Haryana Veterinarian (Dec., 2025) **64(2),** 93-96 ISSN 0033-4359 (P)

Research Article

SHORT AND LONG TERM EFFECT OF SUPPLEMENTATION OF TEA SEED AND TEA SEED SAPONIN EXTRACT ON STATUS AND RETENTION OF CALCIUM AND PHOSPHORUS IN GROWING GADDI GOAT

MANJEET KUMAR*, A. KANNAN1, R. BHAR, G. MAL, A. GAURAV and RAVINDRA JADHAV
Indian Veterinary Research Institute, Regional Station, Palampur, Himachal Pradesh-176061, India

Received: 22.09.2023; Accepted: 28.01.2025

ABSTRACT

The present study was conducted to evaluate blood calcium (Ca) and phosphorus(P) status and their retention during short term and long term trial in growing Gaddi goats supplemented with either tea seed or tea seed saponin (TSS) extract. Eighteen growing goats were distributed into three groups, T_0 (control), T_1 and T_2, were fed a basal diet consisting of concentrate mixture (CM) and oat fodder (50:50). In T_1, apart of the ingredients of CM was replaced by tea seeds (5.2%) and in T_2 byTSS (0.8%). Daily saponin intake in T_1 and T_2 were equal (0.4% of dry matter intake). The results showed that the fecal excretion of Ca was significantly ($P<0.05$) lower in TSS fed groups during both short term as well as long term. However, there was no significant difference in Ca and P intake, urinary excretion and retention in all the groups in both short as well as long term. The blood Ca and P levels remained within normal range and were comparable among the groups. It was concluded that supplementation of both TSS and tea seed did not affect the Ca and P status and also their retention in growing Gaddi goats.

Keywords: Calcium, Goats, Phosphorus, Tea seed saponin

How to cite: Kumar, M., Kannan, A., Bhar, R., Mal, G., Gaurav, A., and Jadhav, R. (2025). Short and long term effect of supplementation of tea seed and tea seed saponin extract on status and retention of calcium and phosphorus in growing Gaddi goat. *Haryana Veterinarian.* **64(2)**: 93-96.

Feed and fodder shortage is the main constraint in the animal production system in Himachal Pradesh. Annual requirement of dry fodder, concentrates and green fodder in Himachal Pradesh for feeding livestock is 40.0, 25.5 and 43.0 lakh tons, whereas availability is only 30.0, 7.65 and 16.0 lakh tons, indicating a deficit of 25.0, 70.0 and 62.8 %, respectively (Vashisht and Pathania, 2001). The shortage is very acute during winter season and mainly met through transportation of feeds from Punjab and other neighbouring states to Himachal Pradesh. Therefore, there is an urgent need for identifying and characterising the available feed resources in the region. Kangra valley is one of the important areas of tea cultivation in the North Western Himalayan Region. The tea plant variety in Kangra region is different from the tea plants of Assam and Nilgris. Moreover, the tea plants of this area produce seeds in abundance as compared to other regions. The saponin content of the tea seed is also high. Recently, due to ban on the use of antibiotics and synthetic chemicals as feed additives, especially in western countries, use of natural ingredients containing phytochemicals is gaining importance. Some reports suggest that feeding tea seed saponin in the diet of ruminants improve animal production (Hu *et al.*, 2006). However, it has been reported that some phytochemicals interfere with calcium and phosphorus metabolism resulting in deficiency or imbalance of one or the other mineral. Some saponins are also known to from insoluble saponin mineral complexes, with iron, zinc and calcium (Millgates and Roberts,1995) resulting in their deficiencies. Deficiency of calcium reduces the ability of immune cells to respond to stimuli (Kimura *et al.*, 2006) and decreases smooth muscle contraction, thus predisposes animals to infection (Goff, 2008). Severe deficiency may lead to anorexia, progressive weakness, reduced milk yield etc. However, there are no reports available regarding the effect of feeding tea seed or tea seed saponin on bloodcalcium and phosphorus levels and their balance or retention in goats. Moreover, the critical minerals in hilly regions are calcium and phosphorus (Kannan and Bhar, 2016). Inview of the above, the present study was undertaken to evaluate the effect of supplementation of tea seed and tea seed saponin extract on calcium and phosphorus status and their retention during short term and long term in growing Gaddi goats.

*Corresponding author: dr.manjeet.13@gmail.com

MATERIALS AND METHODS

Tea (*Camellia sinensis*) seeds used in the study were sourced from tea garden of Department of Tea Husbandry and Agroforestry, Himachal Pradesh Krishi Vishwa Vidyalaya, Palampur. Saponins were extracted from tea seeds as per the procedure of Jadhav *et al.* (2017). On dry matter basis, saponin yield was 15.35%. Detection and purity estimation of saponin was performed on a HPLC.

Eighteen Gaddi goats of about 6-8 months of age were allotted into three groups, T_0 (control), T_1 and T_2, consisting of six animals each in completely randomized design. The goats were fed for maintenance and for the growth rate of 40 g/day as per ICAR (2013) feeding standards,

on basal diet consisting of concentrate mixture (CM) and oat fodder (50:50 ratio) during feeding trial of 120 days. In T_1, apart of the ingredients of CM was replaced by tea seeds (5.2%) and in T_2 by tea seed saponin (0.8%). Daily saponin intake in T_1 and T_2 were equal (0.4% of dry matter intake). Animals were offered concentrate mixture daily at 10 AM and oat fodder 2 hr after CM feeding. Animals were watered 3 times a day. Two metabolism trials were conducted, 1st after 21 days and 2nd after the 90 days of feeding trial. Each metabolism trial consisted of 6 days collection period after adaptation of goats in the metabolic cages. The faeces and urine were collected daily at 10 AM weighed and stored appropriately for further analysis. The samples of feed offered, residue and faeces were dried in an oven at 60º C until constant weight to estimate DM and ground to pass 1mm sieve for further analysis. Mineral extract was prepared from fodder, feed, faeces and urine. Calcium was estimated by the method of Talapatra *et al.* (1940) and phosphorus was estimated by spectrophotometer method (Sastry *et al.*, 1999).

Blood samples were collected in serum tubes from all goats on 0th, 30th, 60th, 90th and 120th day before morning feeding and watering by puncturing jugular vein. Serum tubes without anticoagulant were kept slanted at room temperature for serum separation and were stored at -20º C for further analysis. Serum calcium concentration was measured by O-Cresolphthalein Complexone (OCPC) method. OCPC reacts with Ca in alkaline solution to form a purple coloured complex. The intensity of the purple colour formed is proportional to the Ca concentration and was measured photometrically at 575 nm wave length (Sastry et al., 1999). Serum phosphorus concentration was measured by UV Molybdate method. In acidic medium, inorganic phosphorus reacts with ammonium molybdate to form phosphomolybdate complex. This colorless complex was measured at 340 nm and which was directly proportional to the concentration of inorganic phosphorus in serum(Sastry *et al.*,1999).

The data generated from the experimental study were subjected to statistical analysis by following the standard procedures of Snedecor and Cochran (1989) with the help of SAS 9.2 statistical software (SAS, 2010). Comparison between groups was made by one way analysis of variance by using the model $y_{ij} = \mu + T_i + e_{ij}$, Where y_{ij} = observed value of the response variable for i^{th} treatment; μ = General mean effect; T_i = i^{th} treatment effect; e_{ij} = random error.

The data on blood levels of calcium and phosphorus were analyzed in repeated measures ANOVA. Results were presented as means and standard error of means (SEM). The significance of difference between means was compared using Duncan`s multiple range test. In all cases, statistical differences were accepted if $P<0.05$.

RESULTS AND DISCUSSION

Most of the studies using phytochemicals were done mainly in *in vitro* or for very short term. So, we have studied the effect of feeding tea seed as well as tea seed saponin both during short term as well as long term. The calcium and phosphorus retention after 21 days and 90 days of feeding tea seed or tea seed saponinare presented in Table I and II, respectively. During both short term and long termmetabolic trail, calcium intake and urinary calcium outgo did not differ significantly between T_0, T_1 and T_2, but the faecal calcium outgo was significantly lower ($P<0.05$) in T_2 group. Though the daily calcium retention (g/d and g/kgW0.75) was numerically higher in T_1 and T_2 as compared toT_0, however, it was not significant. Calcium retention as percentage of intake and as percentage absorbed also did not differ in different groups. Johnson *et al.* (1986) observed that some type of saponins improve intestinal permeability thereby increasing their absorption. Saponins from Gypsophila, quillaza, clover and lucerne reduces transmural potential difference (the electrochemical gradient that acts as driving force for active nutrient transport across the brush border membrane of the intestine) in rat, resulting in increase in the permeability of the brush border, which increase uptake of macromolecules. In contrast, feeding saponin rich alfalfa meal resulted in decreased plasma calcium and other minerals in pigs (Pond and Yen, 1984) may be due to formation of insoluble saponin-mineral complex (Millgates and Roberts,1995). However, in the present study no such effect was observed on feeding tea seed and tea seed saponin.

During both short term and long term, P intake, faecal and urinary P outgo did not differ significantly between T_0, T_1 and T_2. Phosphorus retention was 0.55, 0.49 and 0.56 g/day after 21 days and 0.47,0.24 and 0.38 after 90 days of feeding in T_0, T_1 and T_2, respectively. Phosphorus retention as percentage of intake and as percentage absorbed also did not differ among the groups. Other workers also did not observe any negative effect of saponin supplementation on P retention in animals.

Serum calcium and phosphorus levels analyzed on day 0, 30, 60, 90 and 120 during the feeding trial are presented in Table III. The mean blood calcium levels which were 8.18, 8.26 and 8.24 mg/dl initially increased to 9.31, 8.80 and 9.67 mg/dl in T_0, T_1 and T_2, respectively indicating that there is adequate calcium uptake in all the three groups. Similarly, blood phosphorus levels which

Table 1. Intake and retention of calcium and phosphorus in Gaddi goats after 21 days of feeding tea seed and tea seed saponin

Attributes	Treatment			SEM	P Value
	T_0	T_1	T_2		
Diet Ca :P ratio	1.92:1.0	1.98:1.0	1.95:1.0		
Ca					
Ca intake (g/d)	4.58 ± 0.64	5.73 ± 0.46	5.68 ± 0.42	0.31	0.248
Ca excreted through faeces (g/d)	2.09[a] ± 0.16	2.06[a] ± 0.16	1.45[b] ± 0.10	0.15	0.018
Ca excreted through urine (g/d)	1.24 ± 0.08	1.30 ± 0.19	1.78 ± 0.40	0.14	0.272
Ca retention (g/d)	1.26 ± 0.49	2.36 ± 0.71	2.45 ± 0.69	0.36	0.395
Ca retention (% intake)	35.33 ± 10.38	38.01 ± 8.80	43.13 ± 8.28	5.28	0.526
Ca retention (% absorbed)	66.20 ± 11.15	64.30 ± 9.6.5	57.91 ± 9.49	5.70	0.649
Ca retention (g/kg $W^{0.75}$)	0.16 ± 0.04	0.23 ± 0.06	0.26 ± 0.05	0.03	0.268
P					
P intake (g/d)	2.38 ± 0.32	2.89 ± 0.26	2.92 ± 0.26	0.16	0.345
P excreted through faeces (g/d)	1.25± 0.10	1.59± 0.14	1.56± 0.19	0.10	0.059
P excreted through urine (g/d)	0.57 ± 0.09	0.80 ± 0.09	0.78 ± 0.07	0.05	0.168
P retention (g/d)	0.55 ± 0.18	0.49 ± 0.20	0.56 ± 0.27	0.11	0.972
P retention (% intake)	20.24 ± 5.35	14.83 ± 5.16	17.11 ± 7.02	3.17	0.794
P retention (% absorbed)	47.09 ± 20.04	37.30 ± 7.04	42.01 ± 10.26	4.63	0.533
P retention (g/kg $W^{0.75}$)	0.05 ± 0.03	0.04 ± 0.01	0.05 ± 0.02	0.01	0.962

[a,b]Means bearing different superscripts in a row differ significantly.

Table 2. Intake and retention of calcium and phosphorus in Gaddi goats after 90 days of feeding tea seed and tea seed saponin

Attributes	Treatment			SEM	P Value
	T_0	T_1	T_2		
Diet Ca:P ratio	2.31:1.0	2.30:1.0	2.04:1.0		
Ca					
Ca intake (g/d)	6.42 ± 0.33	7.49 ± 0.52	6.19 ± 0.35	0.27	0.106
Ca excreted through faeces (g/d)	3.91[a] ± 0.17	4.25[a] ± 0.24	3.37[b] ± 0.19	0.23	0.040
Ca excreted through urine (g/d)	0.60 ± 0.07	0.75 ± 0.09	0.70 ± 0.15	0.07	0.093
Ca retention (g/d)	1.91 ± 0.28	2.48 ± 0.38	2.12 ± 0.29	0.19	0.282
Ca retention (% intake)	29.70 ± 3.13	32.25 ± 2.80	34.24 ± 2.80	2.27	0.058
Ca retention (% absorbed)	76.09 ± 1.49	76.04 ± 4.03	75.16 ± 2.00	1.77	0.262
Ca retention (g/kg $W^{0.75}$)	0.19 ± 0.02	0.24 ± 0.02	0.20 ± 0.02	0.01	0.095
P					
P intake (g/d)	2.78 ± 0.44	3.26 ± 0.25	3.03 ± 0.20	0.17	0.560
P excreted through faeces (g/d)	1.79 ± 0.26	2.15 ± 0.06	2.29 ± 0.16	0.10	0.163
P excreted through urine (g/d)	0.52 ± 0.19	0.86 ± 0.25	0.36 ± 0.01	0.12	0.221
P retention (g/d)	0.47 ± 0.19	0.24 ± 0.07	0.38 ± 0.11	0.07	0.532
P retention (% intake)	16.65 ± 6.31	7.53 ± 2.52	12.19 ± 3.13	2.30	0.620
P retention (% absorbed)	18.34 ± 26.90	24.59 ± 8.82	45.62 ± 8.75	9.22	0.499
P retention (g/kg $W^{0.75}$)	0.04 ± 0.01	0.02 ± 0.008	0.03 ± 0.01	0.007	0.604

[a,b]Means bearing different superscripts in a row differ significantly.

were 4.75, 4.71 and 4.96 mg/dl initially changed to 4.73, 5.01 and 5.15 mg/dl per cent, respectively indicating that there was adequate intake of P in all the three groups. In contrast with our findings, Pond and Yen (1984) observed decrease in calcium levels due to feeding of high saponin containing alfalfa meal on growing and finishing pig. Southern *et al.* (1988) also observed decrease in mineral levels due to feeding gypsophila saponin in rats. However, in the present study no such effect was observed. As hypothesized earlier by Patra and Saxena (2009) the effect of saponin varies depending on the type of saponin, saponin structure, dose level, animal type and dietary composition. Chemically saponins are high molecular weight glycosides in which sugars linked to a triterpene or steroidal aglycone moiety. A large number of saponins could be possible depending upon the modifications of the ring structure of aglycone moieties and number of sugars added to it and inturn producing different biological properties. The tea seed saponins are triterpenoid saponins and acidic in nature as compared to steroidal saponins which are neutral. The dose level used in the study did not create any deficiency of calcium and phosphorus.

Table 3. Calcium and phosphorus statusin Gaddi goats due to feeding of tea seed and tea seed saponin

Attributes	Treatment			Period (Mean ± SE)	P Value		
	T_0	T_1	T_2		P	T	T*P
Calcium (mg/dl)							
0 day	8.18 ± 0.28	8.26 ± 0.30	8.24 ± 0.26	8.22 ± 0.15	0.067	0.211	0.897
30 day	8.27 ± 0.21	8.49 ± 0.23	8.58 ± 0.36	8.44 ± 0.14			
60 day	8.42 ± 0.23	8.18 ± 0.25	8.85 ± 0.30	8.46 ± 0.15			
90 day	8.65 ± 0.32	8.46 ± 0.29	9.05 ± 0.49	8.70 ± 0.22			
120 day	9.31 ± 0.27	8.80 ± 0.28	9.67 ± 0.50	9.24 ± 0.20			
Treatment (Mean ±SE)	8.57 ± 0.13	8.44 ± 0.16	8.88 ± 0.18				
Phosphorus (mg/dl)							
0 day	4.75 ± 0.27	4.71 ± 0.37	4.96 ± 0.34	4.80 ± 0.18	0.784	0.161	0.935
30 day	4.84 ± 0.10	4.77 ± 0.19	4.66 ± 0.18	4.76 ± 0.08			
60 day	4.72 ± 0.23	5.09 ± 0.27	5.03 ± 0.30	4.94 ± 0.15			
90 day	4.59 ± 0.05	4.89 ± 0.16	5.00 ± 0.22	4.81 ± 0.09			
120 day	4.73 ± 0.12	5.01 ± 0.23	5.15 ± 0.43	4.95 ± 0.15			
Treatment (Mean ±SE)	4.72 ± 0.07	4.90 ± 0.11	4.96 ± 0.13				

Calcium and phosphorus ratio of the diet ranged from 1.92:1.0 to 1.98:1.0 after 21 days of feeding and 2.04:1.00 to 2.31:1 after 90 days of feeding. The ratio was within the acceptable limits for feeding ruminants. Recently it was reported, that when oak leaves (which are rich in the phytochemical tannin) were fed for long term without concentrate supplementation led to calcium deposition in most of the visceral organs (Kannan and Bhar, 2016) in Gaddi goats. No such effect was seen in the study.

On the basis of above said observations from the study, it may be inferred that feeding tea seed or tea seed saponin in the diet did not alter the blood calcium and phosphorus levels. Though fecal excretion of calcium was significantly lower in tea seed saponin fed groups, however, the retention of both the minerals were comparable among the treatments. Therefore, it may be concluded that supplementation of tea seed or tea seed saponin did not affect calcium and phosphorus metabolism in Gaddi goats.

REFERENCES

Goff, J.P. (2008). The monitoring, prevention and treatment of milk fever and subclinical hypocalcemia in dairy cows. *Vet. J.* **176**: 50-57.

Hu, W.L., Liu, J.X., Wu, Y.M., Guo, Y.Q. and Ye, J.A. (2006). Effects of tea saponins on *in vitro* ruminal fermentation and growth performance in growing Boer goat. *Archiv. Anim. Nutr.* **60**: 89-97.

ICAR (2013). Nutrient Requirements of Sheep, Goat and Rabbit. (2nd Edn.), ICAR, New Delhi, India.

Jadhav, R.V., Kannan, A., Bhar, R., Sharma, O.P., Bhat, T.K., Gulati, A., Rajkumar, K., Sharma, R., Mal, G., Singh, B. and Sharma, V.K. (2017). Effect of tea (*Camellia sinensis*) seed saponin supplementation on growth performance, nutrient utilization, microbial protein synthesis and hemato-biochemical attributes of gaddi goats. *Anim. Nutr. Feed Technol.* **17(2)**: 255-268.

Johnson, I.T., Gee, J.M. Price, K.R., Curl, C. and Fenwick, G.R. (1986). Influence of saponins on gut permeability and active nutrient transport *in vitro*. *J. Nutri.* **116**: 2270-2277.

Kannan, A. and Bhar, R. (2016). Mineral imbalances in hills and temperate environments and its impact on farm animal production. In: Micro Nutrients in Animal Nutrition. [Jadhav, S.E., Pattanaik, A.K., Das, A., Garg, A.K. and Verma, A.K. (Eds)]. Centre of Advanced Faculty Training in Animal Nutrition, Division of Animal Nutrition, Indian Veterinary Research Institute, Izatnagar-243 122, India, pp. 10-17.

Kimura, K., Reinhardt, T.A. and Goff, J.P. (2006). Parturition and hypocalcemia blunts calcium signals in immune cells of dairy cattle. *J. Dairy Sci.* **89**: 2588-2595.

Millgates, J. and Roberts, D.C.K. (1995). The nutritional and biological significance of saponins. *Nutr. Res.* **15**: 1223-1249.

Patra, A.K. and Saxena, J. (2009). The effect and mode of action of saponins on the microbial populations and fermentation in the rumen and ruminant production. *Nutr. Res. Rev.* **22**: 204-219.

Pond, W.G. and Yen, J.T. (1984). Effect of level of alfalfa meal in a corn soybean meal diet on growing-finishing swine. *Nutr. Rep. Intern.* **29**: 1191-1201.

SAS (2010). SAS User's Guide. Verson 9.2. SAS Institute Inc. Cary, NC.

Sastry, V.R.B., Kamra, D.N. and Pathak N.N. (1999). Laboratory Manual in Animal Nutrition. Indian Veterinary Research Institute, Izatnagar, India.

Snedecor, G.W. and Cochran, W.G. (1989). Statistical Methods, (9th Edn.), Oxford and IBH Pub. Co., New Delhi.

Southern, S., Johnson, I.T., Gee, J.M. and Price, K.R. (1988). The effect of gypsophila saponins in the diet on mineral status and plasma cholesterol in the rat. *Br. J. Nutr.* **59**: 49-55.

Talapatra, S.K., Ray, S.C. and Sen. K.C. (1940). The analysis of mineral constituents in biological material. Part I. Estimation of phosphorus, chlorine, calcium, magnesium, sodium and potassium in feedstuffs. *Indian J. Vet. Sci. Anim. Husb.* **10**: 243-258.

Vashisht, G.D. and Pathania, M.S. (2001). Requirement and availability of feed and fodder resources for bovine population in Himachal Pradesh. *Indian J. Anim. Sci.* **71**: 367-374.