda *et al.* (2009) (39.5%), Akande and Odunsi (2012) (39.58%) and Ani and Okorie (2009) (40.9%). Similarly the CF content was higher than the reported values of previous workers (2.5-12.5%) and lower than that reported by Okoye *et al.* (1987) (37.7%), which could be due to variation in the degree of decortication. The CSC and GNC were ghani pressed and thus had higher crude fat. The ricin content in CSC was 476 ppm and was higher than the values reported by Anandan *et al.* (2005) (388 ppm) and Gowda *et al.* (2009) (314 ppm) and lower than that reported by Diniz *et al.* (2010) (1143.7 ppm) (Table 1).

The ricin and allergen in CSC suppresses the appetite of the animals. Feed intake decreased ( $p < 0.01$ ) in layers with inclusion of even 3.5% of raw CSC and further with increase in proportion

Table 1: Ingredients and chemical composition of experimental diets

Ingredients (%)	Diet			
	GNC based	CSC based	GNC	CSC
Sorghum stover	40.000	40.000		
Maize	30.500	33.000		
Groundnut cake	13.000	--		
Castor seed cake	--	10.000		
Deoiled rice bran	9.500	10.000		
Molasses	5.000	5.000		
Di-calcium phosphate	0.839	1.032		
Calcite powder	0.645	0.383		
Salt	0.500	0.500		
*Trace mineral and vitamin mixture	0.055	0.055		
<b>Chemical composition** (%)</b>				
Crude protein	10.810	10.440	36.17	44.11
Ether extract	1.770	1.020	10.42	8.98
Crude fibre	24.650	26.010	22.06	25.66
Total ash	11.730	9.300	6.25	6.05
Calcium	1.580	1.270	0.36	0.69
Phosphorous	0.420	0.420	0.25	0.51

GNC: Groundnut cake, CSC: Castor seed cake, \*Trace mineral and vitamin premix provided ( $\text{mg kg}^{-1}$ ), iron: 30, copper: 10, manganese: 40, zinc: 30, cobalt: 0.2, molybdenum: 0.5, iodine: 0.25, selenium: 0.2, vitamin A and E were provided to supply 940 and 20I U  $\text{kg}^{-1}$  diet, respectively. \*\*Estimated

of CSC up to 14% (Olayeni *et al.*, 2006). Feed intake, body weight and feed efficiency depressed in broilers with dietary inclusion of 10-25% raw (Okoye *et al.*, 1987) or 5-15% fermented (Oso *et al.*, 2011) CSC. In rabbits too feeding of 5-25% boiled CSC based diets decreased the daily feed intake and average daily gain (Adedeji *et al.*, 2006). In the present study, lambs fed GNC and CSC diets grew with an average daily gain of 75.33 and 74.28 g, respectively with no significant difference. No adverse effect of feeding CSC as sole protein supplement in diet was observed on body weights, DM intake and nutrient efficiency (DM, CP and ME) except for the higher TDN intake (Table 2). Similarly, no effect on DMI, was reported in sheep fed 30% CSC containing concentrate mixture (Garg *et al.*, 2005) or 13.5% CSC in total mixed ration (Gowda *et al.*, 2009), kids fed 15% CSC (Oliveira *et al.*, 2015b), buffaloes fed 30% CSC containing concentrate mixture (Reddy *et al.*, 1986). In accordance, Diniz *et al.* (2010) reported no effect on body weight gain, DMI and feed efficiency in beef cattle fed 9.14% raw CSC based diets.

The digestibility of nutrients, except for CF digestibility did not differ with inclusion of CSC in place of GNC. All lambs were on positive nitrogen, calcium and phosphorous balance and the balances were comparable between CSC and GNC fed lambs (Table 3). De Oliveira *et al.* (2010) reported no effect on nutrient digestibility except ether extract when fed 15% CSC replacing SBM in sheep consuming 2.46 g ricin kg<sup>-1</sup> b.wt. Similarly, Gowda *et al.* (2009) and Garg *et al.* (2005) did not observe any significant differences in digestibility of nutrients in adult sheep with incorporation of 12.3% CSC in total mixed ration or 30% CSC in concentrate mixture of ration. On similar lines, Purushotham *et al.* (1994) observed no significant difference in the digestibility of nutrients in sheep fed concentrate mixture containing up to 30% steam treated CSC. Reddy *et al.* (1986) reported no adverse effect on digestibility of DM, organic matter, CF and nitrogen free extract in buffaloes fed 30% CSC in the concentrate mixture. While, Kumar (1998) reported a significant depression in digestibility of DM, OM and NFE in lambs fed concentrate mixture containing 36-37.5% CSC, which might be due to higher level of inclusion of CSC. The nutrient digestibility in lactating cows and TDN content of the diet reduced with replacement of SBM with calcium oxide (CaO) treated CSC beyond 33% with no effect on nitrogen balance at even 100% replacement in studies of Cobianchi *et al.* (2012). De Barros *et al.* (2011) also observed quadratic decrease in nitrogen balance in heifers with replacement of SBM with CaO treated CSC beyond 33% (16.5% CSC in supplement mixture) i.e., at 67 and 100% replacement. A linear reduction in DM and CP retentions and increase in CF retention was observed in cockerels with inclusion of fermented

Table 2: Performance of lambs fed castor seed cake based diets

Attributes	GNC	CSC	SEM	p-value
<b>Growth performance</b>				
Initial body weight (kg)	27.57	27.51	1.154	0.981
Final body weight (kg)	38.87	38.66	1.099	0.927
Average daily gain (g)	75.33	74.28	4.423	0.911
<b>Nutrient intake (g or kcal kg<sup>-1</sup> W<sup>0.75</sup>)</b>				
Dry matter	97.49	102.02	2.351	0.356
Crude protein	10.53	10.61	0.248	0.879
Total digestible nutrients	52.20 <sup>b</sup>	60.12 <sup>a</sup>	1.690	0.012
Metabolizable energy (kcal)	200.80 <sup>a</sup>	184.19 <sup>b</sup>	5.999	0.018
<b>Nutrient efficiency (g or kcal intake/g gain)</b>				
Dry matter	18.08	20.84	1.101	0.402
Crude protein	1.95	2.06	0.105	0.610
Total digestible nutrients	9.68	11.70	0.629	0.111
Metabolizable energy(kcal)	37.26	34.06	1.873	0.415

<sup>ab</sup>Means with different superscripts in a row differ significantly: p<0.05, p<0.01, GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean

Table 3: Nutrient digestibility and balances in lambs fed castor seed cake based diets

Nutrients	GNC	CSC	SEM	p-value
<b>Nutrient digestibility (%)</b>				
Dry matter	56.24	59.59	1.085	0.128
Organic matter	58.96	63.24	1.231	0.079
Crude protein	58.46	61.53	1.239	0.236
Ether extract	59.57	53.44	3.424	0.402
Crude fibre	40.90 <sup>b</sup>	53.10 <sup>a</sup>	2.436	0.003
Nitrogen free extract	68.67	69.54	1.139	0.725
Gross energy	63.75	67.03	1.202	0.187
<b>Balance (g)</b>				
Nitrogen	9.48	9.62	0.663	0.922
Calcium	9.51	6.47	0.979	0.125
Phosphorus	2.80	2.84	0.202	0.926
<b>Nutritive value (%)</b>				
Digestible crude protein	6.32	6.40	0.122	0.752
Total digestible nutrients	53.86 <sup>b</sup>	59.10 <sup>a</sup>	1.170	0.013
Digestible energy (Mcal kg <sup>-1</sup> )	2.51 <sup>a</sup>	2.10 <sup>b</sup>	0.079	0.001
Metabolisable energy (Mcal kg <sup>-1</sup> )	2.06 <sup>a</sup>	1.72 <sup>b</sup>	0.065	0.001
<b>Plane of nutrition (g or kcal kg<sup>-1</sup> W<sup>0.75</sup>)</b>				
Dry matter	98.23	90.08	3.209	0.223
Crude protein	11.83 <sup>a</sup>	10.21 <sup>b</sup>	0.400	0.033
Digestible crude protein	6.21	5.78	0.254	0.435
Total digestible nutrients	53.00	53.19	1.858	0.964
Digestible energy	246.90 <sup>a</sup>	188.80 <sup>b</sup>	13.009	0.014
Metabolisable energy (kcal)	202.50 <sup>a</sup>	154.80 <sup>b</sup>	10.670	0.014
Water intake (L kg <sup>-1</sup> DMI)	2.34 <sup>a</sup>	3.29 <sup>b</sup>	0.206	0.009

<sup>ab</sup>Means with different superscripts in a row differ significantly: p<0.05, p<0.01, GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean

CSC from 5-15%, replacing SBM (Oso *et al.*, 2011). Nutrient retentions also decreased with inclusion of 10-20% dehulled and cooked CSC in broiler chicks (Ani and Okorie, 2009). The DCP content of diet was comparable but CSC based diet had higher TDN content.

Purushotham *et al.* (1985b) reported increased number of lymphocytes, reduced number of neutrophils, leucocytes and higher hematocrit value in adult sheep fed daily with 250 g concentrate mixture containing 30% CSC. Similarly, a higher leucocyte and lower packed cell volume, TEC and Hb% in broilers was observed with feeding of diets containing 10-20% heat treated and fermented CSC (Akande and Odunsi, 2012). In the present study, inclusion of CSC containing 476 ppm ricin at 10% in diet had no adverse effect on hematological constituents in lambs estimated at 75 day of experimental feeding (Table 4) and the observed values were within normal limits. The results corroborated with the findings of Gowda *et al.* (2009), Adedeji *et al.* (2006) and Oso *et al.* (2011) in sheep fed 12.5% CSC, rabbits fed 15% CSC diets or broiler fed 15% fermented CSC diets, respectively.

The biochemical constituents (total protein, albumin and globulin, creatinine) were not affected by CSC inclusion in diet of lambs (Table 4). While, Aslani *et al.* (2007) reported higher blood creatinine in lambs fed raw CSC, respectively. Such depressing effect on biochemical constituents with feeding of CSC was not observed in the present study and these findings corroborated with those of Gowda *et al.* (2009) and Adedeji *et al.* (2006).

Feeding of diets containing antinutritional/toxic factors affects immunity in various livestock species and poultry. Incorporation of 40% expeller or solvent extracted karanj seed cake in diets of broilers reduced humoral immune response (Panda, 2004) due to presence of karanjin. Similarly, immunological reactivity was lowered due to feeding of cotton seed meal based diets owing to presence of gossypol (Nagalakshmi *et al.*, 2001). In the present study, antibody titres against specific antigen *B. abortus* and non-specific antigen like chicken RBC was depressed (p<0.01) in lambs fed CSC based diets compared to those fed GNC based diets (Table 5). The ricin in CSC is

Table 4: Hematological, biochemical constituents and immune response in lambs fed castor seed cake based diets

Constituents	GNC	CSC	SEM	p-value
<b>Hematological constituent</b>				
Haemoglobin (g dL <sup>-1</sup> )	13.37	13.16	0.432	0.815
RBC (x10 <sup>6</sup> μL <sup>-1</sup> )	7.34	8.44	0.639	0.413
WBC (x10 <sup>3</sup> μL <sup>-1</sup> )	9.00	9.48	0.447	0.612
Lymphocytes (%)	31.83	41.00	1.339	0.684
Neutrophils (%)	59.17	55.67	1.400	0.227
<b>Biochemical constituent</b>				
Total protein (g dL <sup>-1</sup> )	5.35	5.14	0.117	0.393
Albumin (g dL <sup>-1</sup> )	1.70	1.82	0.044	0.168
Globulin (g dL <sup>-1</sup> )	3.65	3.32	0.122	0.182
Creatinine (mg dL <sup>-1</sup> )	0.64	0.92	0.174	0.450

GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean, RBC: Red blood cells, WBC: White blood cells

Table 5: Humoral immune response in lambs fed castor seed cake based diets

Diets	Days post sensitization					Average
	7	14	21	28	35	
<b>Brucella abortus (log<sub>2</sub> titres)</b>						
GNC	8.82	8.57	7.57	7.32	7.32	7.92±0.193 <sup>a</sup>
CSC	8.72	8.52	7.12	6.52	5.92	7.36±0.241 <sup>b</sup>
Average	8.77±0.263 <sup>z</sup>	8.54±0.131 <sup>z</sup>	7.34±0.212	6.92±0.221 <sup>x</sup>	6.92±0.300 <sup>x</sup>	
<b>Chicken RBC (log<sub>2</sub> titres)</b>						
GNC	6.40	4.80	4.60	4.40	5.00	5.04±0.248 <sup>a</sup>
CSC	5.60	3.60	3.60	3.40	3.20	3.88±0.273 <sup>b</sup>
Average	6.00±0.365 <sup>z</sup>	4.20±0.416 <sup>y</sup>	4.10±0.348 <sup>y</sup>	3.90±0.347 <sup>y</sup>	4.10±0.458 <sup>y</sup>	

<sup>abxyz</sup>Means with different superscripts in a row or column differ significantly: p<0.01, GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean

highly cytotoxic and potent immunotoxin (Brandt *et al.*, 2005) resulting in immunosuppression. The peak antibody titres against *B. abortus* were on 7th day PS and later decreased. The IgG levels lowered and expressing of mRNA for HSP 70.1 decreased in goats fed either treated or detoxified CSC (Arruda *et al.*, 2013) corroborating with present findings. Naidu *et al.* (1988) observed formation of antitoxins against ricin in rams fed 30% CSC containing concentrate mixture along with mixed grass hay. Similarly, antigenic response was elicited against ricin in adult sheep fed CSC based diet (Gowda *et al.*, 2009). The skin induration against PHA-P mitogen, indicative of cell mediated immunity was depressed with CSC feeding at 24, 48 and 96 h post sensitization (Table 6). The DTH response was at peak at 24 h PS and thereafter gradually decreased at 48, 72 and 96 h PS. At low intakes of ricin, antibodies against ricin are produced but as the intake of ricin increases, it depresses the immune response and has ill effects in the body. The ricin intake was only 23.17 mg day<sup>-1</sup> in studies of Gowda *et al.* (2009) as against the 57.98 mg day<sup>-1</sup> ricin intake in the present study, which attributed to the depressing effect on immune response.

Gowda *et al.* (2009) recorded the dressing percentage of 45.8-47.6% in CSC fed sheep which was similar to the dressing percentage of 48.35% observed in the present study with feeding of CSC diet (Table 7). No effect on dressing percentage and weights of kidney, liver, heart and spleen with feeding of CSC diets corroborated with findings of Gowda *et al.* (2009). The carcass characteristics in terms of dressing percentage, proportion of meat, bone and fat, proportion of edible and non-edibles, whole sale cuts, organ weights and chemical composition of *Longissimus dorsi* muscle were not influenced by inclusion of CSC as sole protein supplement. Similarly, Uechiewcharnkit and Kanthapainit (1997) in fattening pigs and Diniz *et al.* (2010) in beef cattle observed no effect on preslaughter weight, dressing percentage, back fat thickness, proportion of various cuts and meat, fat and bone proportion when fed 8-16% detoxified CSC or



Table 6: Cell mediated immune response (increase in skin thickness, mm) against PHA-P in lambs fed castor seed cake based diets

Diets	Hours post sensitization			
	24	48	72	96
GNC based diet	2.840 <sup>a</sup>	1.960 <sup>a</sup>	1.300	0.820 <sup>a</sup>
CSC based diet	1.800 <sup>b</sup>	0.760 <sup>b</sup>	1.140	0.180 <sup>b</sup>
SEM	0.282	0.329	0.203	0.179
p value	0.049	0.042	0.721	0.030

<sup>ab</sup>Means with different superscripts in a column differ significantly: p<0.05, GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean

Table 7: Carcass characteristics of lambs fed castor seed cake based diets

Attributes	GNC	CSC	SEM	p-value
Pre slaughter weight (PSW) (kg)	33.05	34.22	0.876	0.545
Empty body weight (kg)	31.52	33.15	0.852	0.379
Carcass weight (kg)	15.35	16.54	0.480	0.244
Dressing percentage, (% PSW)	46.46	48.35	0.759	0.237
Loin eye area (cm <sup>2</sup> )	24.79	23.93	0.910	0.672
Meat (%)	61.48	61.86	1.198	0.887
Bone (%)	28.70	28.61	1.374	0.976
Fat (%)	9.86	11.60	0.599	0.158
Meat: Bone	2.14	2.26	0.155	0.730
Edible (%)	49.21	51.06	0.784	0.268
Non-edible (%)	19.51	18.68	0.507	0.458
Non edible: edible	2.52	2.75	0.091	0.216
<b>Cuts (% CW)</b>				
Leg	34.16	33.70	0.592	0.727
Loin	10.07	10.19	0.285	0.842
Rack	12.75	13.99	0.648	0.377
Breast and shank	16.38	15.11	0.494	0.222
Neck and shoulder	26.62	26.99	0.598	0.784
<b>Organ weights (% PSW)</b>				
Heart	0.39	0.38	0.009	0.541
Liver	1.36	1.36	0.004	0.976
Kidney	0.22	0.23	0.006	0.502
Spleen	0.30	0.27	0.012	0.211
Testis	0.73	0.72	0.061	0.956
Lungs and trachea	1.31	1.24	0.043	0.456
<b>Chemical composition of LD (% fresh basis)</b>				
Moisture	74.88	71.82	0.770	0.031
Protein	20.37	20.20	0.689	0.914
Fat	2.45	4.75	0.922	0.237
Ash	4.51	4.98	0.552	0.700

GNC: Groundnut cake, CSC: Castor seed cake, SEM: Standard error mean

9.14% raw CSC diets, respectively. Oliveira *et al.* (2015b) observed no effect on proximate composition of *L. dorsi* muscle and pH of carcass with feeding of castor deoiled cake to young goats for 165 day.

The lambs fed either of the protein sources did not reveal any changes of gross pathological significance in any of the organs with few exceptions of mild to moderate congestion of lungs, liver and kidney in lambs fed CSC. The heart, spleen and testes of lambs fed GNC did not reveal any changes of pathological significance. Purushotham *et al.* (1985a) reported congested intestinal mucous membrane with thick mucous exudate, congested kidney with hemorrhages in sheep fed 30% CSC replacing GNC for 8 months. Microscopically, Rao *et al.* (1986) reported dose related acute tubular necrosis in kidneys, congestion in portal tracts and sinusoids with occasional focal necrosis in liver and lymphocytic infiltration in intestines in sheep fed 30% CSC in concentrate mixture. Hepatic necrosis and acute tubular necrosis in kidneys was observed by Aslani *et al.* (2007) in sheep

fed castor beans in miscellaneous garden waste. Similarly, Okoye *et al.* (1987) reported focal areas of necrosis and loss of hepatocytes with congested tissues around blood vessels, focal accumulation of lymphocytes and hyperplasia of bile ducts in liver, necrotic lesions with depletion of lymphocytes and macrophages in spleen, degeneration and severely congested kidney with pyknotic renal epithelial cells in some areas of kidneys of broilers fed 10-25% roasted CSC. In the present study also, lambs fed CSC diet showed few marked changes of pathological significance in liver, kidney, lungs and intestines. In liver, marked areas of necrosis (mild to moderate), moderate to severe congestion of blood vessels and hemorrhages at few places were observed (Fig. 1). Marked structural loss with increased kupffer cell activity at few places and reticulo endothelial cell hyperplasia was observed. In kidney, swollen tubules, increase in glomerular tuft and mononuclear cell infiltration predominantly neutrophils were observed (Fig. 2). In lungs, the thickness of alveolar septa with rupture of alveolar walls and infiltration of mononuclear cells around the areas of bronchus was seen. The intestines showed broad villi with infiltration of neutrophils, goblet cell activity was mildly decreased (Fig. 3). Though no effect on growth performance, nutrient utilization, hematology and biochemical constituents was observed, the histopathological lesions in above organs indicated that antinutritional factors with inclusion of 10% CSC in complete diets of lambs

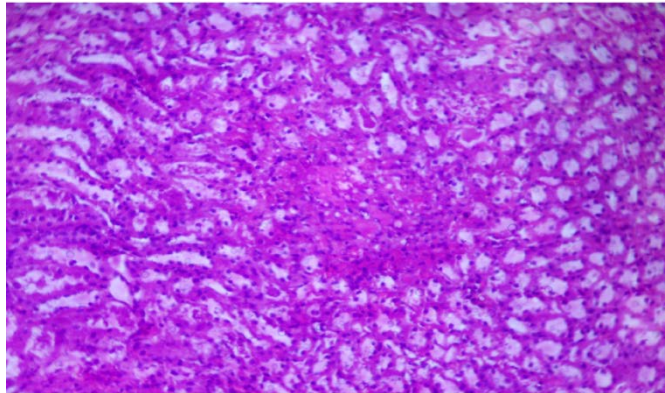


Fig. 1: Liver of sheep fed castor seed cake based complete diet showing focal areas of degeneration and lymphocytic infiltration with increased sinusoidal space H and E×200

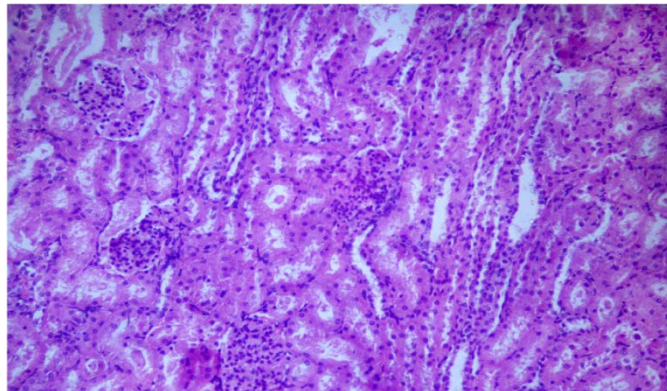


Fig. 2: Kidney of sheep fed with castor seed cake based complete diet showing lymphocytic infiltration in glomeruli and tubules. H and E×200

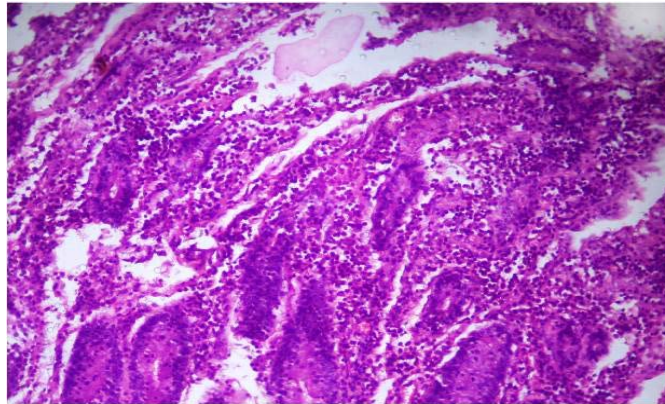


Fig. 3: Intestine of sheep fed with castor seed cake based complete diet showing mononuclear infiltration in the sub-mucosa H and E×200

had adverse effect on these vital organs and could affect the performance if fed for further longer duration beyond 5 months. On the other hand, Gowda *et al.* (2009) observed no deleterious effect on the histological architecture of liver, kidney, spleen and intestines of sheep fed 12.3% CSC in total mixed rations, which might be due higher age of animals (2.5 years) compared to 9 months old lambs used in present study.

## CONCLUSION

The results suggested that replacing groundnut cake completely by castor seed cake had no effect on growth performance, nutrient digestibility and carcass characteristics in lambs. But the humoral and cell mediated immune response was depressed with feeding of castor seed cake diets. Though, no effect on growth performance, nutrient utilization, hematology and biochemical constituents was observed, the histopathological lesions in liver, kidney, lungs and intestines indicated that antinutritional factors (ricin) with inclusion of 10% CSC in complete diets of lambs had adverse effect on their vital organs and could affect the performance if fed for further longer duration beyond 5 months.

## ACKNOWLEDGMENT

The financial help provided by Indian Council of Agricultural Research, Government of India, New Delhi under 'Young Scientist Award' to carry out the above research work is greatly acknowledged.

## REFERENCES

- AOAC., 2000. Official Methods of Analysis. 16th Edn., Association of Official Analytical Chemists, Washington, DC., USA.
- Adedeji, J.A., D.F. Apata, O.A. Aderinola, T.A. Rafiu and S.R. Amao, 2006. Performance and haematological/serum characteristics of rabbits fed boiled castor seed cake based diet. *World J. Zool.*, 1: 91-93.
- Akande, T.O. and A.A. Odunsi, 2012. Nutritive value and biochemical changes in broiler chickens fed detoxified castor kernel cake based diets. *Afr. J. Biotechnol.*, 11: 2904-2911.

- Albretsen, J.C., S.M. Gwaltney-Brant and S.A. Khan, 2000. Evaluation of castor bean toxicosis in dogs: 98 cases. *J. Am. Anim. Hosp. Assoc.*, 36: 229-233.
- Anandan, S., G.K.A. Kumar, J. Ghosh and K.S. Ramachandra, 2005. Effect of different physical and chemical treatments on detoxification of ricin in castor cake. *Anim. Feed Sci. Technol.*, 120: 159-168.
- Ani, A.O. and A.U. Okorie, 2009. Response of broiler finishers to diets containing graded levels of processed castor oil bean (*Ricinus communis* L.) meal. *J. Anim. Physiol. Anim. Nutr.*, 93: 157-164.
- Arruda, I.J., L.M. Silva, C.H.A. Oliveira, F.V. Rodrigues and A.M. Silva *et al.*, 2013. Embryo production and gene expression in superovulated goats supplemented with de-oiled castor cake before and after detoxification treatment. *Anim. Prod. Sci.*, 57: 893-898.
- Aslani, M.R., M. Maleki, M. Mohri, K. Sharifi, V. Najjar-Nezhad and E. Afshari, 2007. Castor bean (*Ricinus communis*) toxicosis in a sheep flock. *Toxicon*, 49: 400-406.
- Audi, J., M. Belson, M. Patel, J. Schier and J. Osterloh, 2005. Ricin poisoning: A comprehensive review. *JAMA.*, 294: 2342-2351.
- Blaxter, K.L. and J.L. Clapperton, 1965. Prediction of the amount of methane produced by ruminants. *Br. J. Nutr.*, 19: 511-522.
- Bonsnes, R.W. and H.H. Taussky, 1945. On the colorimetric determination of creatinine by the Jaffe reaction. *J. Biol. Chem.*, 158: 581-591.
- Brandt, N.N., A.Y. Chikishev, A.I. Stonikov, Y.A. Savochnikina, I.I. Agapov and A.G. Tonevitzky, 2005. Ricin, ricin agglutinin, and the ricin binding subunit structural comparison by Raman spectroscopy. *J. Mol. Struct.*, 735-736: 293-298.
- Cobianchi, J.V., A.S. de Oliveira, J.M.S. Campos, A.V. Guimaraes and S.C.V. Filho *et al.*, 2012. [Productive performance and efficiency of utilization of the diet components in dairy cows fed castor meal treated with calcium oxide]. *Revista Brasileira de Zootecnia*, 41: 2238-2248.
- Coles, E.H., 1986. *Veterinary Clinical Pathology*. 4th Edn., W.B. Saunders Company, Philadelphia, PA., USA., ISBN-13: 978-0721618289, pp: 40.
- Culling, C.F.A., 1963. *Hand book of Histopathological Technique*. London Butterworths, Co. Ltd., London.
- De Barros, L.V., M.F. Paulino, E. Detmann, S.C.V. Filho and S.A. Loipes *et al.*, 2011. Replacement of soybean meal by treated castor meal in supplements for grazing heifer during the dry-rainy season period. *Revista Brasileira de Zootecnia*, 40: 843-851.
- De Oliveira, A.S., J.M.S. Campos, M.R.C. Oliveira, A.F. Brito and S.C.V. Filho *et al.*, 2010. Nutrient digestibility, nitrogen metabolism and hepatic function of sheep fed diets containing solvent or expeller castor seed meal treated with calcium hydroxide. *Anim. Feed Sci. Technol.*, 158: 15-28.
- Diniz, L.L., S.C.V. Filho, J.M.S. Campos, R.F.D. Valaderesi and L.D. Silva *et al.*, 2010. Effects of castor meal on the growth performance and carcass characteristics of beef cattle. *Asian-Aust. J. Anim. Sci.*, 23: 1308-1318.
- Duncan, D.B., 1955. Multiple range and multiple F tests. *Biometrics*, 11: 1-42.
- FAO., 2014. Castor seed production. Statistical Database of the Food and Agriculture Organization of the United Nations, <http://faostat.fao.org/>.
- Garg, A.K., P. Singh, V.R.B. Sastry and D.K. Agrawal, 2005. Replacement effect of groundnut-cake with castor bean-meal (*Ricinus communis*) in concentrate mixture of adult sheep. *Indian J. Anim. Sci.*, 75: 688-690.

- Gowda, N.K.S., D.T. Pal, S.R. Bellur, U. Bharadwaj and M. Sridhar *et al.*, 2009. Evaluation of castor (*Ricinus communis*) seed cake in the total mixed ration for sheep. *J. Sci. Food Agric.*, 89: 216-220.
- Gustafsson, E.J., 1976. Improved specificity of serum albumin determination and estimation of acute phase reactants by use of the bromocresol green reaction. *Clin. Chem.*, 22: 616-622.
- ICAR., 1998. Nutrient requirement of Livestock and Poultry. Indian Council of Agricultural Research, New Delhi, pp: 6.
- Kabat, E.A., M. Heidelberger and A.E. Bezer, 1947. A study of the purification and properties of ricin. *J. Biol. Chem.*, 168: 629-639.
- Kumar, S.K., 1998. Utilization of castor (*Ricinus communis*) bean meal for feeding sheep. M.V.Sc. Thesis, Indian Veterinary Research Institute, Deemed University, Izatnagar
- Nagalakshmi, D., V.R.B. Sastry, D.K. Agrawal and R.C. Katiyar, 2001. Haematological and immunological response in lambs fed on raw and variously processed cottonseed meal. *Asian-Aust. J. Anim. Sci.*, 14: 21-29.
- Naidu, M.M., M.S. Rao, G.V. Raghavan, M.R. Reddy and N.P. Purushotham, 1988. Studies on the utilization of castor bean meal by incorporating iodine in sheep rations. *Indian Vet. J.*, 65: 1100-1102.
- Okoye, J.O.A., C.A. Enunwaonye, A.U. Okorie and F.O.I. Anugwa, 1987. Pathological effects of feeding roasted castor bean meal (*Ricinus communis*) to chicks. *Avian Pathol.*, 16: 283-290.
- Olayeni, T.B., L.O. Ojedapo, O.S. Adedeji, T.A. Adedeji and S.A. Ameen, 2006. Effects of feeding varying of castor fruit meal (*Ricinus communis*) on performance characteristics of layers. *J. Anim. Vet. Adv.*, 5: 515-518.
- Oliveira, C.H.A., A.M. Silva, L.M. Silva, M.F. van Tilburg and C.C.L. Fernandes *et al.*, 2015a. Growth, testis size, spermatogenesis, semen parameters and seminal plasma and sperm membrane protein profile during the reproductive development of male goats supplemented with de-oiled castor cake. *Reprod. Toxicol.*, 53: 152-161.
- Oliveira, C.H.A., A.M. Silva, L.M. Silva, M.F. van Tilburg and C.C.L. Fernandes *et al.*, 2015b. Meat quality assessment from young goats fed for long periods with castor de-oiled cake. *Meat Sci.*, 106: 16-24.
- Oliveira, N.D., K.V. Fernandes, L.M. Crespo and L.T. Machado, 2011. Use of chemical treatment with calcium compounds to inactivate toxins and allergens from castor bean cake. *FASEB J.*, 25 (Meeting Abstract Supplement): 765.15. [http://www.fasebj.org/cgi/content/meeting\\_abstract/25/1\\_MeetingAbstracts/765.15](http://www.fasebj.org/cgi/content/meeting_abstract/25/1_MeetingAbstracts/765.15)
- Oso, A.O., W.A. Olayemi, A.M. Bamgbose and O.F. Fowoyo, 2011. Utilization of fermented castor oil seed (*Ricinus communis*, L.) meal in diets for cockerel chicks. *Arch. Zootec.*, 60: 75-82.
- Panda, A.K., 2004. Nutritional performance and immunocompetence of broiler chickens fed processed karanj (*Pongamia glabra*) cake as partial protein supplement. Ph.D. Thesis, Deemed University, Indian Veterinary Research Institute, Izatnagar, India
- Purushotham, N.P., A. Djajanegara and A. Sukmawati, 1994. Utilization of castor bean meal in concentrate mixture for sheep. *Proceedings of the 7th AAAP Animal Science Congress*, 11-16 July, 1994, Sustainable Animal Production and environment, Bali, Indonesia, pp: 477.
- Purushotham, N.P., G. Veeraraghavan, M.M. Naidu and M. Mahender, 1985a. Haematological studies on experimental feeding of castor bean meal (*Ricinus communis*) in sheep. *Indian Vet. J.*, 62: 379-382.

- Purushotham, N.P., G.V. Raghavan, M.S. Rao, M.R. Reddy and M. Mahender, 1985b. Studies on the pathology of experimental feeding of castor bean meal (*Ricinus communis*) in sheep. *Indian Vet. J.*, 62: 116-118.
- Quist, C.F., E.W. Howerth, D.I. Bounous and D.E. Stallknecht, 1997. Cell-mediated immune response and IL-2 production in white-tailed deer experimentally infected with hemorrhagic disease viruses. *Vet. Immunol. Immunopathol.*, 56: 283-297.
- Rao, M.S., N.P. Purushotham, G.V. Raghavan, M.R. Reddy and M. Mahendar, 1986. Studies on the pathology of experimental feeding of detoxified castor bean meal (*Ricinus communis*) in sheep. *Indian Vet. J.*, 63: 944-946.
- Reddy, G.R., M.R. Reddy and G.V.N. Reddy, 1986. Nutrient digestibility and rumen metabolism in buffaloes fed castor-bean-meal in the concentrate feeds. *Indian J. Anim. Sci.*, 56: 567-572.
- Reinhold, J.G., 1953. *Standard Methods of Clinical Chemistry*. 1st Edn., Academic Press, New York, pp: 88.
- Schalm, O.W., N.C. Jain and E.J. Carrol, 1986. *Schalm's Veterinary Hematology*. 4th Edn., Lea and Febiger, Philadelphia, PA., USA., ISBN-13: 9780812109429, pp: 15.
- Talapatra, S.K., S.C. Roy and K.C. Sen, 1940. The analysis of mineral constituents in biological materials. 1. Estimation of phosphorus, chlorine, calcium, magnesium, sodium and potassium in food-stuffs. *Indian J. Vet. Sci.*, 10: 243-258.
- Uechiewcharnkit, K. and C. Kanthapanit, 1997. Effects of various levels of detoxified and deallergized castor meal ration on growth performance and carcass characteristics of fattening swine. *Thammasat Int. J. Sci. Technol.*, 2: 56-60.
- Ulanova, R. and J. Kravchenko, 2013. Lactic acid bacteria fermentation for detoxification of castor bean meal and processing of novel protein feed supplement. *Int. J. Eng. Sci. Innovative Technol.*, 2: 618-624.
- Waller, G.R. and S.S. Negi, 1958. Isolation of ricin, ricinine and the allergenic fraction from castor seed pomace from two different sources. *J. Am. Oil Che. Soc.*, 35: 409-412.
- Wegmann, T.G. and O. Smithies, 1966. A simple haemagglutination system requiring small amounts of red cells and antibodies transfusion. *Transfusion*, 6: 67-73.