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**Suraj Koujalagi**

M.V.Sc., Subject Matter  
Specialist, ICAR- Krishi Vigyan  
Kendra, BIRDS Campus,  
Tukkanatti, Belagavi- 1,  
Karnataka, India

**Sushma Chhabra**

Ph.D., Professor, Department of  
Veterinary Medicine,  
GADVASU, Ludhiana, Punjab,  
India

**SNS Randhawa**

Ph.D., Professor and Head,  
Department of Microbiology,  
Khalsa College of Veterinary  
Sciences, Amritsar, Punjab,  
India

**Randhir Singh**

Ph.D. Assistant Professor  
(Veterinary Medicine),  
Department of TVCC,  
GADVASU, Ludhiana, Punjab,  
India

**DK Gupta**

Ph.D. Associate Professor,  
Department of Veterinary  
Medicine, GADVASU, Ludhiana,  
Punjab, India

**Corresponding Author:**

**Randhir Singh**

Ph.D. Assistant Professor  
(Veterinary Medicine),  
Department of TVCC,  
GADVASU, Ludhiana, Punjab,  
India

## Effect of herbal vitamin E and organic selenium complex supplementation on oxidative stress, milk quality and somatic cell count in transition dairy COWS

**Suraj Koujalagi, Sushma Chhabra, SNS Randhawa, Randhir Singh and DK Gupta**

### Abstract

Twenty dairy animals were randomly grouped into 2 groups with 10 animals each in control and supplemented group to investigate the effect of *E-Sel Power* (natural vit-E and organic selenium complex) on oxidative stress, milk quality and somatic cell count (SCC) in cross bred transition cows. Supplemented group was fed *E-Sel Power* @ 7g / 100kg body weight per day per animal during transition period for 3 weeks before and 3 weeks after parturition. Animals were sampled thrice during different stages of periparturient period viz. (i) Being Dry (BD) - 15 days before drying off (ii) Far Off Dry (FOD) - >10 days following drying off (iii) Fresh - 3-30 days in milk. Serum biochemical parameters were estimated viz. Reduced Glutathione (GSH), Superoxide Dismutase (SOD) and Lipid peroxidation (LPO) to assess the status of oxidative stress. Milk parameters viz. fat, solid not fat (SNF), lactose, density, California Mastitis Test (CMT), somatic cell count (SCC), pH and electrical conductivity (EC) were estimated. Plasma Selenium and Vit-E were also estimated, levels of which increased significantly in *E-Sel Power* supplemented group. Overall, *E-Sel Power* was found effective in reducing oxidative stress, preventing mastitis and improving production and quality of milk. Thus, the above study reveals that feeding herbal supplement *E-Sel Power* @ 7g/100 kg body weight during transition period in dairy animals helps prevent sub-clinical mastitis by marked reduction in oxidative stress and lipid peroxidation as well as increase in the blood levels of antioxidant enzymes viz. GSH and SOD, facilitating the lowering of the CMT, EC & SCC with increased% of fat, protein and SNF contents in milk along with improvement in udder immunity & health.

**Keywords:** Cow, oxidative stress, selenium, vitamin E

### Introduction

Mastitis prevalence has been reported to the extent of 44.67 percent as national average and therefore, a major deterrent to successful dairy farming due to production loss as well as cost of treatment for full recovery of the affected dairy animals. The estimated total economic losses due to mastitis in cows and buffaloes have been reported to be Rs. 2646.00 crores and Rs. 1723.32 crores, respectively due to sub-clinical mastitis and Rs. 987.60 crores and Rs. 696.29 crores, respectively due to clinical mastitis, thereby giving an overall economic loss of Rs. 6053.21 crores per year as per National Mastitis Council, 1996 [1].

A substantial decline in the plasma vitamin E and selenium levels is encountered during periparturient period, which depresses the functional capacity of peripheral blood lymphocytes and thereby increases the susceptibility to infections. It is, therefore, important to supplement antioxidant like Vitamin-E and selenium to dry cows to counter their decreased levels and oxidative stress at parturition. Supplementation of vitamin E and selenium results in significant reduction in infected quarters at calving and clinical mastitis and also improves the killing ability of blood neutrophils. Further, improvement in milk yield has also been reported due to supplementation of vitamin E in cows. Therefore, there is a need to maintain adequate levels of Vitamin-E and Selenium in peri-parturient cows. Antioxidants like Vitamin E and selenium are important for proper udder health, immune function, growth and reproduction of dairy animals. Supplementation of Vitamin E and Selenium above normal requirements is considered essential for optimization of animal health and animal product quality due to their ability to impact the lipid oxidation in biological systems.

Peri-parturient cows undergo intense mammary growth, copious synthesis and secretion of carbohydrates, fats and proteins as well as marked accumulation of colostrum and milk. Since, colostrum is rich in vitamin E, therefore, cows require increased supply of these vitamins prior to parturition. Selenium (Se) is an essential metalloid trace element, naturally occurring and required in small amounts, as a part of selenoproteins and selenoenzymes. Selenium levels in plants is related to selenium levels in surrounding soils. The normal content of selenium in forages ranges from 0.1 to 0.5 ppm. The average concentrations of selenium in water does not exceed 10mg/L [2]. Selenium deficiency is common in animals without supplementary feeding, especially with forages grown on neutral and acidic soils. Selenium forms a vital constituent of biologically important enzyme glutathione peroxidase (GPX). This enzyme reduces peroxides in cells, thus preventing oxidative injury to cells. Selenium deficiency causes reproductive disorders, as well as increased susceptibility to mastitis and its deficiency affects IgG and T cell function. Herbal E- Sel Power is a combination of natural vitamin E and selenium and its inclusion in feed can help in improving immunity, reducing oxidative stress and infection, preventing mastitis, improving milk shelf life and calf health which are the future producers. Thus, the present study was planned to investigate the effect of E-Sel Power (natural vit-E and organic selenium complex) for prevention of oxidative stress and improved udder health in cross bred cows during peri-parturient period.

#### Materials and Methods

Twenty animals in third trimester of pregnancy were selected randomly from a single dairy farm of 3000 cow population located at Latala village of Ludhiana district, Punjab. The research work was carried out in the Department of Veterinary Medicine, College of Veterinary Sciences, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab.

**Animals:** Twenty cross bred cows in transition period were selected and randomly divided in 2 groups of 10 animals each as control and treatment group.

**Experiment Design:** Ten cattle of treated group were supplemented with E-Sel Power (Indian Herbs Specialities Pvt. Ltd.) at the rate of 7g per 100 kg body weight per day for 42 days i.e. from 3 weeks before parturition to 3 weeks post parturition and were sampled three times consecutively for estimation of hemato-biochemical parameters. E-Sel Power powder contains natural forms of tocopherols along with organic complex of Selenium. 100 g E-Sel Power has Vitamin E like anti-oxidant activity equivalent to 11000 IU synthetic Vitamin E & 0.3ppm Selenium in the organic complex form.

**Blood Collection:** From each animal, blood samples were collected from jugular vein after cleaning area with spirit swab thrice during different stages of peri-parturient period viz., (i) 3 weeks before parturition, (ii) Soon after parturition and (iii) 3 weeks after parturition.

**Milk Collection:** From each animal, milk samples were collected twice during different stages viz., (i) 1 week before dry off (Being Dry) and (ii) 3 weeks post parturition. 20-30 ml of fresh milk was collected after thorough cleaning of teats with spirit swab and letting go few strips of milk from each

quarter for estimation of various milk components viz., Fat percentage, SNF, protein, lactose, pH, EC, CMT and SCC.

**Concentrations of Selenium:** One milliliter of plasma of each sample was digested in 4 ml of double distilled nitric acid over a hot plate and heated below 80° C till digestion until volume reduced to 1 – 2 ml. The digested samples were diluted with double glass distilled water and the final volume of digestate was made to 10 ml. Concentration of selenium were estimated by atomic absorption spectrophotometer (Perkin Elmer Analyst 700, USA).

#### Oxidative Stress Parameters

**Estimation of Lipid Peroxidation (LPO):** Lipid peroxidation was estimated by methods of Placer<sup>3</sup>. The method is based on the principle that the reaction of malonylaldehyde (MDA), an end product of lipid peroxidation with thiobarbituric acid (TBA) yields a pink coloured trimethine complex, which was measured by spectrophotometer at 548 nm.

**Superoxide Dismutase (SOD) Activity:** The activity of SOD in haemolysate was measured by method of Nishikimi<sup>4</sup>. The assay is based on the principle that SOD inhibits the reduction of nitro blue tetrazolium (NBT) by reduced NBT with reduced nicotinamide adenine dinucleotide (NADH) mediated by phenazinemethosulphate (PMS) under aerobic conditions.

**Estimation of Reduced Glutathione (GSH):** Reduced glutathione was estimated by the method of Hafeman<sup>5</sup>.

**Milk Analysis:** Milk samples were collected at 2 different stages, (i) 1 week before dry off (Being Dry), (ii) 3 weeks post parturition. Milk composition and quality parameters were analyzed using Lactoscan LA milk analyzer from Milktronic Ltd., Bulgaria. Milk pH (Mettler), EC (Mettler) and SCC (Somascope, Delta Instruments) and CMT was recorded.

**Estimation of Vitamin E:** Vitamin E reduces ferric ions to ferrous ions quantitatively, which combines with bathophenanthroline to form an orange colored complex<sup>6</sup>. The color was read at 536 nm.

**Statistical Analysis:** Data was analyzed statistically using SAS software (Microsoft). The data was presented as mean ± S.E. Two-way analysis of variance (ANOVA) with interaction followed by Duncan's multiple range test (DMRT) was used to estimate significant difference at 5 per cent level of significance.

#### Results

**Vitamin E:** The Vitamin E levels in serum of control and E-Sel Power supplemented group at different sampling periods during transition period are given in Table 1. In control group, the mean values ( $\mu$  mol/l) of vitamin E varied non-significantly from 30.49±1.93 (FOD) to 31.64 ± 1.55 (> 21 DIM); the lowest concentrations were noticed during fresh period with the mean values of 29.81 ± 1.86. In E-Sel Power supplemented group the mean values increased significantly ( $p < 0.05$ ) from 31.66 ± 1.99 (FOD) to 37.76± 1.26 (>21DIM).

**Selenium:** These animals of organised dairy farms were supplemented with mineral mixture (Minfa Gold, *Intas Pharmaceuticals*), which had selenium also in it. Therefore, there was increase in mean plasma Selenium levels (ppm) in control group from  $0.04 \pm 0.01$  FOD to  $0.10 \pm 0.01$  during >21 DIM (table 2). However, due to organic selenium complexed with Vitamin-E, there was significant increase in mean Selenium levels in E-Sel Power supplemented group from  $0.07 \pm 0.01$  (FOD) to  $0.31 \pm 0.04$  during >21 DIM. Though, there was over four-fold increase in the mean selenium values in E-Sel Power group but the mean selenium values in the dairy animals were within permissible limits.

#### Effect on milk constituents

**California Mastitis Test:** There was significant difference in treatment group compared to the control group. The mean CMT values declined from  $3.11 \pm 0.2$  (being dry) to  $0.55 \pm 0.28$  during > 21 DIM in E-Sel Power supplemented group (table 3).

**Electrical Conductivity and pH:** The mean EC values (mS/cm) significantly declined from being dry ( $7.07 \pm 0.25$  to  $5.97 \pm 0.32$ ) during >21 DIM in E-Sel Power supplemented group, where as in control group, non-significant decline was noticed from being dry ( $6.99 \pm 0.32$ ) to ( $6.55 \pm 0.12$ ) during > 21 DIM. Similarly, for the pH values, there was significant difference in the treatment group as compared to the control group, where the mean pH declined significantly ( $p < 0.003$ ) from  $6.9 \pm 0.07$  (being dry) to  $6.61 \pm 0.05$  during > 21 DIM in E-Sel Power supplemented group, where as in the control group, increase was noticed from  $6.69 \pm 0.12$  (being dry) to  $6.84 \pm 0.06$  during > 21 DIM (table 3).

**Somatic Cell Count:** In the current study, the SCC ( $\times 10^6$  cells/l) reduced from  $4.16 \pm 1.05$  being dry to  $0.74 \pm 0.52$  at >21DIM in E-Sel Power supplemented group while there was reduction in SCC from  $3.63 \pm 0.98$  being dry to  $1.37 \pm 0.7$  only at >21DIM in control group (table 3).

**Density:** There was significant decline ( $p < 0.05$ ) in the mean density values (%) in control group from  $34.75 \pm 0.880$  being dry to  $28.45 \pm 0.77$  during >21 DIM. On other hand there was non-significant decline in the mean density values in E-Sel Power supplemented group from  $32.36 \pm 2.35$  being dry to  $29.07 \pm 1.24$  >21 DIM (table 3).

**Lactose:** The decrease in the mean lactose values (%) was noticed in control group from  $4.01 \pm 0.22$  (being dry) to  $3.91 \pm 0.27$  during >21 DIM, while in E-Sel Power supplemented group, there was increase in the mean lactose values from  $4.17 \pm 0.21$  (being dry) to  $4.71 \pm 0.18$  during >21 DIM (table 3).

**Fat:** There was non-significant increase in the fat value (%) in control group from  $3.59 \pm 0.17$  (being dry) to  $3.72 \pm 0.49$  during >21 DIM, while in E-Sel Power supplemented group there was significant increase ( $p < 0.05$ ) in the mean fat percent from  $3.92 \pm 0.35$  (being dry) to  $4.51 \pm 0.54$  during >21DIM (table 3).

**Solid Not Fat:** In control group, there was significant increase in the mean SNF values (%) from  $7.29 \pm 0.22$  (being dry) to  $8.93 \pm 0.16$  during >21 DIM, while in E-Sel Power supplemented group, it increased from  $8.17 \pm 0.48$  (being dry)

to  $9.08 \pm 0.22$  during > 21 DIM. Further, there was higher (+ 8.22%) level of SNF in E-Sel Power supplemented group as compared to control group, during >21 DIM (table 3).

#### Oxidative stress

**Reduced Glutathione:** The mean levels of GSH, SOD and LPO of cattle of control group and E-Sel Power supplemented group are presented in Table 4. There was significant decline ( $p < 0.05$ ) in mean GSH levels (mM) of control group from  $2.78 \pm 0.06$  (FOD) to  $2.42 \pm 0.07$  during fresh period. Further decline was noticed during > 21 DIM to  $2.31 \pm 0.04$ . While in E-Sel Power supplemented group, the mean GSH levels declined from  $2.66 \pm 0.07$  (FOD) to  $2.42 \pm 0.07$  during fresh period but there was increase of mean GSH levels to  $2.49 \pm 0.09$  during > 21 DIM.

**Superoxide Dismutase:** There was significant decline ( $p < 0.05$ ) in the mean SOD values (U/mg Hb) from  $59.41 \pm 3.57$  (FOD) to  $48.33 \pm 0.59$  during >21 DIM in control group. In E-Sel Power supplemented group, there was significant increase in the mean SOD values from  $56.54 \pm 3.81$  (FOD) to  $63.74 \pm 3.07$  during >21 DIM.

**Lipid Peroxidation:** There was significant increase in the mean LPO values (nmol/g Hb) from  $156.72 \pm 1.87$  (FOD) to  $191.98 \pm 3.82$  during fresh period to  $264.66 \pm 8.06$  ( $p < 0.05$ ) during >21 DIM in control group. In E-Sel Power supplemented group, the mean LPO values increased from  $150.21 \pm 4.15$  (FOD) to  $194.34 \pm 5.38$  ( $p < 0.05$ ) during fresh period, but there was decline in mean LPO values to  $182.95 \pm 20.03$  during >21 DIM.

#### Discussion

**Vitamin E:** Similar to the present study, previous studies [7, 8] reported decrease in serum vitamin E levels during periparturient period in unsupplemented cows, and as risk factor for intra-mammary infection during first week of lactation. Researchers [8] concluded that cows with  $< 2.5 \mu\text{g/ml}$  of plasma concentration of vitamin -E were 2.8 times more prone to mastitis and the *in vitro* killing activity of neutrophils was maximized, when plasma tocopherol concentration was about  $3.4 \mu\text{g/ml}$  at parturition and cows with  $> 3.0 \mu\text{g/ml}$  had no risk for intramammary infections.

Vitamin E inhibits auto-oxidation of polyunsaturated fatty acids in neutrophil membranes and enhances neutrophil function. Dietary supplementation of vitamin E increased intracellular killing by bovine neutrophils [9]. Vitamin E supplementation is associated with higher chemotactic activity and faster migration of neutrophils towards the mammary gland [10], because membrane-bound u-PA is involved in extravasation and migration of neutrophils to inflamed tissues.

**Selenium:** Previous studies [11] reported the mean plasma selenium concentrations of 0.06 ppm in non selenotic cows, whereas, the mean plasma selenium concentrations of 0.60 ppm in selenotic cows. Selenium deficiencies in cows were associated with decreased intracellular kill by neutrophils and Selenium supplementation increased intracellular kill of *S. aureus* by neutrophils [12]. Vitamin E and selenium act synergistically; the blood concentration of one affects the other. Vitamin E inhibits peroxidation of polyunsaturated fatty acids by sequestering free radicals; whereas selenium, as a component of glutathione peroxidase, reduces the

hydroperoxides formed to the less reactive alcohol. However, prior to supplementation of E-Sel Power to dairy animals, it must be ensured that such animals are not supplied with any other mineral mixture having selenium content in that or have not been given parenteral injection of selenium.

### Milk Constituents

**California Mastitis Test:** Previous studies [13] reported that when California Mastitis Test (CMT) score increased from 1, 2 and 3 a corresponding 10-11, 16-21 and 21-25 percent decrease in the milk yield was observed. Infection and tissue repair were common even in well-managed dairy herds, and cows might experience some degree of immune response, especially after calving. Stress, disease, and induction of the immune response increases requirement of nutrients, including vitamins and essential trace elements. Inadequacies of these nutrients, required for both immunity and antioxidant defense, could impair function of both systems.

**Electrical Conductivity and pH:** The results indicated efficacy of E-Sel Power (Herbal vitamin E and organic Selenium) supplementation as decreased leakage of sodium, chloride and immunoglobulins from blood to milk in turn decreases pH and EC values of milk. Several studies provided evidence that vitamin E supplementation slows down oxidative deterioration of milk [14]. There is no optimal milk vitamin E concentration established by the dairy industry that will help milk maintain its freshness and block auto-oxidation processes by stabilizing pH and hence EC, until milk is consumed. Previous studies [10] suggested that supplementation with 1000 IU vitamin E/day may be helpful in herds with milk with off-flavors by stabilizing the ion concentrations in the milk ultimately reducing pH and EC.

**Somatic Cell Count:** Similar to the present study, previous studies [15] reported that, heifers produced milk with low SCC at day 14 of lactation when fed with vitamin E. A decrease in milk production was reported when SCC was  $0.5 \times 10^6$ /ml. As the SCC increased from  $110^6$ ,  $2 \times 10^6$  and  $4 \times 10^6$ /ml milk, a decrease in milk production of 8, 15 and 27 percent, respectively was recorded. Vitamin E and Se supplementation can reduce both the incidence of clinical mastitis and the duration of symptoms of this disease. A reduction in SCC and percentage of PMN during the first period of the subsequent lactation (especially at 30 DIM) could have been a positive

consequence of the supplemental vitamin E and Se, which prevents the suppression of neutrophil function during the early postpartum period [16]. In fact, PMN represent the cell class mainly involved in the inflammatory process and are important in the defense of the mammary gland [10]. The phagocytosis of pathogens results in a respiratory burst with an increase in oxygen consumption and production of reactive oxygen species that kill pathogens. In fact, Se as a constituent of GSH-Px, together with vitamin E, share a biological role as antioxidants and partially neutralize the negative effects of reactive oxygen species in the cells, and reduce the somatic cell count.

### Oxidative stress

**Reduced Glutathione:** Similar to the present study, researchers [17] in their previous study, found that the initial glutathione peroxidase activity in heifers increased from  $14.6 \pm 0.9$  to  $21.6 \pm 1.6$  U/mg Hb six weeks after supplementation with vitamin E and Selenium. Miller [19] reported that the reduction in Se intake, at a time when there was excessive exposure to stimulators of ROS production resulted in relative deficiency of GPx activity and development of oxidative stress. Selenium deficiencies in cows were associated with decreased blood concentrations of GSH-Px and intracellular killing by neutrophils. Selenium supplementation also maintained whole blood GSH-Px activity. The Se containing enzyme GSH-Px can protect neutrophils by detoxifying peroxides in the cytosol [19].

**Superoxide Dismutase:** Similar to the present study, Singh [20] reported significantly low mean SOD levels from far off dry to fresh stage in buffaloes. Similarly, other researchers [21] reported significant increase in ROS and decrease in GPx and SOD activity during postpartum period. Thus, it was concluded that the treatment followed was effective in improving the SOD activity significantly.

**Lipid Peroxidation:** Previously researchers [19] reported significantly high LPO levels in early lactating cows than advanced pregnant cows. Similar to the present study, other researchers [21] also reported significantly high mean LPO levels from far off dry to fresh stage in buffaloes. Vitamin E is concentrated in tissues producing steroid hormones, in which it protects highly sensitive steroidogenic activities of cytochrome P-450 against lipid peroxidation.

**Table 1:** Mean Vitamin E levels in serum of control and Vitamin E-Sel. supplemented group at different sampling periods during transition period (Mean  $\pm$  Standard error)

Parameter	Period	Control	E-Sel Power supplemented	% change over control
Vitamin E ( $\mu$ mol/l)	Far off dry (FOD)	30.49 $\pm$ 1.93 <sup>Ax</sup>	31.66 $\pm$ 1.99 <sup>Ay</sup>	-
	Fresh	29.81 $\pm$ 1.86 <sup>Ax</sup>	33.28 $\pm$ 1.81 <sup>Axy</sup>	+ 11.64%
	> 21 Days in milk (DIM)	31.64 $\pm$ 1.55 <sup>Bx</sup>	37.76 $\pm$ 1.26 <sup>Ax</sup>	+ 19.34%
Values bearing different superscript in capital letters (A, B) across the row vary significantly ( $p < 0.05$ ). Values bearing different superscript in small letters (x, y) down the column vary significantly ( $p < 0.05$ ).				

**Table 2:** Mean Selenium values in plasma of control and Vitamin E-Sel. Supplemented group cows at different sampling periods (Mean  $\pm$  Standard error)

Parameter	Period	Control	E-Sel Power supplemented	% change over control
Selenium (ppm)	Far off dry (FOD)	0.04 $\pm$ 0.01 <sup>Ay</sup>	0.07 $\pm$ 0.01 <sup>Ay</sup>	+ 75%
	> 21 Days in milk (DIM)	0.10 $\pm$ 0.01 <sup>Bx</sup>	0.31 $\pm$ 0.04 <sup>Ax</sup>	+ 210%
Values bearing different superscript in capital letters (A, B) across the row vary significantly ( $p < 0.05$ ). Values bearing different superscript in small letters (x, y) down the column vary significantly ( $p < 0.05$ ).				

**Table 3:** Mean values of milk constituents in control and Vitamin E-Sel. supplemented group cows at different sampling periods (Mean  $\pm$  Standard error)

Parameters	Period	Control	E-Sel Power supplemented	% change over control
CMT (5 Point Scale)	Being Dry	3 $\pm$ 0.41 <sup>Ay</sup>	3.11 $\pm$ 0.2 <sup>Ax</sup>	
	> 21 Days in milk (DIM)	1.18 $\pm$ 0.38 <sup>Ax</sup>	0.55 $\pm$ 0.28 <sup>Ay</sup>	(-) 53.38
EC (mS/cm)	Being Dry	6.99 $\pm$ 0.32 <sup>Ax</sup>	7.07 $\pm$ 0.25 <sup>Ax</sup>	
	> 21 DIM	6.65 $\pm$ 0.12 <sup>Ax</sup>	5.97 $\pm$ 0.32 <sup>Ay</sup>	(-) 10.22
SCC (x10 <sup>6</sup> cells/l)	Being Dry	3.63 $\pm$ 0.98 <sup>Ax</sup>	4.16 $\pm$ 1.05 <sup>Ax</sup>	
	> 21 DIM	1.37 $\pm$ 0.7 <sup>Ax</sup>	0.74 $\pm$ 0.52 <sup>Ay</sup>	(-) 45.75
SCS	Being Dry	6.37 $\pm$ 0.17 <sup>Ay</sup>	6.43 $\pm$ 0.16 <sup>Ax</sup>	
	> 21 DIM	5.77 $\pm$ 0.18 <sup>Ax</sup>	5.38 $\pm$ 0.17 <sup>Ay</sup>	(-) 6.75
pH	Being Dry	6.69 $\pm$ 0.12 <sup>Ax</sup>	6.9 $\pm$ 0.07 <sup>Ax</sup>	
	> 21 DIM	6.84 $\pm$ 0.06 <sup>Ax</sup>	6.61 $\pm$ 0.05 <sup>By</sup>	(-) 3.36
Density (%)	Being Dry	34.75 $\pm$ 0.88 <sup>Ay</sup>	32.36 $\pm$ 2.35 <sup>Ax</sup>	
	> 21 DIM	28.45 $\pm$ 0.77 <sup>Ax</sup>	29.07 $\pm$ 1.24 <sup>Ax</sup>	+ 2.18
Lactose (%)	Being Dry	4.01 $\pm$ 0.22 <sup>Ax</sup>	4.17 $\pm$ 0.21 <sup>Ax</sup>	
	> 21 DIM	3.91 $\pm$ 0.27 <sup>Bx</sup>	4.71 $\pm$ 0.18 <sup>Ax</sup>	+ 20.46
FAT (%)	Being Dry	3.59 $\pm$ 0.17 <sup>Ax</sup>	3.92 $\pm$ 0.35 <sup>Ay</sup>	
	> 21 DIM	3.72 $\pm$ 0.49 <sup>Bx</sup>	4.51 $\pm$ 0.54 <sup>Ax</sup>	+ 21.23
Protein (%)	Being Dry	3.62 $\pm$ 0.15 <sup>Ax</sup>	3.36 $\pm$ 0.24 <sup>Ax</sup>	
	> 21 DIM	3.44 $\pm$ 0.06 <sup>Bx</sup>	3.66 $\pm$ 0.07 <sup>Ax</sup>	+ 6.39
SNF (%)	Being Dry	7.29 $\pm$ 0.22 <sup>Ay</sup>	8.17 $\pm$ 0.48 <sup>Ax</sup>	
	> 21 DIM	8.39 $\pm$ 0.16 <sup>Bx</sup>	9.08 $\pm$ 0.22 <sup>Ax</sup>	+ 8.22

Values bearing different superscript in capital letters (A, B) across the row vary significantly ( $p < 0.05$ ).  
Values bearing different superscript in small letters (x, y) down the column vary significantly ( $p < 0.05$ ).  
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**Table 4:** Mean oxidative stress parameter values at different sampling periods of control cows and cows supplemented with Vitamin E-Sel. (Mean  $\pm$  Standard error).

Parameters	Period	Control Group	Treatment Group (E-Sel Power)	% change over control
Reduced Glutathione GSH (mM)	Far off dry (FOD)	2.78 $\pm$ 0.06 <sup>Ax</sup>	2.66 $\pm$ 0.07 <sup>Ax</sup>	
	Fresh	2.42 $\pm$ 0.07 <sup>Ay</sup>	2.42 $\pm$ 0.07 <sup>Ay</sup>	
	> 21 Days in milk (DIM)	2.31 $\pm$ 0.04 <sup>Ay</sup>	2.49 $\pm$ 0.09 <sup>Axy</sup>	+ 7.79%
Superoxide Dismutase SOD (U/ mg Hb)	FOD	59.41 $\pm$ 3.57 <sup>Ax</sup>	56.54 $\pm$ 3.81 <sup>Ax</sup>	
	Fresh	57.84 $\pm$ 2.27 <sup>Ax</sup>	57.09 $\pm$ 2.25 <sup>Ax</sup>	
	> 21 DIM	48.33 $\pm$ 0.59 <sup>Ay</sup>	63.74 $\pm$ 3.07 <sup>Bx</sup>	+ 31.88%
Lipid Peroxidation LPO (n mol/g Hb)	FOD	156.72 $\pm$ 1.87 <sup>Az</sup>	150.2 $\pm$ 4.15 <sup>Ay</sup>	
	Fresh	191.98 $\pm$ 3.82 <sup>Ay</sup>	194.34 $\pm$ 5.38 <sup>Ax</sup>	
	> 21 DIM	261.66 $\pm$ 8.06 <sup>Ax</sup>	182.95 $\pm$ 20.03 <sup>Bxy</sup>	(-) 29.51%

Values bearing different superscript in capital letters (A, B) across the row vary significantly ( $p < 0.05$ ).  
Values bearing different superscript in small letters (x, y) down the column vary significantly ( $p < 0.05$ ).

## Conclusion

The present study reveals that feeding herbal E-Sel Power @ 7g per 100 kg body weight (or 30g/day) for 3 weeks before expected parturition and 3 weeks after parturition in dairy animals increases the levels of serum Vitamin E and plasma Selenium leading to marked reduction in oxidative stress and lipid peroxidation as well as increase in the blood levels of antioxidant enzymes viz. GSH and SOD facilitating the lowering of the CMT, EC SCC in milk and improvement in udder immunity and health as well as in milk quality with increased fat, protein and SNF contents in milk.

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