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Research Article

OXIDATIVE STATUS AND ACUTE PHASE RESPONSE OF SOWS DURING TRANSITION PERIOD

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ABSTRACT: The present study was conducted in pregnant sows to evaluate the oxidative status and inflammatory markers during the transition period. Blood samples were collected from sows on the days -30, -10, -3, 0, +3, and +10 of farrowing. Markers of oxidative status such as malondialdehyde (MDA), lipid hydroperoxides (LHP), total antioxidant capacity (TAC), glutathione (GSH), superoxide dismutase (SOD), glutathione-S-transferase (GST), catalase, glutathione peroxidase (GPx) and C- reactive protein (CRP) were evaluated. Lipid hydroperoxide levels significantly increased, while TAC and GSH levels significantly decreased on days 0 and +3 compared to day -30 of farrowing. The highest activity of SOD was observed on day -10, while GST activity was at its peak on day 0. Before farrowing, glutathione peroxidase was the active peroxidase enzyme; catalase was more active after farrowing. C-reactive protein (CRP) levels increased significantly during the transition period compared to day -30 of farrowing. Depleted levels of antioxidant molecules after farrowing emphasize the significance of antioxidant supplementation to maintain the increased antioxidant enzyme activity. Elevated serum CRP levels during the transition period indicate the risk of development of metabolic disturbances like mastitis, metritis, and agalactia in transition sows.

Keywords: Oxidative stress markers, Antioxidant status, Antioxidant enzyme activities, Inflammatory marker, Transition period, Sows.

INTRODUCTION

The transition phase from late gestation to early lactation (day -10 to day +10) is considered one of the most critical periods for health and productivity in pigs [1]. The metabolic adaptations during the transition phase could in turn result in altered oxidative/ antioxidative status, which influences the ability of sows to produce milk and, in turn, affects the growth of their piglets [2]. During the puerperium, physiological events like an increase in myometrial activity, subsequent relaxation, and dilatation of the cervix and the caudal part of the birth canal during expulsion occur which increase the acute phase proteins (APP) in serum [3]. Serum CRP is an APP that is considered a marker of early stages of MMA (mastitis, metritis, agalactia) in transition sows [4]. The suboptimal transition from the late-gestation period to lactation can impair production and reproductive performance and cause economic losses [5]. A better understanding of the redox and inflammatory status during the transition period may facilitate the development of management and nutritional solutions to prevent metabolic disorders.

Though such studies have been carried out abroad on well-known exotic breeds, there is a lack of information about the transition physiology of cross-breed sows developed in India. SVVU-17 is the crossbreed variety of pigs developed at AICRP on pigs, SVVU, Tirupati. Thus, the present investigation was carried out to analyze the oxidative and inflammatory status of SVVU-17 sows during the transition period.

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MATERIALS AND METHODS

Sample collection

Ten healthy pregnant sows (75% LWY crossbred pig variety SVVU T17) in second parity with an average litter size of 8-12 piglets were selected for the current investigation. The sows were maintained under standard management practices at AICRP on pigs, S.V.V.U, Tirupati, Andhra Pradesh. The care of sows was undertaken as per the guidelines of the International Animal Ethics Committee. The committee for the purpose of control and supervision of experiments on animals (CPCSEA), India has approved the protocol of the experiment (No.281/go/REeBi/S/2000/CPCSEA/ CVSc/TPTY/010/).

Blood samples were collected from the selected sows on days 30, 10, and 3 before farrowing, on the day of farrowing, and 3 and 10 days after farrowing. The bleeding was done by venipuncture of the ear vein and blood was collected separately into K2 EDTA and clot activator vials. Whole blood in EDTA vials was used for hematology studies. The blood collected in clot activator vials was centrifuged at 2500 rpm for 5 min to separate serum and stored at -80ºC for further analysis.

Analysis of oxidative status and C-reactive protein

Blood collected in EDTA vials was used for the preparation of hemolysate. Blood samples were centrifuged at 3000 rpm for 15 min for separation of the RBC pellet. The resulting erythrocyte sediment was washed thrice with 0.9% w/v NaCl solution, each time by mixing and centrifuging the suspension at 2000 rpm for 5 min and the supernatant was discarded. The washed red cells are lysed by adding 4 parts of chilled distilled water to give stock hemolysate solution (20% v/v). The hemolysate was then analyzed to estimate the oxidative stress and antioxidant status.

The concentration of MDA [6] and LHP [7] was measured to estimate lipid peroxidation by spectrophotometry. Total antioxidant capacity [8], glutathione [9], and the protein content [10] of the hemolysate were estimated using a UV-Vis spectrophotometer. The activity of antioxidant enzymes was analyzed spectrophotometrically by the respective standard protocols, *i.e.*, SOD [11], catalase [12], GST [13], and GPx [14].

The serum samples were analyzed for C - reactive protein, an inflammatory marker by ELISA using a CRP test kit (Arkray Healthcare Pvt. Ltd, India.).

Statistical analysis

All the data obtained were subjected to One way ANOVA [15] followed by Duncan's multiple range test (SPSS version 22).

RESULTS AND DISCUSSION

Lipid hydroperoxides are primary lipid oxidation products formed during peroxidative processes of unsaturated phospholipids, glycolipids, and cholesterol while malondialdehydes are the major secondary oxidation products or end products of lipid peroxidation [16]. Earlier studies reported a rise in the LHP levels on day +10 compared to day -14 of farrowing in sows [17]. No change was reported in MDA levels during the peripartum period of sows [18]. In the present study, significantly ($p<0.05$) higher LHP levels were observed during day 0 and +3 compared to day -30 of farrowing while there was a non-significant rise in MDA levels during the transition period compared to day -30 of farrowing (Table 1). Similar findings were reported earlier in cows where an increase in LHP during the transition period was associated with no significant change in MDA levels, which might be due to the interference of the antioxidant system during the process of lipid peroxidation [19].

In the present study, the results showed significantly lower TAC (Table 1) and GSH levels (Fig. 1A) on day 0 and +3 compared to day -30 of farrowing. The results are by previous studies [20, 21] in sows where TAC concentrations decreased around parturition. Similar results were reported in camels [22], where GSH levels gradually decreased from late pregnancy till early lactation. Decreased GSH levels around calving during the transition period of cows, might be due to the consumption of GSH in the chain reactions of lipid peroxidation [19]. A similar trend was observed in the present study where decreased GSH levels were associated with increased levels of LHP. The present study showed increased SOD activity on day -10, -3 compared to day -30 of farrowing and decreased activity from the day of farrowing until day +10 of farrowing (Fig. 1B). An increased SOD activity during late gestation until parturition was reported in previous studies on sows [20, 21, 23]. Increased SOD activity during the late gestation period with a rapid decline in its activity after calving in cows could be due to homeostatic regulation during late pregnancy and reduced availability of Zn/Cu in the early postpartum period [19].

A progressive increase in GST activity was observed during the current study from day -30 to day 0 of

Table 1. Markers of oxidative status during the transition period in sows.

Parameters	Day -30	Day -10	Day -3	Day 0	Day +3	Day +10
MDA (µg/ml hemolysate)	2.06^{a} ±0.17	2.36^{a} ±0.31	2.69^{a} ±0.24	2.80^{a} ±0.17	2.48^{a} ±0.16	2.32^{a} ±0.32
Lipid hydroperoxides (X 10^{3} mM/ml hemolysate)	21.10^{a} ±2.90	21.33^{ab} ±1.42	22.09^{ab} ±0.34	26.45^{bc} ±1.91	29.00^{c} ±1.01	25.82^{abc} ±1.01
Total Antioxidant Capacity (X 10^{-3} mM GSH equivalents/mg protein)	61.40^{b} ±9.84	57.97^{b} ±9.51	45.57^{ab} ±6.42	26.50^{a} ±4.38	34.92^{a} ±4.55	42.10^{ab} ±1.59

*Values are Mean ± SE (n=10); a, b, c different superscripts row wise differ significantly (p<0.05).

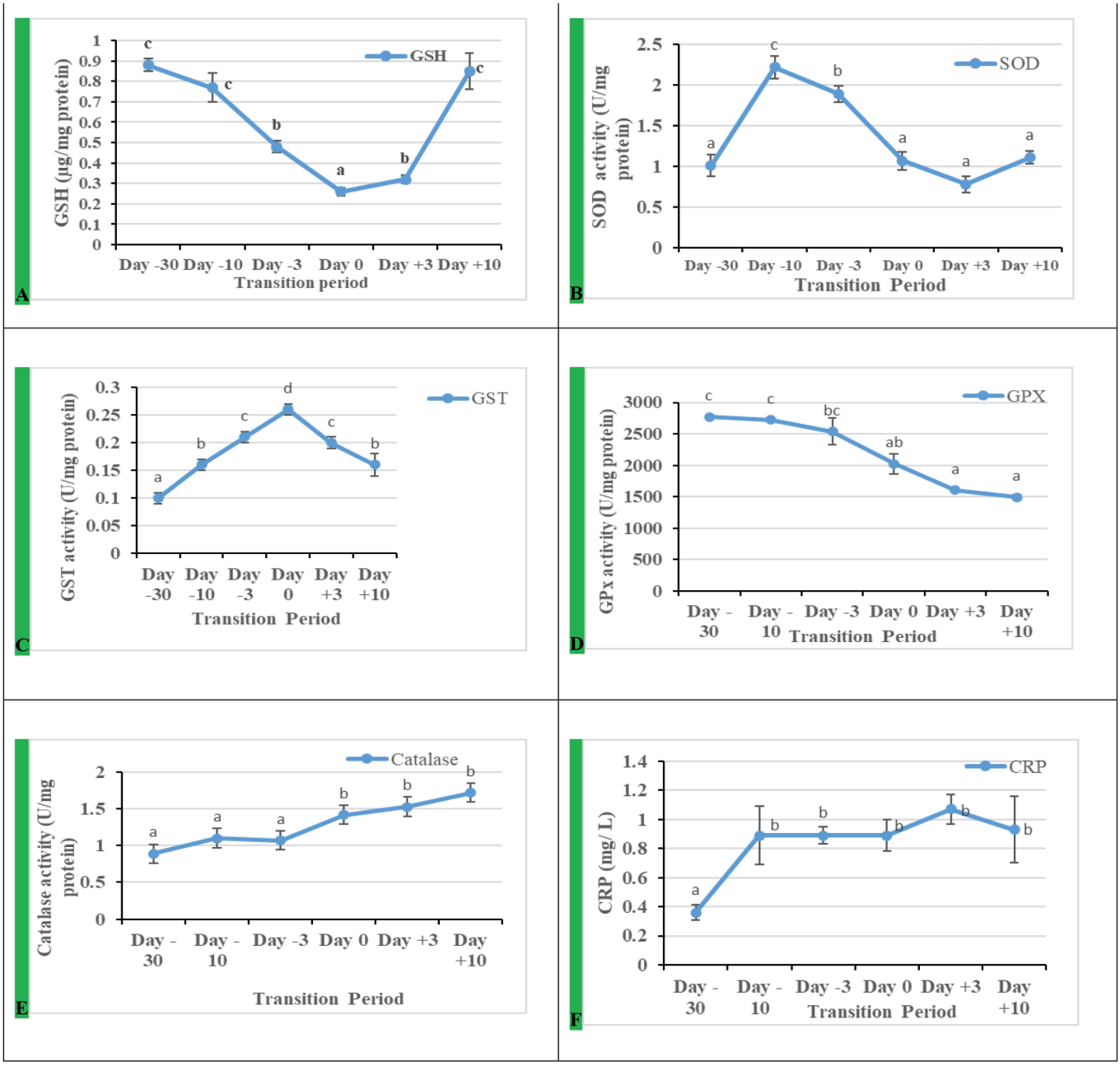

Fig. 1. Antioxidant status and inflammatory status of sows during transition period. (Values are Mean ± SE (n=10), a, b, c different superscripts differ significantly at p<0.05).

farrowing and decreased thereafter (Fig. 1C). The increase in GST activity might be due to its involvement in buffering undesirable oxidative reactions before and on the day of farrowing, where GSH levels were decreased [24]. Decreased activity of GPx during the early postpartum period (day +3 to day +10), is in coincidence with previous studies [20, 21, 25] in sows. Similar results were reported earlier [26], where GPx activity decreased in early lactating cows compared to the advanced pregnant cows. Decreased erythrocytic GPx activity around calving and early lactation in cows was attributed to the susceptibility of enzymes to oxidative reactive molecules and decreased trace mineral availability around parturition [19].

The results showed lower catalase activity from day -30 to -3 compared to the other time points of the transition period (Fig. 1E), which is similar to a previous study [26], where lower catalase activity was reported in the advanced pregnant cows compared to the early lactation group. This may be due to metabolic demands associated with late pregnancy that increase the production of reactive oxygen species, which in turn reduce the catalase activity during late pregnancy. Several other authors have reported a decrease in catalase activity during the transition period of sows [17, 21, 25].

Acute phase proteins in serum are considered markers of infections, stress, and inflammatory events [27]. Serum CRP levels increased ($p<0.05$) around 3 folds during the transition period compared to day -30 and a peak value was noticed on the day +3 of farrowing (Fig. 1F). An increase in CRP concentration on the day of parturition compared to day - 28 of farrowing, which remained elevated until day +7 was reported in sows [28]. A similar increase in CRP concentrations at week 1 after farrowing compared to the 4th and 1st week before farrowing in sows was also reported [29]. The inflammation of the reproductive tract and mammary gland caused by the periparturient increase in the number of both non-pathogenic microflora and facultative pathogens could have led to the elevated serum CRP during the transition phase [28, 30].

CONCLUSION

The depleted levels of GSH and TAC from day -3 to day +3 of farrowing in the present study revealed the increase in antioxidant demands to ameliorate the reactive oxygen species generated during the peripartum period in sows. Thus, the supplementation of antioxidants would be necessary to better counteract oxidative stress during the peripartum period. Serum CRP levels remained elevated during the entire transition phase (from day -10 to day +10) which indicates the importance of anti-inflammatory agents during the transition period to prevent the development of metabolic disturbances like MMA, thus improving the growth performance of piglets and economics of pig rearing.

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