



Polyphenols rich plants extract supplementation to enhance the desaturation and antioxidant activity in goat kids

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ABSTRACT

This study aimed to investigate the effect of supplementation of tanniniferous *Terminalia chebula* plant extract at different levels on plasma antioxidative property, immunity and desaturation level in crossbred kids. Crossbred (Alpine × Beetal) kids (18) were divided into 3 equal groups and fed a basal diet containing concentrate mixture and green maize fodder for 90 days. In addition, treatment groups T1 and T2 were supplemented with aqueous extract of *T. chebula* @ 6 and 18 g/kg DM intake, respectively. Average body weight gain per day was higher ($P < 0.05$) in group T2 (58 g) compared to control (44 g) and T1 (55 g) groups. The digestibility of DM, OM and EE were also higher ($P < 0.05$) in group T2 compared to control. Superoxide-dismutase [U/g haemoglobin (Hb)/ min] and catalase (μmol s of H_2O_2 consumed/ min/mg of Hb) activities in T1, T2 groups were 3.37, 14.27 and 0.61, 27.45% lower ($P < 0.05$) than control respectively. Plasma total antioxidant activity on day 90 in T2 group was 9.60% higher than control. Total immunoglobulin concentration was 19.72 and 24.95% higher in T1 and T2 group as compared to control at 90 days of experimental feeding. In T1 and T2 group total monounsaturated fatty acid (MUFA) and polyunsaturated fatty acid (PUFA) contents in muscle were enhanced by 20.25, 24.97 and 26.36, 34.77%, whereas saturated fatty acid (SFA) was reduced by 6.97, 19.65% respectively. Results of the present study indicated that the supplementation of one percent polyphenols through aqueous extract of *T. chebula* could also be used as an efficient strategy to improve the immunity, antioxidative status and unsaturated fatty acid level in small ruminants' meat.

Key words: Antioxidant activity, Desaturation index, Plant extract, Polyphenols, *Terminalia chebula*

In animals various physiological conditions lead to production of reactive oxygen species (ROS), which causes oxidative stress. Adequate feeding regimes, nutrient supply and administration of dietary antioxidants have been suggested effective strategies to reduce metabolic and oxidative stresses in ruminants (Sawadogo *et al.* 2006). Several studies have underlined the utility of dietary antioxidants, derived from plants and their byproducts (Nayana and Janardhanan 2000, Sreepriya *et al.* 2001), which play an important role in strengthening the antioxidants defences. Harnessing the antioxidant, antibacterial and antifungal activity of some of the plant extracts has been heightened interest in the field of herbal medicine (Sreepriya *et al.* 2001).

Plant extracts rich in polyphenols exhibit antioxidant activity, in addition to it they are also reported to affect rumen biohydrogenation (Naik *et al.* 2003, Vasta *et al.* 2009, Rana *et al.* 2011). *Terminalia chebula*, a polyphenol rich plant, has been reported to exhibit a variety of biological activity including its role as antibacterial, antiviral,

antifungal activity (Cheng *et al.* 2003). Studies have further suggested effects of polyphenols on desaturation index in ruminant food products and also on total fatty acid content in ruminant meat (Vasta *et al.* 2009, Rana *et al.* 2011). Ruminant products have higher saturated fatty acid (SFA) content due to biohydrogenation of dietary unsaturated fatty acids, leading to intramuscular fat that is less unsaturated than in non-ruminants. Also lipids are the macromolecules that are most susceptible to peroxidative processes, especially when they are rich in polyunsaturated fatty acid (PUFA) (Gladine *et al.* 2007a, b). Supplementation of plant extracts such as *T. chebula* improves the oxidative stability and enhances unsaturated fatty acid content of ruminant products and could be an effective strategy to improve the healthiness of ruminant products from consumer perspective. In addition, different studies have reported that supplementation of antioxidants is necessary to preserve the health of animals and to provide oxidative stability to their products (Vasta *et al.* 2009, Gladine *et al.* 2007a,b).

Polyphenols can alter the activity and the proliferation of some ruminal microorganisms (Min *et al.* 2003). So, the *T. chebula*, was aimed to investigate the effect of polyphenol rich plant extract supplementation on the antioxidant activity, immune status and ruminal biohydrogenation.

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MATERIALS AND METHODS

Selection of animals and feeding management: Eighteen crossbred (Alpine × Beetal) male goat kids (5–6 months) were selected from the flock and divided into three groups (6 kids each) in a completely randomized block design on the basis of average initial body weight (20.07 ± 0.66 kg) as control, T1 and T2 groups. The kids in T1 and T2 groups were supplemented with aqueous extract of *T. chebula* @ 6 g/kg dry matter (DM) intake and 18g/kg DM intake, respectively in addition to control diet. The aqueous extract prepared from fruit of *T. chebula* contained 546.9 g total phenol and 497.1 g total tannins as tannic equivalent/ kg of DM. The nutrient requirements (i.e. crude protein (CP) 60 g/d and metabolisable energy (ME) 6.52 MJ/d) of kids were met by feeding compound feed and maize fodder during the 90 d feeding trial (NRC 1981). Compound feed consisted of maize grain 570 g/kg, groundnut cake (GNC) 400 g/kg, mineral mixture 20 g/kg and common salt 10 g/kg. Plant extract was mixed homogenously with the compound feed and supplemented in morning during experimental period to groups T1 and T2, respectively. The DM content of offered and refusal feed was measured daily and the actual DM intake was derived. Body weight of the animals was recorded at weekly interval for two consecutive days before offering feed and water.

Chemical and fatty acid analysis of feed sample: The roughage and concentrate were sampled at the beginning, mid and end of the experimental period. All the samples were grounded individually, labelled and analyzed for proximate composition as per AOAC (2005) and cell wall constituents as per Goering and Van Soest (1970).

The feed samples were dried at $55 \pm 1^\circ\text{C}$ and ground to pass through a sieve of 1 mm diameter. The ground feed sample were then processed by following protocol according to O'Fallon *et al.* (2007) with small modifications. In brief, fatty acids in the feed were obtained by hydrolysis with 10 N KOH at 55°C for 1.5 h with vigorous shaking for 5 sec at 20 min interval to properly permeate. After adding internal standard the sample was acidified with 24 N H_2SO_4 . The tubes were mixed by inversion and incubated at 55°C for 1.5h with hand shaking for at every 5 sec at 20 minute interval. Hexane (3 ml) was then added in cooled tubes and each tube was vortex mixed for 5 min. Tubes were then centrifuged at 1,000g for 5 min at 23°C and the hexane layer containing fatty acid methyl ester (FAME) was extracted. The same was concentrated under nitrogen and stored at -20°C until gas chromatography (GC) analysis.

For phenols and tannin analysis, the feed sample was dried at $55 \pm 1^\circ\text{C}$ and ground so as to pass through a sieve of 1 mm diameter. About 50 mg ground sample was transferred to a test tube and 5 ml diethyl ether containing 1% acetic acid (v/v) was added to remove the pigment material. After 5 min, the supernatant was carefully discarded and 5 ml of 70% aqueous acetone was added and kept overnight at 40°C . Samples were then vortexed for 5 min. After centrifugation

(1000 g for 10 min) supernatant was taken for further analysis. Total phenolics and tannins were estimated according to Folin-Ciocalteu method (Makkar *et al.* 1993). Whereas, condensed tannins were analyzed according to Porter *et al.* (1986).

Collection and processing of blood sample: Blood samples were collected from the jugular vein of individual animals in sterile vacutainer tubes with heparin in the morning at day 0 and also on the last day of the experiment. Plasma was prepared after centrifugation of blood at 1000 g for 15 min at 23°C and stored at -20°C for later blood biochemical and fatty acid analyses.

Estimation of blood biochemical parameters: The haemoglobin (Hb) content was estimated by cyanomethaemoglobin method as per Drabkin (1944). Superoxide dismutase (SOD) was assayed by the method of Marklund and Marklund (1974). The activity of catalase was estimated spectrophotometrically using the method described by Aebi (1984). Total antioxidant activity was measured by ferric reducing antioxidant power (FRAP) assay of Benzie and Strain (1999). Alkaline phosphatase level in plasma was determined by p-nitrophenyl phosphatase (pNPP) method (Klin 1970) using a kit. Total immunoglobulins in the plasma sample were estimated by Zinc turbidity method (McEwan and Fisher 1970).

Slaughter method and sampling: Animals were slaughtered at 90 days of experimental period. Rumen content was taken within 15 min of slaughter and filtered through cheesecloth. An aliquot of rumen fluid was immediately stored at -20°C for fatty acid analysis. Both left and right halves of *longissimus dorsi* muscle (about 100 g) were excised within 15 min of slaughtering and were immediately immersed in liquid nitrogen for 90 sec and were then tightly wrapped in aluminium foil and stored at -80°C for subsequent analysis.

The experiment involving blood sample collection, slaughter, transport or invasive procedures on live animals was conducted according to guidelines of CPCSEA (Committee for the Purpose of Control and Supervision on Experiments on Animals, India) in order to avoid any unnecessary discomfort to the animals.

Fatty acid analysis: Rumen fluid, plasma and intramuscular fatty acids were methylated by direct transesterification method of O'Fallon *et al.* (2007). For the methyl ester formation 1.5 ml plasma, rumen fluid and 1.5 g muscle samples were taken and 0.5 µl of methyl ester sample was injected in GC. Methyl esters were separated by using a GC and equipped with a capillary column (60m × 0.25mm × 70µm-BPX70). Helium was used as carrier gas at constant inlet pressure (205 kPa). The Injector and detector temperature were 260°C and 270°C respectively and the split ratio was 1:10. The initial oven temperature was 120°C and increased by $2^\circ\text{C}/\text{min}$ to 240°C for 55 min. The identification of individual fatty acid was based on a commercial standard mixture and published isomeric profiles.

Statistical analysis: Statistical analysis of the data was

by ANOVA as per Snedecor and Cochran (1994) with the SPSS (1998). The effect of treatments on fatty acid profile was analysed by one way ANOVA, whereas, effect of treatment and period as well as their interaction on blood biochemical parameters was by two way ANOVA.

RESULTS AND DISCUSSION

Chemical composition of *T. chebula* extract: Chemical composition of feedstuffs supplemented to ration of kids during the experimental period of 90 days is presented in Table 1. Chemical composition of aqueous extract of *T. chebula* contained DM 973.8, OM 962.8, CP 26.7, EE 0.42, NDF 45.2, ADF 08.1, hemicellulose (HC) 37.1, total ash 37.2, total phenol 546.9 and total tannins 497.1 g/kg and revealed lower contents of CP, NDF and ADF whereas the extract was rich in soluble sugars and other water soluble metabolites (phenols and tannins).

Fatty acid composition of diet: Fatty acid composition of feeds offered to the kids was examined (Table 2). Maize fodder was a rich source of C18:3 (linolenic acid) (42.57%) as compared to C18:2 (linoleic acid) (16.85%) and C18:1 (oleic acid) (12.23%) whereas the concentrate mixture was rich in C18:2 (41.80%), C18:1 (35.45%) as compared to C18:3 (2.18%).

Effect of supplementation of *T. chebula* on dry matter (DM) intake and nutrient digestibility: The average DM intake (Table 3) in kids was similar ($P>0.05$) in control, T1 and T2 groups. However, the DM digestibility was increased

Table 3. Average nutrient intake of kids in different group

Particulars	Control	T1	T2	SEM
DM intake (kg/d)	0.66	0.67	0.67	0.006
DM intake (kg)/ 100 kg BW	3.05	3.03	2.96	0.034
DM intake (g)/ kg W ^{0.75}	66.01	65.91	64.67	0.659
CP intake (g/d)	111.26	112.77	112.52	1.014
ME intake (MJ/d)	6.86	6.96	6.91	0.066
Plant extract (g/kg DM intake)	0	6.0	18.0	-
Total phenols (g/kg DM intake)	0	3.2	9.8	-
Total tannins (g/kg DM intake)	0	2.9	8.9	-

SEM, Standard error of difference between means.

Table 4. Effect of the dietary supplementation of *T. chebula* extract on digestibility of nutrients (%) in different groups of kids

Particulars	Control	T1	T2	SEM
DM*	75.96 ^a	78.68 ^b	78.86 ^b	0.752
OM*	81.12 ^a	82.02 ^{ab}	82.36 ^b	0.325
CP	84.45	86.08	85.03	0.821
EE*	84.06 ^a	88.16 ^b	88.79 ^b	0.628
NDF	69.72	71.22	71.94	1.863
ADF	57.47	60.32	61.09	2.538
Hemi-cellulose	81.63	81.66	82.88	1.527

SEM, Standard error of difference between means; ^{a, b} Values with different superscripts in a row differ ($P<0.05$).

($P<0.05$) by dietary supplementation of *T. chebula* extract in treatment groups T1 and T2 (78.68, 78.86%) compared to control (75.96%; Table 4). Similarly digestibility of OM and ether extract in groups T1 (82.02 and 88.16%) and T2 (82.36 and 88.79%) was higher ($P<0.05$) than in control (81.12 and 84.06%) whereas the digestibility of CP was similar ($P<0.05$) in control, T1 and T2. The digestibility of NDF was (69.72, 71.22 and 71.94%), ADF (57.47, 60.32 and 61.09%) and HC (81.63, 81.66 and 82.88%) in control, T1 and T2 group, respectively statistically similar ($P>0.05$). Patra *et al.* (2009) found significantly higher DM and OM digestibility following the supplementation of diet with *T. chebula* (10 g/kg feed) in sheep. It was widely known that tannins affect the digestibility of nutrient either by binding digestive enzymes or feed nutrients (Min *et al.* 2005). Rai and Shukla (1977) also reported increased EE digestibility following the feeding of salseed meal (11% tannins) based diet. It was concluded that dietary supplementation of *T. chebula* extract, irrespective of its level has potential as feed additive for improving nutrient digestibility in small ruminants.

Effect of plant extract on growth performance: The daily body weight gain in respective groups was 44.05, 55.45 and 58.33 g (Table 5). The average body weight gain/ week and per day in group T2 was higher ($P<0.05$) than control group. Higher weight gain/ day ($P<0.05$) in group T2 compared to control showed better nutrient digestibility and subsequent utilization of nutrients at higher dose (1.79% DM) of *T. chebula* extract supplementation.

Table 1. Chemical composition of feed and fodder (g/kg DM basis)

Particulars	Compound feed	Maize fodder
DM	890.0	184.6
OM	928.4	908.8
CP	204.4	129.5
EE	42.2	19.4
NDF	283.0	540.9
ADF	103.1	381.0
Hemicellulose	179.9	159.9
Total Ash	71.6	91.2

Table 2. Fatty acid composition of feed and fodder (% DM basis)

Fatty acid	Compound feed	Maize fodder
C14:0	0.07	3.27
C16:0	15.59	21.92
C16:1	0.07	1.73
C18:0	5.65	11.04
C18:1	35.45	12.23
C18:2	41.80	16.85
C18:3	2.18	42.57
C20:0	0.45	0.41

C14:0 Myristic acid, C16:0 palmitic acid, C16:1 palmitoleic acid, C18:0 stearic acid, C18:1 oleic acid, C18:2 linoleic acid, C18:3 linolenic acid, C20:0 arachidic acid.

Table 5. Growth performance of kids as influenced by the dietary supplementation of *T. chebula* extract

Particulars	Control	T1	T2	SEM
Initial body weight (kg)	20.03	20.11	20.04	0.661
Final body weight (kg)	23.65	24.35	24.75	0.724
Gain in weight (g/d)	44.05 ^a	55.45 ^{ab}	58.33 ^b	2.658
Gain in weight (g/week)	308.33 ^a	391.67 ^{ab}	408.33 ^b	18.610
Feed conversion efficiency	0.06	0.07	0.08	-

^{a, b} Values with different superscripts in a row differ ($P < 0.05$).

Effect of *T. chebula* on blood biochemical parameter

Haemoglobin (Hb): The average Hb concentration on 90 d was statistically similar among different groups (Table 6).

Superoxide dismutase (SOD) and catalase activities: After 90 d of experimental feeding (Table 5), the SOD activities in treatment groups T1 and T2 supplemented with *T. chebula* extract were 3.37 and 14.27% lower ($P < 0.05$) than control group respectively (Table 6). Catalase activities in treatment groups T1 and T2 group were 0.61 and 27.45% lower ($P < 0.05$) than in the control group after 90 d of experimental feeding. These observations revealed that the dietary supplementation of *T. chebula* might increase the capability of kids to withstand the day to day physiological stresses as SOD and catalase are important free radical scavenging enzymes (Nordenson and Beckman 1981).

Plasma total antioxidant activity: Plasma total antioxidant activities i.e. FRAP value on 0 day in control and treatment groups T1 and T2 were not different among groups. However, at the end of the experimental feeding, there was a significant ($P \leq 0.05$) increase in FRAP value in group T2 than in control and T1 (Table 6).

Alkaline phosphatase activity: Alkaline phosphatase activity in plasma was similar ($P > 0.05$) at 0 day and at 90 d in control, T1 and T2, respectively (Table 6). The present

study indicated that the supplementation of *T. chebula* had no effect on liver function irrespective of its level. Reed (1995) found that higher doses of tannins are harmful for the animals and the major lesion associated with the hydrolysable tannins poisoning is necrosis of the liver. Alkaline phosphatase level is most helpful in detecting hepatobiliary disorder in small animals (Raymond *et al.* 2004).

Total immunoglobulin: The total immunoglobulin concentration at 90th day of experimental feeding was 19.72 and 24.95% higher ($P < 0.05$) in T1 and T2 group, respectively as compared to control (Table 6). On 90th d, the total immunoglobulin concentration in *T. chebula* extract supplemented kids (group T2) was increased ($P < 0.05$) than control. Thus, supplementation of *T. chebula* extract to kids had helped them to maintain the level of total immunoglobulin with advancing age due to its antioxidative property.

Animal nutritionists are currently focusing on new bio-efficient antioxidants particularly on natural antioxidants for consumer safety concerns. Restriction over the use of synthetic antioxidants like BHT and BHA in food further strengthens the concept of using naturally occurring compounds as antioxidants (Kuo *et al.* 2005). Studies have indicated that there is an inverse relationship between the dietary intake of antioxidant rich foods and the incidence of different diseases (Halliwell 1991). Although the protective effects have been primarily attributed to the well-known antioxidants such as vitamin C, vitamin E and β -carotene. Polyphenols can be an interesting choice among natural antioxidants, since they are widely distributed in plants and exhibit various antioxidant properties (Salah *et al.* 1995, Bravo 1998). Polyphenolic compounds of *T. chebula* especially gallic acid, caffeic acid and flavonoids are well documented as dietary antioxidants (Cai *et al.* 1997, Panizzi *et al.* 2002, Cheng *et al.* 2003, Bonjar, 2004, Chattopadhyay and Bhattacharyya 2007). The present study

Table 6. Influence of dietary supplementation of *T. chebula* extract on blood antioxidative and immunity parameters in kids.

Particulars	Hb (g/ L)		SOD (Units/ g Hb/ min)		Catalase (mmol of H ₂ O ₂ consumed/ min/mg Hb)		FRAP (mmol/ L)		ALP (U/ L)		Total Ig (mg/ ml)	
	0	90	0	90	0	90	0	90	0	90	0	90
Control	70.66	75.18	1721.17	2132.02 ^a	159.18	158.43 ^a	2564.15	3445.71 ^a	1111.78	1359.49	18.39	15.11 ^a
T1	73.43	74.79	1659.98	2059.96 ^b	148.47	157.46 ^a	2370.50	3470.09 ^a	1039.10	1336.44	18.33	18.09 ^{ab}
T2	73.11	68.27	1702.83	1827.64 ^a	140.68	114.94 ^b	2210.43	3776.67 ^b	1064.73	1453.22	17.88	18.88 ^b
SEM	1.154		10.533		5.039		88.152		54.714		0.431	
Treatment	0.480		<0.001		0.042		0.041		0.638		0.021	
Day	0.883		<0.001		0.567		≤ 0.001		0.001		0.340	
Treatment x Day	0.258		<0.001		0.350		0.284		0.214		0.150	

^{a, b, c} Values with different superscripts in a column differ significantly; Hb, haemoglobin; SOD, superoxide dismutase; FRAP, ferric reducing antioxidant power assay; ALP, alkaline phosphatase; Total Ig, total immunoglobulins.

Control basal diet containing roughage: concentrate = 55:45; T1, Control + *T. chebula* extract @ 6 g/ kg DM intake; T2, Control + *T. chebula* extract @ 18 g/ kg DM intake.

reports definitive results in terms of increased FRAP value. The increase in FRAP value in *T. chebula* supplemented groups, in group T2 may be attributed to the effect on production of ROS. It has also been demonstrated by Greenrod and Fenech (2003) that the main phenolic and alcoholic components of wine can reduce the DNA damaging effects of two important oxidant i.e. hydrogen peroxide and ionizing radiation. Colitti and Steafanon (2006) found that polyphenols directly affect the ROS generation. Reduced production of ROS and oxidative stress in kids supplemented with *T. chebula* may well be the reason for increased plasma total antioxidant activity in terms of FRAP value.

A positive correlation between SOD and catalase activity has been recorded in present study as reported earlier by Clemens and Waller (1987), Frei *et al.* (1994) and Kehrer and Smith (1994). These result indicated that *T.chebula*, a polyphenolic compound, acted as a potent antioxidant, besides their anti-lipid peroxidation and anti-superoxide formation activities, *T.chebula* was also investigated, for its free radical scavenging activity (Cheng *et al.* 2003). Fruit extract of *T. chebula* have been found to be effective in scavenging superoxide anions and peroxide radicals and thus have been reported to prevent the effect of free radicals on biological membrane (Subramanian *et al.* 2005). More recently Hazra *et al.* (2010) have also indicated that the fruit extracts are better hydroxyl radical scavengers than standard mannitol, with *T. chebula* being the best in comparison to *Terminalia bellerica* and *Embllica officinalis*.

A free radical scavenging enzyme SOD has been termed as an important defense system in animal body against oxygen metabolites (Nordenson and Beckman 1981). High SOD activity in control group compared to treatment groups in the present study could be attributed to an increased hydroxyl radical and hydrogen peroxide levels in the control compared to treatment groups. Sgorlon *et al.* (2006) also reported higher transcriptional activation of SOD mRNA following the administration of grape skin extract (87% polyphenols), as polyphenols in grape skin were more active in regulation of mRNA expression of detoxifying enzymes. Along with decreased SOD in treatment groups, our results indicated that dietary supplementation of *T. chebula* extract decreased the catalase activity in treatment groups as compared to control. Catalase converts hydrogen peroxide into water molecule in cytosol and thereby reduces oxidative stress. Although there was no significant change in catalase activity in T1 group, a decline in T2 group indicated that *T. chebula* supplementation helped to maintain antioxidant status and thus reduced oxidative stress in kids at different levels.

Effect of dietary supplementation of T. chebula extract on ruminal pH: The pH of rumen liquor of kids from different experimental groups prior to slaughter is presented in Table 7. Mean rumen pH in control, T1 and T2 was 6.21, 6.25 and 6.20, respectively. Statistical analysis of data reveal that supplementation of aqueous extract of *T. chebula* at different levels had no effect on ruminal pH. Priolo *et al.*

Table 7. Influence of dietary supplementation of *T. chebula* on ruminal pH of kids

Animal No.	Control	T1	T2	SEM
1	6.44	6.15	6.18	0.075
2	6.03	6.30	6.26	0.069
3	6.38	6.12	6.25	0.061
4	6.17	6.32	6.15	0.044
5	6.04	6.29	6.25	0.063
6	6.18	6.30	6.10	0.047
Mean $\pm$ SE	6.21	6.25	6.20	0.012

(2000), Vasta *et al.* (2009) and Hervas *et al.* (2004) also did not observe any effect of supplementation of polyphenols enriched extract or polyphenols on the ruminal pH.

Effect on fatty acid profiles in rumen fluid, blood plasma and muscle of kids fed T. chebula: Saturated fatty acid content in rumen fluid of kids in T1 and T2 group was lowered ($P<0.01$) by 20.43% and 22.54% as compared to control (Table 8). The monounsaturated fatty acid (MUFA) in group T1 and T2 was 6.12% and 16.14% ($P<0.01$) higher as compared to control. The level of PUFA in rumen fluid was not influenced by the dietary supplementation of *T. chebula* extract. There was no effect of treatments on the ruminal pH.

Plasma SFA content (g/100 g of FAME) was lower ($P<0.01$) in group T2 (39.61) than control (48.38) and T1 (46.29) groups which indicates the dose dependent effect of *T. chebula* on the plasma saturated fatty acids. Concentration of MUFA in T1 and PUFA in T2 group were higher ($P<0.01$) than in the control group.

In muscle, the total SFA level in group T2 (37.94 g/100 g of FAME) was lower ($P<0.01$) than control, however, it was comparable in control and T1. The total MUFA in groups T1 and T2 were 20.25 and 24.97 % higher ($P<0.01$) than control thereby improving the desaturation index (Σ MUFA/ Σ SFA) in group T2 (0.73 ± 0.038) compared to control ($0.47^a \pm 0.01$). The level of PUFA was higher ($P<0.05$) in group T2 than control.

Increase in PUFA concentration and decreased SFA content in *T. chebula* extract supplemented groups as indicated by fatty acid profile of blood and muscle along with increase in antioxidant activity had an immuno-modulating effect in kids as can be inferred from the significant increase in the total immunoglobulin concentration. Additionally, oxidative damage to the cellular membrane can compromise the integrity of the immune cells (T and B-lymphocytes) and immune cells are more sensitive to oxidative damage because their cell membrane contains high concentration of PUFA which are more susceptible to lipid peroxidation and immune cells also produce large amounts of ROS when stimulated (Dayong and Simin 1999). Results from the present study further indicated that the polyphenols present in *T. chebula* extract could be responsible for its pharmacological activity in kids. There have been reports on antioxidant properties of the plant extract however, our study provides a definitive report about

Table 8. Effect of *T. chebula* extracts supplementation on fatty acid profile (g/100 g of FAME) of kids

Particulars	Fatty acid	Control	T1	T2	SEM
Rumen liquor	Σ SFA	49.72 ^d	39.56 ^c	38.51 ^c	1.978
	Σ MUFA	8.49 ^c	9.01 ^{cd}	9.86 ^d	0.214
	Σ PUFA	3.18	3.99	4.38	0.387
Plasma	Σ SFA	48.38 ^d	46.29 ^d	39.61 ^c	1.421
	Σ MUFA	23.15 ^c	36.79 ^d	32.40 ^{cd}	2.347
	Σ PUFA	21.62 ^c	23.71 ^{cd}	27.78 ^d	1.038
<i>Longissimus dorsi</i> muscle	Σ SFA	47.22 ^d	43.93 ^{cd}	37.94 ^c	1.509
	Σ MUFA	22.27 ^c	26.78 ^d	27.83 ^d	0.953
	Σ PUFA	4.40 ^c	5.56 ^{cd}	5.93 ^d	0.308
	ΣMUFA /ΣSFA	0.47 ^a	0.61 ^{ab}	0.73 ^b	0.026

^{a, b} Values with different superscripts in a row differ (P<0.05). ^{c, d} Values with different superscripts in a row differ (P<0.01).

the correlation between free radical scavenging capacity and antioxidant activity of *T. chebula*.

Enhancement of UFA levels in muscle of kids by supplementation of aqueous extract of *T. chebula* at higher dose may be due to the presence of bioactive compound(s) polyphenols (Min *et al.* 2005) which influence the rumen biohydrogenation and fatty acid composition in muscles of kids (Vasta *et al.* 2009, 2010, Rana *et al.* 2011). It can also be inferred from the present investigation that *T. chebula* extract increased total UFA content by altering rumen biohydrogenation and protect this UFA in plasma and tissues from lipid peroxidation with enhanced enzymatic/non-enzymatic antioxidative activity and free radical scavenging capacity, which can ultimately result in increased nutraceutical value of the animal product.

In conclusion, supplementation of aqueous extract of *T. chebula* @ 18 g/kg DM in the diet of kids increased the immunity and antioxidative status. Polyphenols/ tannins influenced the fatty acid biohydrogenation thereby enhanced the MUFA (24.97%), PUFA (34.77%) whereas SFA (19.65%) were lowered in muscle. It is evident from the present experiment that the supplementation of polyphenols enriched extract of *T. chebula* preserves the health of ruminants and increase the nutraceutical and therapeutic value of the meat. Furthermore, evaluation of *in vivo* antioxidant activity of this plant extract has also provided interesting results that might be beneficial for the pharmacological use of this plant in clinical trials. However, detailed clinical work and pharmacological evidences, at molecular level, are required to establish the possible correlation among the mentioned activities of the extract.

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