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Effect of dietary supplementation of silymarin and nano-zinc on protein metabolism of Sahiwal calves

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Abstract
This study assessed the effect of dietary supplementation of silymarin and nano-zinc on protein metabolism of Sahiwal Calves. Twenty-eight 6-15 months old Sahiwal calves were randomly allocated to four groups and fed a basal diet without any supplementation (control) or supplemented respectively with silymarin (500 mg/kg DM, T1), nano-zinc (40 mg/kg DM. T2), and both silymarin and nano-zinc (500 mg/kg DM + 40 mg/kg DM, T3) for 120 days of trial period. Blood was sampled on 0, 30, 60, 90, and 120 days of trial period for the analysis of protein metabolites. The mean protein level was significantly (P<0.05) higher in treatments than control groups but the level was observed highest statistically (P<0.05) in treatment that offered both silymarin and nano-zinc in the diet. However, no statistical difference in protein level was observed between T1 and T2. The mean plasma albumin and albumin concentration on 0, 30, 60, 90, and 120 days were statistically similar in each group. The mean plasma concentration of globulin was statistically (P<0.05) enhanced in the T3 group which is supplemented by silymarin and nano-zinc as compared to all other groups. The mean creatinine concentration was statistically (P<0.05) higher in the T2 and T3 as compared to control and T1 groups and no statistical difference observed between control and T1 groups. The mean plasma urea nitrogen and plasma urea nitrogen on all days of the study were statistical similar among groups, Result of the present study is concluded that dietary supplementation of silymarin and nano-zinc improved the function of kidney and immunity of Sahiwal growing calves.

Keywords: Silymarin, nano-zinc, sahiwal calves, protein metabolites

Introduction
Sahiwal is one of the best milch cattle breed in total of the 50 descriptive indigenous cattle breeds of India, (NBAGR, Karnal 2020). Cattle milk production contributes 48.71% out of total milk production of India which makes it highest milk producer country in the world. Sahiwal breed population is 5.99 million which contributes 4.2% out of total indigenous cattle of India (Anonymous, 2019) [1]. Silymarin is flavonoide mixture extracted by the 'milk thistle (*Silybum marianum*) plant, which belongs to the Asteraceae family from Tracheophyta Phylum. Silidianin, isosilibinin, Silibinin and Silichristin are 4 isomers of Silymarin compound, (Lee *et al*., 2003) [7]. It was used to from ancient times in Egypt and Europe as a traditional medicinal plant for jaundice disease treatment, due to its protecting, detoxifying, and regenerating property for the beneficial cells of gallbladder, spleen, and the entire body (Denev *et al* 2020, Karayiannis, 2003) [2, 4]. It is a good antioxidant, eliminating free radicals directly and activities of enzymes involved in ROS generation (Yin *et al*., 2011) [19]. Silymarin supplementation effect in periparturient dairy cow metabolism shows the most potential use of it, and satisfied that it is a safe natural product, which plays significant role in the metabolic adaptation of dairy cows at the initial stage of lactation period. Supplementation of these substances shows a good BCS (body condition ratio), a higher and earlier milk peak, and no negative conditions revealed by the use of this product. Blood and milk parameters observation also gives out not any adverse effects of feeding cows of this natural compound (Tedesco *et al.*, 2004) [17]. Zinc is also beneficial for enhancing the growth and development of animals. Zinc is an important trace element for many physiological activities which influences growth, health, and reproduction by various types of metabolic happening. Zinc is required in nucleic acid metabolism, carbohydrate metabolism, and protein synthesis (Hidayat *et al.*, 2018) [3]. Zinc also act as a helping content for the carbohydrate metabolites by promoting the glucose transportation, assists the conversion of glucose into lipids, and glucose oxidation by glycolysis and hexose monophosphate shunt pathways (Zaboli *et al.*, 2013) [20].

Supplementation of zinc in animals diet is gives positive effect on growth and immune status (Singh *et al.*, 2018) [15]. Conversion in nanoparticles from micro particles (diameter range under 100nm) helps to increase the surface area for easily absorbance and facilitates greater solubility which responses best utilization for animals (Zaboli *et al.*, 2013) [20]. The objective of this study was to assess the protein metabolites in Sahiwal calves fed diet containing silymarin and nano-zinc.

Materials and Methods

The procedure and protocol of this trial was sanctioned by Institutional Animal Ethics Committee working under the control of the CPCSEA rules laid down by the Government of India.

The present trial was performed at the Livestock Research Centre, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut (UP). For this trial 28 Sahiwal calves were sorted out according to weight (82±0.50 kg) and age (6-15 months) and randomly assigned into 4 groups (n = 7). The basal diet contained concentrate, green fodder, and straw in the ratio of 48:32:20, respectively and was prepared as per suggestion of NRC (2001). Basal diet offered to animals two time in a day on 08.00 am and 06.30 pm without any supplementation (control) or supplemented with silymarin (500 mg/kg DM, T1), nano-zinc (50 mg/kg DM, T2) or both silymarin and nano-zinc (500 mg/kg DM + 50 mg/kg DM, T3) for 120 days of the experimental period. The accurate doses of silymarin and nano-zinc calculated and weighed and mixed in little quantity of concentrate mixture and fed to animals according to their body weight of concern groups individually. Fresh and clean tap water was to the animals *ad-libitum*.

Blood was drawn from the jugular vein puncture in EDTA containing vials on 7.00 am at 30 days interval (0, 30, 60, 90, and 120) during whole trial period. After blood collection at farm, blood containing vials kept up in ice boxes and then carried to laboratory for further analysis. Blood samples centrifuged at 3000 rpm for 30 minutes at 4 ºC temperature. After centrifugation the blood plasma was harvested in Eppendorf tubes and stored these tubes at -20 ºC for the further analysis of total protein, albumin, creatinine, and blood urea nitrogen. The plasma samples were analyzed for total protein, albumin, creatinine, and plasma urea nitrogen using commercial ERBA kits (ERBA diagnostics PvT. Ltd., Mannheim Germany). Globulin content in the blood plasma was calculated by the arithmetic mean difference of total protein and albumin concentration.

The all data by different parameters collected during experiments was analyzed with help of MIXED model of SPSS software V 19.0. The pair wise comparison of means at monthly intervals were performed by the 'Duncan's Multiple Range Test'.

Results

Table 1: Effect of silymarin and nano-zinc supplementation on total protein and albumin of sahiwal calves.

Variable	Days	Treatment				SEM	P- Value		
		Control	T1	T2	T3		T	D	T×D
Total protein (g/100 ml)	0	4.38	4.44	4.37	4.41	0.12			
	30	4.50^{a}	4.58^{a}	4.75^{a}	5.46^{b}	0.12			
	60	4.49^{a}	4.87^{ab}	5.02^{b}	5.60^{c}	0.12			
	90	4.50^{a}	5.41^{b}	5.40^{b}	5.90^{b}	0.16			
	120	4.62^{a}	5.81^{b}	6.11^{b}	6.16^{b}	0.12			
	Mean	4.50^{a}	5.02^{b}	5.13^{b}	5.51^{c}	0.13	.000	.000	.000
Albumin (g/100 ml)	0	3.26	3.26	3.36	3.27	0.05			
	30	3.40	3.19	3.40	3.26	0.05			
	60	3.27	3.23	3.20	3.18	0.03			
	90	3.31	3.31	3.23	3.27	0.07			
	120	3.32	3.45	3.34	3.23	0.05			
	Mean	3.31	3.29	3.31	3.24	0.05	.181	.048	.140

T_1, silymarin (500 mg/kg DM/d) supplemented group; T_2, nano-zinc (40 mg/kg DM/d) supplemented group; T_3, silymarin (500 mg/kg DM/d) and nano-zinc (40 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 $P<0.05$

The result of the total protein of all groups of Sahiwal calves has been presented in Table 1.The mean protein and protein concentration on 30 and 60 days of the trial were significantly ($P<0.05$) higher in T_3 as compared to the control, T_1, and T_2 group, however, no statistical difference was reported between T_1 and T_2 groups. On day 0 of the study, the plasma concentration of total protein was statistically similar in all four groups.

The influence of silymarin and nano-zinc supplementation on albumin is given in Table 1. The albumin concentration was statistically similar in all four groups during 0, 30, 60, 90, and 120 days of the trial period. Likewise, the mean value of albumin was also not varied statistically among the groups.

The impact of silymarin and nano-zinc supplementation on globulin of Sahiwal calves is depicted in Table 2. The mean value of globulin and globulin concentration on day 60 were statistically ($P<0.05$) higher in the group that received both supplemental silymarin and nano-zinc as compared to all other groups, though, there was no statistical difference reported between silymarin and nano-zinc supplemented groups.

Table 2: Effect of silymarin and nano-zinc supplementation on globulin, creatinine, and plasma urea nitrogen (PUN) of Sahiwal calves.

Variable	Days	Treatment				SEM	P- Value		
		Control	T1	T2	T3		T	D	T×D
Globulin (g/100 ml)	0	1.12	1.18	1.01	1.15	0.14			
	30	1.10^{a}	1.39^{a}	1.35^{a}	2.20^{b}	0.14			
	60	1.23^{a}	1.64^{b}	1.82^{b}	2.42^{c}	0.11			
	90	1.18^{a}	2.10^{b}	2.17^{b}	2.63^{b}	0.18			
	120	1.30^{a}	2.36^{b}	2.77^{bc}	2.93^{c}	0.13			
	Mean	1.19^{a}	1.73^{b}	1.82^{b}	2.27^{c}	0.14	.000	.000	.000
Creatinine (mg/100 ml)	0	1.85	1.86	1.85	1.81	0.05			
	30	1.96^{b}	1.83^{b}	1.73^{b}	1.43^{a}	0.08			
	60	2.01^{b}	1.83^{ab}	1.73^{ab}	1.42^{a}	0.09			
	90	2.03^{b}	1.88^{b}	1.47^{a}	1.35^{a}	0.08			
	120	2.01^{c}	1.92^{c}	1.66^{b}	1.43^{a}	0.05			
	Mean	1.97^{c}	1.87^{c}	1.69^{b}	1.49^{a}	0.07	.000	.107	.017
Plasma urea nitrogen (SUN) (mM)	0	5.22	5.27	5.25	5.29	0.15			
	30	5.21	5.22	5.13	5.06	0.1			
	60	5.45	5.10	5.09	4.99	0.12			
	90	5.02	5.06	5.14	5.07	0.14			
	120	4.97	4.97	5.06	5.06	0.13			
	Mean	5.17	5.12	5.13	5.09	0.13	.838	.125	.808

T_1, silymarin (500 mg/kg DM/d) supplemented group; T_2, nano-zinc (40 mg/kg DM/d) supplemented group; T_3, silymarin (500 mg/kg DM/d) and nano-zinc (40 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 $P<0.05$

The value of creatinine in all four groups over a 120 days of study period is presented in Table 2. The mean concentration of creatinine showed a significant ($P<0.05$) difference among the groups and was reported the 1.97, 1.87, 1.69, and 1.49 in the control, T_1, T_2, and T_3 groups, respectively. Similarly, the value of creatinine on day 120 of the trial period was declined significantly ($P<0.05$) in groups that received either nano-zinc or both silymarin and nano-zinc as compared to control and group that received only silymarin.

The effect of silymarin and nano-zinc supplementation on plasma urea nitrogen of sahiwal calves has been shown in Table 2. The mean plasma urea nitrogen and urea nitrogen on all month of study period were statistically similar among the groups.

Discussion

In corroboration with the present finding, Sahad *et al.*, (2020) observed silymarin supplementation in the form of milk thistle seed powder @1% of feed in quail that increase the total protein and globulin, however, no effect was observed on the albumin level. Schiavone *et al.*, (2007) [12] also reported higher protein levels in broilers that received 40 to 80 ppm silymarin in their diet. Dockalova *et al.*, (2021) reported lower creatine level in sports horse fed diet containing silymarin at the rate of 100g and 400g/day, however, no effect observed on PUN and albumin. Moreover, Ulger *et al.*, (2017) [18] noticed a significant increase in the total protein concentration of silymarin supplemented (20 g/animal/day) dairy cows. Kumar *et al.*, (2018) [6] reported that 40 ppm zinc supplementation to the murrah buffalo calves increase the total protein, globulin and albumin significantly ($P<0.05$). Swain *et al.*, (2019) observed higher protein and globulin level in the blood of nano-zinc fed goat. Similar to present findings, Nagalakshmi *et al*., (2009) [9] reported 30 ppm zinc either inorganic or organic sources supplemented in lambs improved globulin levels. Khaleghipour *et al.*, (2020) [5] was observed that silymarin extract supplementation in Japanese quals at dose 500 mg/day had negative effect on blood urea nitrogen and had no effect on creatinine concentration. No significant effect of nano-zinc (50 ppm) supplementation was observed on creatinine level of goat (Swain *et al.*, 2019). In contrast to the present finding, Mishra *et al.*, (2014) [8] observed no positive effect nano zinc (30 ppm) on total protein albumin globulin and plasma urea nitrogen of layer chicks. Parashuramulu *et al.*, (2015) [11] showed that, 80 and 140 ppm zinc supplementation to Murrah buffalo calves resulted non-significant effect for the total protein, albumin, and globulin. Sethy *et al.*, (2016) [13] reported no effect of zinc supplementation on total protein, albumin, globulin and blood urea nitrogen of black Bengal kids.

The results showed that supplementation of silymarin and nano-zinc caused an increase in total protein and globulin, however, a decrease in creatinine concentrations of sahiwal calves. Therefore, supplementation of silymarin and nano-zinc might be improved immunity and kidney function of Sahiwal calves.

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References

1. Anonymous, 20th Livestock census 2019. Department of Animal Husbandry, Dairying and Fisheries, Govt. of India, New Delhi, India, 2019.
2. Denev P, Ognyanov MH, Georgiev YN, Teneva DG, Klisurova DI, Yanakieva IZ. Chemical composition and antioxidant activity of partially defatted milk thistle (*Silybum marianum* L.) seeds. Bulgarian Chemical Communications. 2020;1:182-187.
3. Hidayat C, Jayanegara A, Wina E. Supplementation of dietry nano zn phytogenic on performance antioxidant activity and population of intestinal pathogenic bacteria in broilers chiks. Tropical animal science journal. 2018;44:90-99.
4. Karayiannis P, Hepatitis B. virus: old, new and future

approaches to antiviral treatment. Journal in antimicrobial research. 2003;5:761-785.
5. Khaleghipoura B, Khosraviniaa H, Toghiyanib M, Azarfar A. Efficacy of silymarin-nanohydrogle complex in attenuation of aflatoxins toxicity in Japanese quails. Italian Journal of Animal Science. 2020;19:351-359.
6. Kumar A, Sahu DS, Chandra G, Yadav SP, Kumar R, Jaisawal, *et al.* Effect of different sources of zinc on growth performance and haemato-biochemical profiles of Murah buffalo calves. The Indian Journal of Animal Nutrition. 2018;35:409-414.
7. Lee DY, Liu Y. Molecular structure and stereochemistry of silybin A, silybin B, isosilybin A and isosilybin B, isolated from *Silybum marianum* (milk thistle). Journal of Natural Products. 2003;66:1171-1174.
8. Mishra A, Swain RK, Mishra SK, Panda N, Sethy K. Growth Performance and Serum Biochemical Parameters as Affected by Nano Zinc Supplementation in Layer. Chicks The Indian Journal of Animal Nutrition. 2014;31:384-388.
9. Nagalakshmi D, Dhanalakshmi K, Himabindu D. Effect of dose and source of supplemental zinc on immune response and oxidative enzymes in lambs. Veterinary Research Communications. 2009;33:631-644.
10. NBGAR. Karnal, ICAR$_S$ Breed Registration Committee Approved 7 new breeds of Cattle on 24th Jan 2020 at New Delhi.
11. Parashuramulu S, Nagalakshmi D, Kumar MK. Effect of zinc supplementation on haematology and serum biochemical constituents in Murrah buffalo calves. Indian Journal of Animal Research. 2015;49:482-486.
12. Schiavone A, Righi F, Quarantelli A, Bruni R, Serventi P, Fusari A. Use of *Silybum marianum* fruit extract in broiler chicken nutrition: influence on performance and meat quality. Journal of Animal Physiology and Animal Nutrition. 2007;10:256-262.
13. Sethy K, Behera K, Mishra SK, Swain RK, Satapathy D, Sahoo JK. Grwoth, feed conservation efficiency haemato biochemical profile and immune status of Black Bengal male goats supplemented with inorganic and organic zinc in diet. Animal Science Reporter. 2016;3:10.
14. Shahad BA, Allaw AA. Effect of adding of the milk thistle (*Silibum marianum*) seed powder in the traits of biochemical blood of the quail. Plant Archives. 2020;1:962-964.
15. Singh KK, Maity SB, Maity A. Effect of nano zinc oxide on zinc bioavailability and blood biochemical changes in pre-ruminant lambs. Indian Journal of Animal Sciences. 2018;88:805-807.
16. Swain PS, Rao SBN, Rajendran D, Soren NM, Pal DT, Bhat SK. Effect of supplementation of nano zinc on rumen fermentation and fiber degradability in goats. Animal Feed Science and Technology. 2018;18:297-309.
17. Tedesco D, Tava A, Galletti S, Tameni M, Varisco G, Costa A, Steidler S. Effects of Silymarin, a Natural Hepatoprotector. Journal of Dairy Science. 2004;87:2239-2247
18. Ulger I, Onmaz AC, Ayaşan T. Effects of silymarin (*Silybum marianum*) supplementation on milk and blood parameters of dairy cattle. South African Journal of Animal Science. 2017;47:758-765.
19. Yin F, Liu J, Ji X, Wang Y, Zidichouski J, Zhang J. Silibinin: A novel inhibitor of Aβ aggregation. Neurochemistry International. 2011;58:399-403.
20. Zaboli K, Aliarabi H, Bahari AA, Abbasalipourkabir R. Role of dietary nano-zinc oxide on growth performance and blood levels of mineral. Journal of Pharma. and Health Science. 2013;2:19-26.