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Effect of dietary incorporation of green pea (*Pisum sativum* L.) Pods on body weight gain, nutrient utilization and haemato-biochemical constituents in crossbred female calves

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Abstract
A feeding trail was conducted to discern the effect of dietary incorporation of green pea pods vis-a-vis conventional mixed green fodder in crossbred female calves on growth, nutrient utilization and certain haemato-biochemical constituents for 60 days. Sixteen crossbred female calves (9-12 months of age with average body weight of 105±6.31 kg) were divided into two groups of eight each after an adaptation period of 10 days. The crossbred female calves of group I were fed with concentrate mixture @ 1.0 kg per day and mixed green fodder (containing green oats, green berseem and green mustard approximately in 1:1:1 ratio) with wheat straw @ 10:1 ratio on fresh basis and the calves in group II were fed with concentrate mixture @1.0 kg per day and green pea pods mixed with wheat straw in 10:1 ratio on fresh basis. The mixed green fodder contained 25.97% dry matter and 13.78% crude protein on dry matter basis, whereas green pea pods contained 29.87% dry matter and 17.25% crude protein on dry matter basis. The average daily body weight gains in calves of group I and group II were 695.83±73.58 g and 650.00±39.96 g, respectively. Calves of group I had significantly (P<0.05) higher dry matter intake per kg metabolic body size than of group II. The digestible crude protein intake of group II in which calves were fed with green pea pods was significantly (P<0.01) higher than group I fed with mixed green fodder. The digestibility coefficients of crude protein and crude fibre were significantly (P<0.01) higher in calves of group II than in calves of group I, whereas digestibility coefficient of ether extract was significantly (P<0.005) higher in calves of group I than of group II. Haemato-biochemical constituents were similar in both the groups of calves. It was inferred that green pea pods left after shelling of pea seeds can be used as livestock feed, as the nutritional value was similar to those of conventional mixed green fodder.

Keywords: Crossbred female calves, growth performance, nutrient utilization, green pea pods

Introduction
India has emerged as leading milk producing country in the world and ranks first in milk production, however, the productivity per animal is low (Hedge, N.G., 2006) [9]. The total livestock population is 535.78 million in the country reflected an increase of 4.6% over livestock census 2012. In India the productive life of animal-is low because of late maturity, improper nutrition and lower growth rate during early stages-of growth and development, which is attributed, either due to underfeeding or imbalanced feeding as the success-of dairy farm mainly depends on how well young stock is managed and raised. Feeding plays a very important role in growth, development and-productivity of dairy animals. By 2050 the world will need to feed an additional 2 billion people and require 70 per cent more meat and milk. The increasing future demand for livestock products, driven by increases in income, population and urbanization will impose a huge demand on feed resources. In India availability of feed stuffs both in quantitative and qualitative forms has been the major constraint for sustaining positive growth in Indian Livestock sector. To meet this demand, huge quantity of feed resources will be required, challenging sustainability of the feed production systems. In India a shortage of 25, 159 and 117 million tonnes of concentrates, green forages and crop residues, respectively, constituting a shortage of 32, 20 and 25 per cent of the requirement has been estimated (Gorti R.K *et al.*, 2012) [7]. The major constraint in the development of livestock sector is poor availability of nutrients (Sarwar *et al.*, 2002) [18]. There is a need of 13.5 and 110.3 million tons of crude protein (CP) and total digestible nutrient (TDN), respectively (Anonymous, 2006) [1] to fulfill the requirement of livestock therefore

improvement in livestock demands the efficient use of available feed resources. Using agricultural waste such as green pea pods in animal nutrition is an interesting way to reduce feed cost and environmental pollution. Such unconventional resources can act as an excellent source of nutrients and help to bridge the gap between demand and supply of feedstuffs for livestock. In addition their use can also reduce the cost of feeding and give higher profits to farmers. Field pea (Pisum sativum) is a cool-season legume crop that is palatable and nutritious as seed and forage. The pea pods contain protein 10.8%, fat 1.3%, sucrose 7.9%, glucose 11.9%, fructose 1.2%, starch 3.7%, and dietary fibre 58.6% on dry matter basis as well as iron (1.20%) which is higher than potassium (1.03%)(Mateos-Aparicio *et al.*,2010) [14]. Pea pod contains dietary fibre as a major constituent. Dietary fibre plays an important role in many physiological processes and in the prevention of diseases (Jones *et al*., 2006). After shelling peas, the left over material is empty pea pods, which contain 19.8 percent CP and 1.0 percent EE. These are rich in total soluble sugars, total phenolics, macro- and micro-elements. The fractionation of true proteins revealed that albumins had the highest concentration followed by glutelins, globulins and prolamins (Bakshi, M.P.S *et al.*, 2007) [3].

Presently there is very less information available about feeding of pea pods in crossbred female calves. Therefore the objective of the present study was to investigate the effect of dietary incorporation of pea pods on growth, nutrient utilization and haemato-biochemical constituents in crossbred female calves.

Materials and Methods

The feeding trial of 60 days duration was conducted on sixteen crossbred female calves (9-12 months of age with average body weight of 105.75±6.31 kg) which were selected from the herd of Instructional Dairy Farm, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar. After an adaptation period of 10 days, the calves were weighed for 3 consecutive days and were divided into two groups of 8 each in such a way that the average body weight in both the groups remains uniform. During adaptation period, all calves were provided concentrate mixture along with mixed green fodder (containing green oats, green berseem and green mustard approximately in 1:1:1 ratio) in order to get the animals acclimatized to the standard feed and experimental sheds. Calves were weighed for 3 consecutive days in the morning before feeding and watering. Thereafter, the calves were divided into two groups i.e. group I and group II of eight calves each in such a way that the average body weight in both the groups was approximately similar. The crossbred female calves of group I were fed with concentrate mixture @ 1kg and mixed green fodder (containing green oats, green berseem and green mustard approximately in 1:1:1 ratio) along with wheat straw at the rate 10:1 ratio on fresh basis. The calves in group II were fed concentrate mixture @ 1 kg plus green pea pods and wheat straw mixed in in 10:1 ratio on fresh basis.

Digestion trial: A 6-day digestion trial was conducted at 45 days of feeding in both the groups in which quantitative collection of total faeces voided on 24 hourly bases was collected. Representative samples of offered and leftover green pea pod, concentrate mixture and mixed green fodder as well as faeces were sampled and analysed for proximate principles viz., dry matter, crude protein, ether extract, crude fibre, nitrogen-free extract, total ash as per the procedure of AOAC (2003) [2].

Collection of blood and serum samples: Blood samples were collected from all the experimental female calves of both the groups on 0 day and 60th day of the feeding trial. About 5 ml blood samples were collected aseptically from jugular vein using sterile disposable syringes. Collected blood samples were divided in two parts. One part (about 2 ml) in heparinised vials for analysis of haematological estimations and second part (about 3 ml) was collected in sterilized vials for serum separation. For separation of serum, collected sample was allowed to stand at room temperature in slanting position for clot formation for three to four hours. Freshly collected blood samples were subjected to haematological studies for estimation of haemoglobin concentration by method described by Drabkin and Austin (1932) [6], Packed cell volume (PCV) was estimated by using haematocrit method as described by Dacie and Lewis (1975) [5], Total erythrocyte counts (TEC) and total leukocyte counts (TLC) were done by using haemocytometer as per the method described by Schalm *et al*. (1975) [19]. Differential leukocyte counts (DLC) were done as per the method described by Schalm *et al*. (1975) [19]. Serum samples were analyzed for total protein, albumin, globulin, albumin globulin ratio, cholesterol, glucose, triglycerides and blood urea nitrogen estimation by the method described by (Young, 1997) [23] using diagnostic kit manufactured by ERBA diagnostics Ltd. Difference between treatments means were compared using Duncan's multiple range test (Kramer, 1957) [11].

Results and Discussion

The chemical compositions of the experimental feeding stuffs fed to experimental crossbred female calves are presented in Table 1. The concentrate fed to both the groups at the rate of 1 kg to each calf contained crude protein 20.37%, ether extract 3.72%, crude fibre 6.34%, nitrogen-free extract 59.82%, ash 9.75%, organic matter 90.25% and total carbohydrates 66.16% on dry matter basis.

Nutrient intake and utilization

The data on average daily dry matter intake during digestion trial in crossbred female calves is presented in Table 2. During digestion trial, the total dry matter intake and total dry matter intake per 100 kg body weight was similar in group I and II. The dry matter intake per kg metabolic body size of calves of group I and II was 116.92±2.28 and 109.29±1.48 g, respectively. The dry matter intake per kg metabolic body size in calves differed significantly (P<0.05) between both the groups and it was significantly higher in group I fed mixed green fodder than the group II fed with pea pods. The present findings are in agreement with the observation of Kumar *et al*. (2015) who reported that the crossbred calves fed with total mixed ration comprising mixed green fodder had higher dry matter intake, dry matter intake per 100 kg body weight and dry matter intake per kg metabolic body size during digestion trial. During digestion trial there was no significant difference in daily total digestible nutrients (TDN) intake, total digestible nutrients (TDN) intake per 100 kg body weight and the total digestible nutrient intake/ $W^{0.75}$ kg between both the groups of calves. The digestible crude protein intake in group I was 0.41±0.01 kg and group II whereas in group II it was 0.49±0.003 kg. The digestible crude protein intake in calves of group II was significantly (P<0.01) higher than group I.

The findings are in agreement with Hayashi *et al.* (2007) [8] who reported that field pea hay increased the digestible crude protein intake in Murrah buffaloes. The digestible crude protein intake per 100 kg body weight was 304.85±13.94 g in group I and 363.64±16.77 g in group II respectively. The digestible crude protein intake per 100 kg body weight of group II was higher than in group I ($P<0.05$). The digestible crude protein intake / kg $W^{0.75}$ of group I and group II was 10.38±0.29 g, 12.39±0.39 g, respectively. The digestible crude protein intake/ kg $W^{0.75}$ was significantly ($P<0.01$) higher in group II in which calves were fed with green pea pods as compared to the calves of group I given mixed green fodder.

The digestibility co-efficient of dry matter, organic matter and nitrogen free extract did not differ significantly with each other in both the groups (Table 3). Bastidia Gracia *et al.* (2011) [4] also reported that the total dry matter digestibility and organic matter digestibility were not different in sheep fed with field pea hay over sorghum grain/ oat straw diet. The digestibility for crude protein in group I was 57.48±0.72% and in group II was 66.33±0.95% and it differed significantly between both the groups ($P<0.01$) which might be due to higher intake of crude protein in group I as compared to group II. The digestibility for ether extract in group I was 70.03±1.22% and in group II was 63.85±1.52%. Significant difference was observed in ether extract digestibility between both the groups. Calves of group I had higher ether extract digestibility than the calves of group II ($P<0.05$), which might be due to higher intake of ether extract in animals of group I. The digestibility for crude fibre was 46.08±2.88 % and 59.71±1.53 % in group I and II, respectively. There was significant ($P<0.01$) difference in crude fibre digestibility between both the groups. Crude fibre digestibility of calves of group II fed with green pea pods was higher than the calves of group I fed with mixed green fodder, which may be due to the fact that calves fed with pea pods is fermented better than the mixed green fodder which was evident from the composition of residue left by the calves. Residue of group I contained mainly green fodder/ stem but in group II it was mainly wheat straw and they consumed most of the green pea pod and the fibre of green pea pods was probably digested in a better way as compared to group I calves consumed mixed green fodder and wheat straw. Stein *et al.* (2004) [21] reported that the nutrients of field pea were highly digestible by growing pigs.

Body weight gain: The growth trial of 60 days was conducted to study the effect of feeding green pea pods on body weight and growth of crossbred calves. The average body weights of crossbred calves at fortnightly intervals during experimental feeding period are presented in Table 2. The initial body weight of calves of group I and II was 105.75±6.31 and 105.75±5.68 kg, respectively. There was no significant difference in initial body weight and final body weight between both the groups.

The average body weight gain in crossbred calves of group I was 41.75±4.41 kg and of group II was 39.00±2.40 kg. The average daily body weight gain in crossbred calves of group I was 695.83±73.58 g and group II was 650.00±39.96 g (Table 2). There was no significant difference in average daily body weight gain between the crossbred calves of group I and II.

Blood haematological and biochemical constituents:

The average values of blood glucose concentration, total serum protein concentration, serum albumin, serum globulin, albumin: globulin ratio, triglycerides and cholesterol concentrations was found similar in both the groups at 0 day and at 60 day (Table 4). The blood glucose concentration, concentration, total serum protein concentration, serum albumin, serum globulin, albumin: globulin ratio, triglycerides and cholesterol concentrations did not differ significantly ($P>0.05$) between both the groups of crossbred calves at 0 day and 60 day. The average blood urea concentration at 0 day was 23.78±0.13 mg/dl in group I and 24.00±0.17 mg/dl in group II, whereas at 60 day it was 23.79±0.13 mg/dl in group I and 24.22±0.13 mg/dl in group II, respectively. The average blood urea concentration did not differ significantly in both the groups at 0 day. The blood urea concentration was higher in group II than in group I at 60th day. The blood urea concentration of group II fed with green pea pods had higher value than the group I fed with mixed green fodder which may be due to higher DCP intake in group II as compared to group I. The values of glucose concentration, total protein concentrations, serum albumin, serum globulin concentration, albumin : globulin ratio, triglyceride, cholesterol and blood urea concentration were within normal range as mentioned by Kaneko *et al*. (2008) [10]. Petit *et al*. (1997) [15] also reported higher serum urea concentration in cows fed extruded peas as compared to cows fed with soybean meal. Petit *et al*. (1997) [15] also reported that there was no effect of serum total protein concentration in the cows fed with extruded peas and soybean meal. Volpelli *et al.* (2012) [22] reported that when the cows fed with pea and faba beans as compared to soybean meal, there is significant increase in blood urea level of cows fed with pea and faba beans. Masoero *et al.* (2006) [13] reported no differences in total protein, albumin, globulin, triglycerides in animals fed expanded pea diet. Kumar *et al*. (2015) [12] also reported no significant difference in glucose, serum total protein, albumin, globulin, albumin : globulin ratio, triglycerides and cholesterol.

The haematological parameters viz; haemoglobin content, packed cell volume, total erythrocyte counts, total leucocyte counts and per cent neutrophils, leucocytes, monocytes, eosinophils and basophils did not differ significantly between the two groups of calves which indicated that feeding of moderate diet maintain the haematological values. The findings of the present study were within the normal range for cattle as reported by Kaneko *et al*., (2008) [10]. Similar values were obtained of haemoglobin, packed cell volume, total erythrocyte counts, total leukocyte counts, neutrophills, lymphocytes, eosinophills, monocytes and basophills as mentioned by Reece *et al*., (2015) [17]. Kumar *et al.*, (2015) [12] also reported similar observations regarding haematological parameters of crossbred calves. The value of haemoglobin was within the normal range and did not differ significantly between the groups of crossbred calves as reported by Radostits *et al*., (1995) [16].

Conclusion

It could be concluded that the dietary incorporation of green pea pods had positive effects on growth, nutrient utilization and haemato-biochemical constituents in crossbred female calves and green pea pods left after shelling of pea seeds can be used as livestock feed, as the nutritional value was similar to those of conventional mixed green fodder.

Table 1: Chemical composition of feeding stuffs (% Dry matter basis) fed to experimental crossbred female calves during experimental feeding period.

Nutrients	Feeding stuffs			
	Green pea pods	Mixed green fodder	Wheat straw	Concentrate Mixture
Dry matter	29.87	25.97	91.87	89.73
Crude protein	17.25	13.78	3.42	20.37
Ether extract	2.36	3.50	1.22	3.72
Crude fibre	27.02	24.53	33.90	6.34
Nitrogen free extract	49.12	49.56	50.09	59.82
Ash	4.25	6.75	11.37	9.75
Organic matter	95.75	93.25	88.63	90.25
Total carbohydrates	76.14	75.97	83.99	66.16

Table 2: Average body weight gain, daily dry matter and nutrient intake in crossbred calves during digestion trial

Particulars	Group I	Group II
Average initial body weight (kg)	105.75±6.31	105.75±5.68
Average final body weight (kg)	147.50±9.15	144.75±6.31
Average total body weight gain(kg)	41.75±4.41	39.00±2.40
Average body weight gain (g/day)	695.83±73.58	650.00±39.96
Nutrient intake		
Average body weight (kg)	138.56±8.15	138.06±6.09
Average metabolic body size (kg)	40.28±1.83	40.22±1.37
Total dry matter intake (kg)	4.70±0.20	4.33±0.15
Dry matter intake/100 kg bodyweight (kg)	3.42±0.09	3.20±0.06
Dry matter intake/ $W^{0.75}$ kg(g)*	116.92±2.28	109.29±1.48
Total digestible nutrient intake (kg)	2.90±0.15	2.99±0.12
Total digestible nutrient intake/100 kg body weight (kg)	2.10±0.05	2.17±0.05
Total digestible nutrient intake/ $W^{0.75}$ kg (g)	71.86±1.42	74.35±1.08
Digestible crude protein intake (kg)**	0.41±0.01	0.49±0.01
Digestible crude protein intake/100 kg body weight (g)*	304.85±13.94	363.64±16.77
Digestible crude protein intake/$W^{0.75}$ kg(g)**	10.38±0.29	12.39±0.39

Table 3: Digestibility coefficients of nutrients in crossbred female calves during digestion trial.

Nutrients	Groups	
	I	II
Dry matter	66.04±0.89	65.73±0.71
Organic matter	68.58±0.78	67.10±0.62
Crude protein**	57.48±0.72	66.33±0.95
Ether extract*	70.03±1.22	63.85±1.52
Nitrogen-free extract	76.93±2.06	79.12±0.79
Crude fibre**	46.08±2.88	59.71±1.53

The mean values in a row differ significantly from each other $^{*}P<0.05$, $^{**}P<0.01$.

Table 4: Blood serum biochemical constituents and haematological parameters in crossbred calves during experimental feeding period at 0 day and 60 day.

Particulars	Groups	
	I	II
Biochemical parameters **Blood glucose (mg/dl)**		
0 day	72.57±1.12	73.39±1.58
60 day	73.28±0.65	74.31±0.77
Total serum protein (g/l)		
0 day	65.98±1.92	65.90±1.50
60 day	69.21±0.64	70.47±1.08
Serum albumin (g/l)		
0 day	32.03±0.91	32.19±0.61
60 day	32.60±0.54	33.99±1.05
Serum globulin (g/l)		
0 day	33.96±1.63	33.71±0.99
60 day	36.61±0.62	36.49±0.75
Albumin: globulin ratio		
0 day	1:0.96±0.05	1:0.95±0.02
60 day	1:0.89±0.02	1:0.93±0.04
Blood urea (mg/dl)		
0 day	23.78±0.13	24.00±0.17
60 day	23.79±0.13	24.22±0.13

Serum triglycerides (mg/dl)		
0 day	12.09±0.05	12.16±0.01
60 day	12.36±0.03	12.42±0.06
Serum cholesterol (mg/dl)		
0 day	95.96±0.45	96.59±0.82
60 day	98.10±0.51	98.30±0.42
Haematological parameters Haemoglobin (g/dl)		
0 day	9.58±0.50	9.84±0.32
60 day	10.60±0.30	11.36±0.28
PCV (%)		
0 day	31.63±1.03	31.13±0.91
60 day	34.50±0.78	35.62±1.00
TEC (%)		
0 day	7.50±0.72	7.26±0.72
60 day	9.38±0.36	9.72±0.37
TLC (%)		
0 day	7.29±0.45	7.35±0.55
60 day	7.76±0.20	8.06±0.43
Neutrophils (%)		
0 day	24.87±3.45	23.50±3.19
60 day	27.25±2.74	28.13±2.86
Lymphocytes (%)		
0 day	59.63±4.71	58.87±1.01
60 day	64.87±3.65	61.87±3.75
Eosinophil's (%)		
0 day	2.25±0.42	1.75±0.25
60 day	2.75±0.37	2.25±0.25
Monocytes (%)		
0 day	2.87±0.29	3.25±0.36
60 day	3.25±0.25	3.25±0.25
Basophils (%)		
0 day	0.13±0.13	0.25±0.16
60 day	0.25±0.16	0.37±0.18

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