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Effect of silymarin and selenium-yeast supplementation on liver functions of Barbari goat during cold stress

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Abstract

The aim of the present study was to assess the effect of silymarin (SM) and selenium-yeast (SY) on renal and hepatic functions of Barbari goat during cold stress. Thirty two Barbari goats (age: 6-8 months) were selected from ILFC- II, SVPUAT, Meerut, Uttar Pradesh, India. The animals were randomly divided into four groups (n=8) according to body weight and age. First group (C) was kept as control group giving only basal diet, second group (T_1) supplemented with 600 mg Silymarin/kg DM/day, third group (T_2) supplemented with 0.3 mg Selenium as Se-Yeast/kg DM/day and fourth group (T_3) supplemented with 600 mg Silymarin + 0.3 mg Selenium as Se-Yeast/kg DM/day along with basal diet for 90 days of experimental periods. Supplementation significantly ($p<0.05$) influenced the level of plasma ALT, AST and ALP in the animals. The mean activity of plasma ALT, AST and ALP was found significantly ($p<0.05$) lower in T_1, T_2 and T_3 groups as compare to control group. Mean daily weight gain (DWG) of the supplemented group was found significantly ($p<0.01$) more than control group. Dry matter intake (DMI) was found significantly ($p<0.01$) higher in supplemented group as compare to control group. The result of this study indicated that supplementation of SM alone and in combination with SY improves liver function by reducing liver enzymes and increasing feed intake in the Barbari goats during cold stress.

Keywords: ALT, AST, ALP, Barbari goat, cold stress

Introduction

Goat rearing is a traditional occupation of small, marginal farmers and landless laborers in the developing countries. It is well suited for effective utilization in the diverse socio-economic situations and livelihoods of the households in the developing countries. It has significant contribution in the economy, food security and poverty alleviation of the rural areas and provides income and insurance against emergency conditions (Al-Atiyat, 2014) [3]. On economic ground goat production performance is used as indicator for the livelihood of the farmers (Al Baqain and Valle Zárate, 2011) [2].

Silymarin (SM) is a mixture of flavonolignans obtained from milk thistle (*Silybum marianum*), it has very strong antioxidant effects (Attia *et al.*, 2019) [4]. The main flavonolignans in silymarin is silybin (50%), sily-chrysanthemum (20%), silydianin (10%) and isosilibine (5%) (Kordi *et al.*, 2013) [11]. Silydianin levels are found maximum in stem and seed of the plant (Shaker *et al.*, 2010) [14]. In spite of antioxidant action, SM causes stimulation of DNA polymerase, stabilization of cell membranes, inhibition of free radicals, and increased cellular glutathione concentration. SM has been used to treat hepatic disease, including hepatotoxic reactions in people and animals. It has also some pharmacological role in animals as antioxidant, hepatoprotectant, anti-inflammatory, antibacterial and antiviral (Abascal, 2003) [1].

Selenium (Se) is an essential trace element, and also an integral component of several antioxidant enzymes such as glutathione peroxidase. Se exerts its biological function when incorporated into proteins as selenocysteine residues (Chauhan *et al.*, 2014) [7]. Selenium-yeast (SY) is an organic form of Se having good bioavailability.

Materials and Methods

The experiment was conducted at ILFC- II, SVPUAT, Meerut. In summer, highest temperature rises up to 45° C and in winter there is remarkable fall of temperature sometimes up to freezing point. The average annual rainfall is about 862.7 mm and the annual relative humidity varies from 67 to 83 percent.

Experimental protocol was approved by under Institutional Animal Ethics Committee (IAEC), constituted under control of the committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA) rules laid down by the Government of India (Certificate no. IAEC/SVPUAT/2022/102).

Experimental animal

The study was conducted in 32 Barbari goats of six to eight month age group of the herd maintained at ILFC- II, Sardar SVPUAT, Meerut, Uttar Pradesh, India. Experimental animals were randomly divided into four groups (n=8) according to age and body weight (age: 6-8 months). First group (C) was kept as control group giving only basal diet, second group (T_1) supplemented with 600 mg SM/kg DM/day, third group (T_2) supplemented with 0.3 mg Se as SY/kg DM/day and fourth group (T_3) supplemented with 600 mg SM + 0.3 mg Se as SY/kg DM/day along with basal diet for 90 days of experimental periods. SY (purity 80%) was purchased from Alenit Chemicals LLP, Delhi. SY (2000 ppm) was provided by Chaitanya Agro-Biotech. Pvt. Ltd., 120/2 Laxmi Nagar, Malkapur, Buldana (Maharashtra) India. The computed and weighed doses of SM, SY alone or both mixed in small amount of concentrate and fed individually to each animal for 90 days of study period. Clean and fresh tap water was offered *ad-lib*. Total duration of the experiment was 90 days. Body weight (BW) and feed consumption was recorded before start of experiment and thereafter repeated at fortnightly intervals with digital electronic balance.

Blood sampling and enzyme estimation

About 5 ml blood was collected in the EDTA coated Vacutainer tube before feeding and watering aseptically from jugular vein with the help of sterilized needles on day schedule of 0, 15, 30, 45, 60, 75 and 90 day. The blood samples centrifuged at 3000 rpm for 30 min for plasma separation. The plasma was stored at -20°C and further analyzed for liver enzymes. The concentrations of plasma alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) were measured by using kits from ERBA diagnostics Mannheim, Germany.

Statistical Analysis

Data was analyzed using MIXED Models of statistical software package SPSS version 20 (SPSS for Windows, V 20.0; SPSS, Inc., Chicago, IL, USA). The model was used as follows:

$$Yij = \mu + Ti + Fj + (T \times F)ij + eij$$

Where, Yij = dependent variable; μ = overall mean of a population; Ti = effect of the treatment (SM and SY) (i = 1….4); Fj = effect of days (j = 1….7); (T × F) ij = effect of the interaction between effect of treatments and days; eij = random error.

Results and Discussion

Monthly maximum and minimum AT and THI of 90 d of the experimental period during winter season are presented in the Fig 1 (A and B).

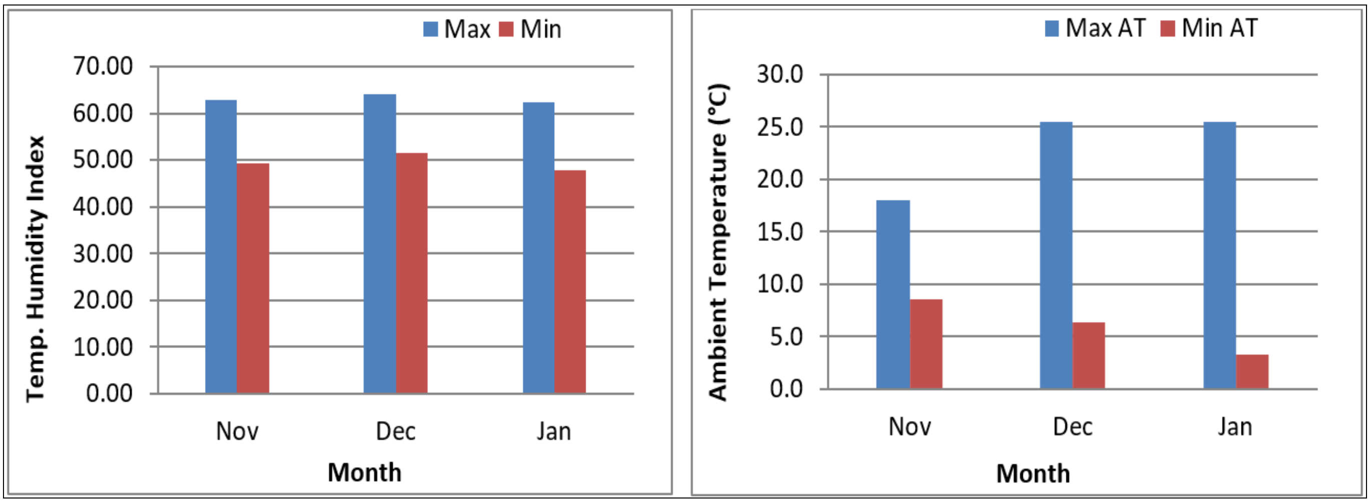

Fig 1: Monthly maximum and minimum (A) ambient temperature (AT) and (B) temperature humidity index (THI) during experimental period in winter season

Feed intake and Growth

The result of growth performance; daily weight gain (DWG), DMI (%) and Feed conversion efficiency (FCE) of Barbari goat during winter season of the experimental period are presented in the Table 1. Supplementation significantly (p<0.05) influenced the level of DWG, DMI and FCE of the animals. The mean values of DWG were significantly (p<0.05) greater in T_1, T_2 and T_3 groups as compare to control group. The goats receiving either SM (600 mg/kg) or Se as SY (0.3 mg/kg) or both showed higher (p<0.05) DWG on 75 and 90 d of the experiment than the control. The mean values of fortnightly DMI (%) were significantly (p<0.05) greater in T_1, T_2 and T_3 groups as compare to control group. The goats receiving either SM (600 mg/kg) or Se as SY (0.3 mg/kg) showed higher (p<0.05) DMI (%) on 45, 60, 75 and 90 d of study than the control group. The mean values of DMI (%) were statistically (p<0.05) vary among the treatment groups. Feed conversion efficiency was found significantly (p<0.05) higher in T_1, T_2 and T_3 groups as compare to control group.

Similar findings on feed intake and body weight gain were also reported by Belvins *et al.* (2010) [5] in broilers supplemented with 1000 mg/kg SM and Wang *et al.* (2019) [18] in fishes supplemented with different doges of SM in feed. In contrast to the present findings, Schiavone *et al.* (2007) [13] reported that BW, DMI and FCE were not affected with supplementation of 40 and 80 mg/kg SM in chickens. Similarly, the growth-promoting effect of SM was also observed by some other researchers Singh (2022) [15] in Murrah buffalo and Dubey (2022) [8] in Sahiwal calves which may be attributed to its hepatoprotective properties.

Table 1: Effect of SM and SY supplementation on DWG, DMI and FCE in cold stressed Barbari goat

Variable	Days	Treatment				SEM	P- value		
		C	T_1	T_2	T_3		T	D	TxD
Daily Weight Gain (g/d)	15	25.00	37.58	33.83	46.75	3.80			
	30	34.25	48.00	44.50	47.25	4.39			
	45	38.33	52.83	48.83	52.67	4.16			
	60	53.42	57.17	49.42	64.17	7.68			
	75	42.33^{a}	67.92^{b}	55.67^{b}	68.67^{b}	5.12			
	90	52.50^{a}	65.75^{b}	62.00^{b}	97.00^{c}	5.44			
	Mean	40.97^{a}	54.88^{b}	49.04^{b}	62.75^{b}	5.10	0.000	0.001	0.105
DMI (%)	15	2.34	2.41	2.39	2.47	0.03			
	30	2.37	2.48	2.44	2.57	0.03			
	45	2.42^{a}	2.55^{ab}	2.51^{ab}	2.61^{b}	0.03			
	60	2.39^{a}	2.63^{bc}	2.54^{b}	2.69^{c}	0.03			
	75	2.47^{a}	2.68^{b}	2.61^{ab}	2.80^{c}	0.04			
	90	2.55^{a}	2.78^{b}	2.69^{b}	2.92^{c}	0.03			
	Mean	2.42^{a}	2.59^{c}	2.53^{b}	2.68^{d}	0.03	0.000	0.000	0.081
Feed conversion efficiency (FCE)	15	0.12	0.16	0.16	0.20	0.03			
	30	0.16	0.21	0.20	0.20	0.03			
	45	0.15	0.19	0.18	0.18	0.02			
	60	0.20	0.18	0.16	0.20	0.03			
	75	0.14	0.19	0.17	0.18	0.02			
	90	0.16	0.17	0.17	0.23	0.02			
	Mean	0.15^{a}	0.18^{b}	0.17^{b}	0.20^{b}	0.02	0.002	.000	0.231

C, control; T1, silymarin (600 mg/kg DM/d) supplemented group; T2, selenium-yeast (0.3 mg/kg DM/d) supplemented group; T3, silymarin (600 mg/kg DM/d) and selenium-yeast (0.3 mg/kg DM/d) supplemented group; SEM, Standard error mean; T, effect of treatment, D, effect of day; T × D, interaction between treatment and day

$^{a, b, c}$Mean bearing different superscripts in a row showed a statistical difference at $p<0.05$

DWG, Daily Weight Gain; DMI, Dry Matter Intake (%); FCE, Feed Conversion Efficiency

Liver functions

The effects of SM and SY supplementation on activity of liver enzymes; ALT, AST and ALP of Barbari goat during 90 days of study period are presented in the Table 2. Supplementation significantly ($p<0.05$) influenced the level of plasma ALT, AST and ALP in the animals. The mean activity of plasma ALT was found significantly ($p<0.05$) lower in T_1, T_2 and T_3 groups as compare to control group. Further, goats receiving either SM (600 mg/kg) alone or in combination with Se as SY (0.3 mg/kg) exhibited statistically lower ($p<0.05$) plasma ALT activity on 45, 60 and 90 d of experiment than the control group. Also there was found significant ($p<0.05$) variation in the mean values of plasma AST activity among the groups. The mean values of AST activity were found significantly ($p<0.05$) lower in T_1, T_2 and T_3 groups as compare to control group. The level of AST was found significantly ($p<0.05$) lowest in T_3 group followed by T_1, T_2 and control group on 30, 45, 60 and 75 d of the study. Further, the mean activity of plasma ALP was found significantly ($p<0.05$) lower in T_1, T_2 and T_3 groups as compare to control group. The overall mean value of ALP was 73.58, 67.15, 67.97 and 65.09 in control, T_1, T_2 and T_3 (IU/L) groups respectively.

Guo *et al.* (2016) [10] reported SM has been effective in reducing blood ALP levels and reduced liver damages in mice. Long term use of SM (120 mg twice daily for 60 d) significantly reduced serum ALT and AST in liver patients (Pares *et al.*, 1998) [12]. It has also role in prevention of the peroxidation processes in liver lesions caused by any drug or toxic substances (Vargas-Mendoza *et al.*, 2014) [17]. SM has been used in the treatment of liver diseases, including alcoholic liver disease, chronic viral and toxic hepatitis (Flora *et al.*, 1998) [9]. Silymarin affects the stability of the liver membrane and prevents the binding of many toxins and drugs to this membrane.

SM stimulates activity of RNA polymerase I enzymes in the nucleus of hepatocytes which increases the production of ribosomes and accelerates the process of protein synthesis (Wang *et al.*, 1996; Sonnenbichler, 1986) [19, 16]. Further, more active structural and functional protein synthesis occurred in the liver which increased rejuvenation capacity of the liver (Blumenthal *et al.*, 2000) [6].

Table 2: Effect of silymarin and selenium-yeast supplementation on ALT, AST and ALP activity of Barbari goat during winter season

Variable	Days	Treatment				SEM	P- value		
		C	T_1	T_2	T_3		T	D	TxD
ALT (IU/L)	0	17.68	17.02	17.90	16.80	1.49			
	15	19.67	18.23	17.79	18.67	1.63			
	30	21.61	18.67	18.34	18.23	1.17			
	45	21.42^{b}	19.12^{ab}	18.01^{a}	17.46^{a}	1.37			
	60	23.10^{b}	18.56^{a}	19.01^{a}	20.22^{ab}	1.24			
	75	23.02	20.11	21.33	19.12	1.32			
	90	23.20^{b}	19.45^{a}	21.00^{ab}	18.67^{a}	1.62			
	Mean	21.39^{b}	18.74^{a}	19.05^{a}	18.45^{a}	1.40	0.001	0.008	0.988
AST (IU/L)	0	44.42	42.98	43.21	43.65	1.81			
	15	45.47	42.54	44.53	40.55	2.61			
	30	50.36^{b}	43.98^{ab}	44.97^{ab}	40.33^{a}	3.08			
	45	48.71^{b}	40.55^{a}	40.77^{a}	41.33^{a}	3.35			
	60	46.12^{b}	39.23^{a}	38.45^{a}	38.56^{a}	1.66			
	75	44.30^{b}	38.12^{a}	39.45^{ab}	37.79^{a}	2.00			
	90	42.10	39.01	41.55	36.13	1.75			
	Mean	45.93^{b}	40.92^{a}	41.85^{a}	39.76^{a}	2.32	0.000	0.005	0.914
ALP (IU/L)	0	74.61	73.76	74.61	72.40	6.15			
	15	77.06	71.22	72.40	65.96	4.76			
	30	75.40	70.20	68.16	67.32	4.45			
	45	74.29^{b}	63.08^{a}	62.91^{a}	64.26^{a}	4.55			
	60	69.86	62.23	65.96	61.38	2.79			
	75	72.45^{b}	63.76^{ab}	64.94^{ab}	60.36^{a}	2.69			
	90	71.39	65.79	66.81	63.93	3.37			
	Mean	73.58^{b}	67.15^{a}	67.97^{ab}	65.09^{a}	4.11	0.003	0.022	1.000

C, control; T1, silymarin (600 mg/kg DM/d) supplemented group; T2, selenium-yeast (0.3 mg/kg DM/d) supplemented group; T3, silymarin (600 mg/kg DM/d) and selenium-yeast (0.3 mg/kg DM/d) supplemented group; SEM, Standard error mean; T, effect of treatment, D, effect of day; T × D, interaction between treatment and day

[a, b, c]Mean bearing different superscripts in a row showed a statistical difference at $p<0.05$

ALT, Alanine aminotransferase; AST, Aspartate aminotransferase; ALP, Alkaline Phosphatase

Conclusions

The result of the study indicated that supplementation of silymarin and selenium-yeast improves liver function by reducing liver enzymes, increasing feed intake, and growth performance of the Barbari goats during cold stress.

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Conflict of Interest

The authors declare that there is no conflict of interest for this manuscript.

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