Effect of Vitamin C on Performance and Antioxidant Capacity of Cattle Fed Varying Concentrations of Dietary Sulfur

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Summary and Implications

Limited research is available concerning the effects of feeding supplemental vitamin C (**VC**) to feedlot cattle. This study concluded that supplementation of VC to cattle receiving a high S diet improved the marbling scores of these steers, which may be related to the greater plasma VC measured in these cattle.

Introduction

Ethanol industry co-products, such as dried distiller's grains with solubles (**DDGS**), are a common addition to Midwest feedlot diets. However, as a result of the sulfuric acid used during the ethanol fermentation process, DDGS are often rich in sulfur (**S**), limiting the inclusion rate in cattle diets. High dietary S has been documented to be detrimental to cattle performance, specifically influencing average daily gain (**ADG**) and hot carcass weight (**HCW**), and may lead to the development of S toxicity.

VC is a potent antioxidant, which is naturally produced in cattle. However, limited research is available concerning the VC needs of feedlot cattle. Previous research noted a decrease in plasma VC during the finishing period of long-fed cattle and observed an increase in marbling scores when a rumen-protected VC source was added to the diet during the finishing period. Additionally, cell culture work suggests VC enhances differentiation of pre-adipocytes to adipocytes, a step needed for marbling to occur.

The objective of this study was to determine the effects of supplementing VC to diets containing varying concentrations of S on the live and carcass-based performance and antioxidant capacity of cattle.

Materials and Methods

Calf-fed steers (n = 120) were started on a receiving ration for 28 d, followed by 3 step-up rations in preparation for the finishing diet. Cattle were limit fed at 1.5% BW and increased 0.25% each day for the first 7 d of the study, to decrease risk for acidosis and development of polioencephalomalacia symptoms (**PEM**, S toxicity). Cattle were blocked by initial BW (781 ± 50 lbs) and randomly assigned to the following treatments: 1) low S (0.2% S), 2) low S diet + VC, 3) medium S (0.4% S), 4) medium S diet + VC, 5) high S (0.06% S); medium S diet + sodium sulfate, and 6) high S diet + VC (**Table 1**). Rumen-protected VC was targeted at 10 g VC per head per day.

Consecutive day weights were taken at the beginning and end of the trial, and every 28 d in between. Jugular blood was collected from 2 steers per pen on d 0, 14, 28, 90, and 143. Plasma was aliquoted and stored for analysis of total antioxidant (**TA**) activity and VC concentration. As a measure of dietary S exposure, rumen hydrogen sulfide (**H_2S**) and whole blood sulfhemoglobin concentrations were determined from one steer per pen on d 0, 14, 28, 90, and 143.

Steers were harvested at Tyson's Fresh Meats in Denison, IA on d 149, when at least 75% of the steers in a pen were estimated by visual appraisal to have 0.5 in of back fat. Cattle were graded according to USDA standards and a strip loin steak, for lipid extraction and fatty acid analysis, was collected from each carcass.

Data were analyzed as a complete randomized block design using the MIXED procedure of SAS. Single DF contrasts were used to compare: A) VC vs. no VC, B) linear effect of S, C) VC within high sulfur, D) VC within medium and high S, and E) VC within low S.

Results and Discussion

Performance results of this study are consistent with previous research findings that high S has negative impacts on the performance of cattle, which was evident by the linear decrease ($P < 0.01$; **Table 2**) in dry matter intake (**DMI**) and ADG when dietary S increased. Within the high S treatment, the addition of VC tended ($P < 0.10$) to improve DMI, while *numerical* benefits were observed in the improvement of final BW and ADG. Overall, the addition of VC to the high S treatment resulted in improved animal performance.

Previous research data also indicate feeding high dietary S may result in lower quality grade (**QG**) and lighter HCW. Results of this study are congruent with those findings, as linear decreases ($P < 0.05$; **Table 3**) in yield grade (**YG**), marbling score, back-fat thickness, and HCW were observed as diets increased in dietary S concentration.

Interestingly, the positive effects of VC were noted within the high S treatment, as the inclusion of VC increased ($P < 0.01$) marbling scores from high Select to low Choice and also increased back-fat thickness. The addition of VC to the high S treatment successfully eliminated the negative impacts imposed by high S, as the quality of the carcasses were comparable to the low and medium S treatments, even though the high S + VC carcasses were lighter by about 30 lbs. Addition of VC to any level of dietary S resulted in fatter cattle, as evident by an increase ($P < 0.01$) in kidney, pelvic, and heart fat (**KPH**). It is unclear what exact mechanism is driving the increase in marbling and back fat in the high S cattle and

KPH fat production in cattle receiving VC (regardless of S inclusion), further research in this area is certainly warranted.

Previous research data indicate the addition of ethanol co-products increases the polyunsaturated fatty acid (**PUFA**) profile of meat, which may be attributed to the higher percentage of fat in those co-products. In the present study, cattle consuming increasing levels of dietary S had a greater percentage ($P < 0.01$; **Table 4**) of PUFAs and omega 6 fatty acids (**n6**), while tending to decrease saturated fatty acids (**SFA**). These findings are consistent with previous research as the low S treatment was primarily a corn-based diet, while the medium and high S treatments were 40% DDGS.

Addition of VC to the high S treatment resulted in less ($P < 0.05$) SFA, while the addition of VC to any level of dietary S tended to lower ($P < 0.10$) the percentage of SFA. The PUFA-to-SFA ratio was increased ($P < 0.05$) with the increase in dietary S, primarily being driven by the VC treatments within the low and high S treatments (ratio of 0.15 and 0.24, respectively). Additionally, within the low S treatment the inclusion of VC increased ($P < 0.05$) omega 3 fatty acids (**n3**) from 0.37% to 0.50%, while the other two treatments differed by 0.01 or 0.03%. This difference among the low S treatment was also the driving factor for the tendency ($P < 0.10$) of VC to positively influence the ratio of n3-to- n6 fatty acids from 0.05 to 0.08, where a higher ratio is better. There was no effect of dietary S or VC inclusion on the meat profile of medium chain fatty acids (**MCFA**), long chain fatty acids (**LCFA**), or the calculated atherogenic index (**AI**).

Previous research data indicated H_2S values peak within the first 28 d, and during this time period cattle are susceptible to the development of PEM symptoms. Hydrogen sulfide values in the current study peaked at 3500 ppm on d 14 (in the high S cattle), and all treatments experienced a decrease in H_2S production between d 90 and 143, due to a new load of DDGS (lower in S than previous loads). Increased inclusion of dietary S resulted in a linear increase ($P < 0.01$) in H_2S concentrations (**Figure 1**). VC supplementation did not affect H_2S concentrations at any level of dietary S ($P = 0.23$)

Sulfhemoglobin is created when S in the blood binds to the iron molecule in hemoglobin in place of oxygen, thereby decreasing the oxygen carrying capacity of the blood. In this study sulfhemoglobin was greater ($P < 0.05$; **Table 5**) in steers receiving medium and high S diets compared to the low S diets. The percent of total hemoglobin that is sulfhemoglobin increased ($P < 0.01$) with the increase in inclusion rate of dietary S, but these cattle were well below the accepted toxic threshold of 1%.

Limited research data are available concerning plasma VC concentrations and feedlot cattle. The research data available noted a decrease in plasma VC during the finishing period. In the present study inclusion of VC in the diet resulted in greater ($P < 0.05$; **Figure 2**) plasma VC concentrations; most notably within the high S treatment ($P < 0.01$) and within the medium and high S treatments combined ($P < 0.05$). Cattle consuming the high S diet experienced a decrease in plasma VC between d 28 and 90, but supplementing the high S cattle with VC prevented the decrease, resulting in the highest VC values of all the treatments. The low and medium S treatments remained relatively consistent throughout the entire study.

Similar to plasma VC there are limited research data available concerning plasma TA capacity of feedlot cattle, but preliminary data suggest increasing dietary S results in a decrease in plasma TA capacity. In the present study, cattle receiving increasing levels of dietary S experienced a linear decrease ($P < 0.05$; **Figure 3**) in TA capacity on d 90 and 143. Antioxidants are critical for maintenance of cellular integrity, as well as supporting the immune response. There was no effect of VC inclusion on TA capacity. As VC is a water soluble vitamin it has a limited storage capacity within the body. Therefore, VC may not have been directly affecting the TA capacity, but may have been contributing to regeneration of vitamin E or assisting in relieving stress on other antioxidants.

In conclusion, supplementation of VC to cattle receiving the low and medium S diets saw varying benefits, suggesting the improvements noted in the high S treatment may have been in response to the stress of the high dietary S. Addition of VC to the high S diet improved the marbling scores of these steers, and while the exact mechanism for this improvement is unknown, it may be related to the greater circulating VC in these cattle. Since TA capacity was decreased by increasing dietary concentrations of S it is possible that plasma VC was being used in these steers in place of other functional antioxidants. This may be why plasma VC is so much lower in steers receiving the high S diet. The addition of VC to the high S diet recovered plasma VC concentrations, meaning these cattle had more VC available for functions related to lipid metabolism, perhaps leading to the observed improvement in marbling scores. Additionally, VC may be beneficial in improving the overall "healthfulness" of meat products, as indicated by the increase in the ratio of n3-to-n6 fatty acids and general decrease in SFA.

Acknowledgements

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Table 1. Composition of finishing diet used in feedlot study (% DM basis).

Item	Low S[1]	Medium S[1]	High S[1,2]
Corn	69.7	48.0	48.0
Corn dried distiller's grains	18.0	40.0	38.9
Chopped hay	9.00	9.00	9.00
Limestone	2.00	2.00	2.00
Salt	0.31	0.31	0.31
Vitamin A premix[3]	0.10	0.10	0.10
Trace mineral premix[4]	0.035	0.035	0.035
Rumensin90[5]	0.016	0.016	0.016
Sodium sulfate[2]	--	--	1.11
Urea	0.80	--	--

[1]Vitashure C (Balchem Corp) included 0.22 % DM to achieve 10 g of vit C per head per day; included in treatments 2, 4, and 6

[2]Sodium sulfate included to achieve a dietary sulfur value of 0.6% at the expense of DDGS

[3]Vitamin A premix contains 4,400,000 IU/kg

[4]Provided per kg of diet: 30 mg Zn as $ZnSO_4$; 20 mg Mn as $MnSO_4$; 0.5 mg I as $Ca(IO_3)_2(H_2O)$; 0.1 mg Se as Na_2SeO_3; 10 mg Cu as $CuSO_4$; and 0.1 mg Co as $CoCO_3$

[5]Provided at 27 g/ton diet

Table 2. Effect of supplemental vitamin C on performance of steers fed low, medium, or high S diets.

Diet Sulfur	Low		Medium		High			
Diet Vit C	-	+	-	+	-	+	SEM	Sig[1-3]
Item								
Pen (Steers)	5 (20)	5 (20)	5 (20)	5 (20)	5 (19)	5 (20)		
Initial BW, lbs	646	648	646	649	647	648	2.98	
Live Performance[4]								
Final BW, lbs	1167	1150	1175	1163	1100	1124	18.4	B**
ADG, lbs/d	3.52	3.43	3.69	3.51	3.10	3.15	0.09	B**
G:F	0.17	0.17	0.17	0.17	0.17	0.17	0.004	
DMI, lbs/d	19.8	19.6	20.4	19.1	17.2	18.2	0.48	B**C†
Carcass-adjusted Performance[5]								
Final BW, lbs	1182	1181	1195	1190	1119	1139	21.5	B**
ADG, lbs/d	3.78	3.67	4.01	3.73	3.43	3.64	0.12	B*
G:F	0.13	0.13	0.15	0.15	0.15	0.16	0.01	B†

[1]Sig: Significance of contrast statements

[2]Contrast Statements: A= Vitamin C vs. No Vitamin C; B = Linear effect of Sulfur; C = Vitamin C within High Sulfur; D = Vitamin C within Sulfur; E = Vitamin C within Corn Diet

[3]†($P \leq 0.10$);*($P \leq 0.05$);**($P < 0.01$)

[4]Live Performance values based on measured live BW and a 4% pencil shrink

[5]Carcass Adjusted Performance values calculated final BW from hot carcass weight divided by the average dressing percent of 64% for all treatments

Table 3. Effect of supplemental vitamin C on carcass characteristics of steers fed low, medium, or high S diets.

Diet Sulfur	Low		Medium		High			
Diet Vit C	-	+	-	+	-	+	SEM	Sig[1-3]
Pen (Steers)	5 (20)	5 (20)	5 (20)	5 (20)	5 (19)	5 (20)		
Calc. YG	3.45	3.28	3.26	3.20	2.90	3.16	0.14	B*
QG[5]	3.48	3.18	3.48	3.03	2.59	3.52	0.18	C**
KPH	2.10	2.34	2.17	2.36	2.22	2.18	0.05	A**E**
Marbling score[4]	514	458	474	438	398	470	14.5	B**C**E*
REA, in[2]	12.49	12.39	12.68	12.68	12.73	12.23	0.17	C*
Fat, in	0.55	0.50	0.50	0.52	0.39	0.48	0.02	B**C**D*
HCW, lbs	748	748	757	752	706	719	13.6	B**

[1]Sig: Significance of Contrast Statement

[2]Contrast Statements: A= Vitamin C vs. No Vitamin C; B = Linear effect of Sulfur; C = Vitamin C within High Sulfur; D = Vitamin C within Sulfur; E = Vitamin C within Corn Diet

[3]†($P \leq 0.10$);* ($P \leq 0.05$);**($P < 0.01$)

[4]Marbling Scores, traces: 200, slight: 300, small: 400, modest: 500, moderate: 600

[5] Quality Grade refers to the plant assigned QG

Table 4. Effect of vitamin C on fatty acid percentages and ratios of cattle fed a low, medium, or high dietary S diet.

Diet Sulfur	Low		Medium		High			
Diet Vit C	-	+	-	+	-	+	SEM	Sig[1-3]
Item								
Steers/trt	4	5	5	5	5	4		
SFA, %	45.38	44.78	44.29	43.21	45.59	42.96	0.96	A†B†C*
MUFA, %	42.57	48.86	47.18	47.85	47.12	47.65	1.43	A†E*
PUFA, %	7.08	6.69	8.53	8.50	8.84	10.05	0.80	B**
PUFA:SFA	0.19	0.15	0.19	0.19	0.20	0.24	0.02	B*
MCFA, %	3.42	3.89	3.39	3.52	3.25	3.43	0.22	
LCFA, %	96.43	96.13	96.21	96.04	96.02	96.12	0.28	
n3, %	0.37	0.50	0.53	0.54	0.52	0.48	0.04	E*
n6, %	6.71	6.19	8.00	7.92	8.31	9.55	0.72	B**
n3:n6	0.05	0.08	0.06	0.07	0.06	0.05	0.004	A†B*E**
AI	0.66	0.67	0.63	0.62	0.62	0.63	0.03	

[1]Sig: Significance of Contrast Statement

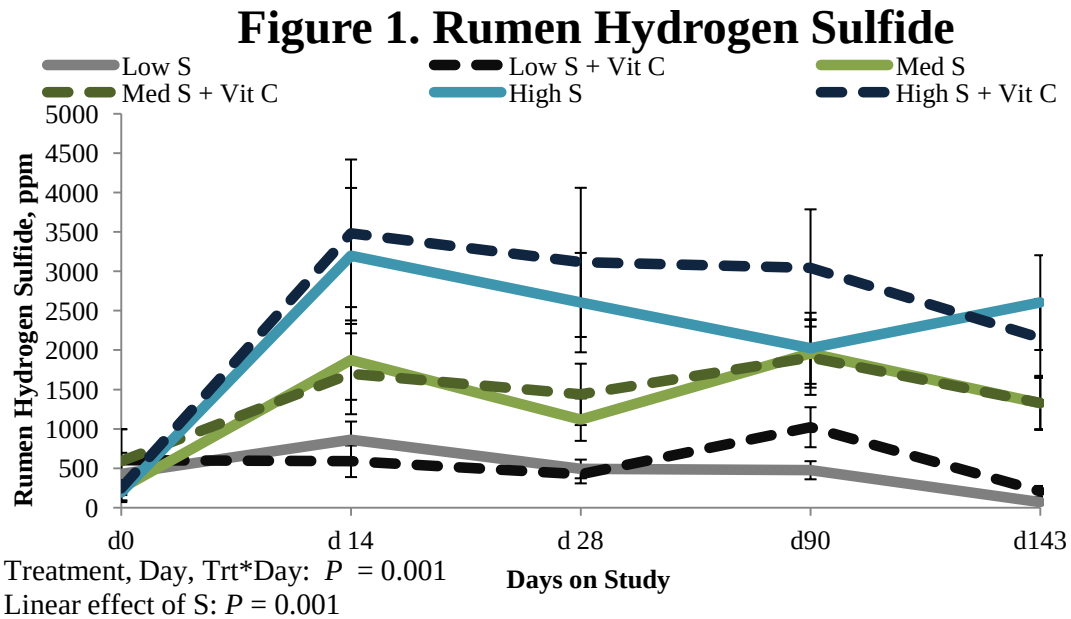
[2]Contrast Statements: A= Vitamin C vs. No Vitamin C; B = Linear effect of Sulfur; C = Vitamin C within High Sulfur; D = Vitamin C within Sulfur; E = Vitamin C within Corn Diet

[3]†($P \leq 0.10$);*($P \leq 0.05$);**($P < 0.01$)

Table 5. Effect of vitamin C on sulfhemoglobin concentrations of cattle fed a low, medium or high S diet.

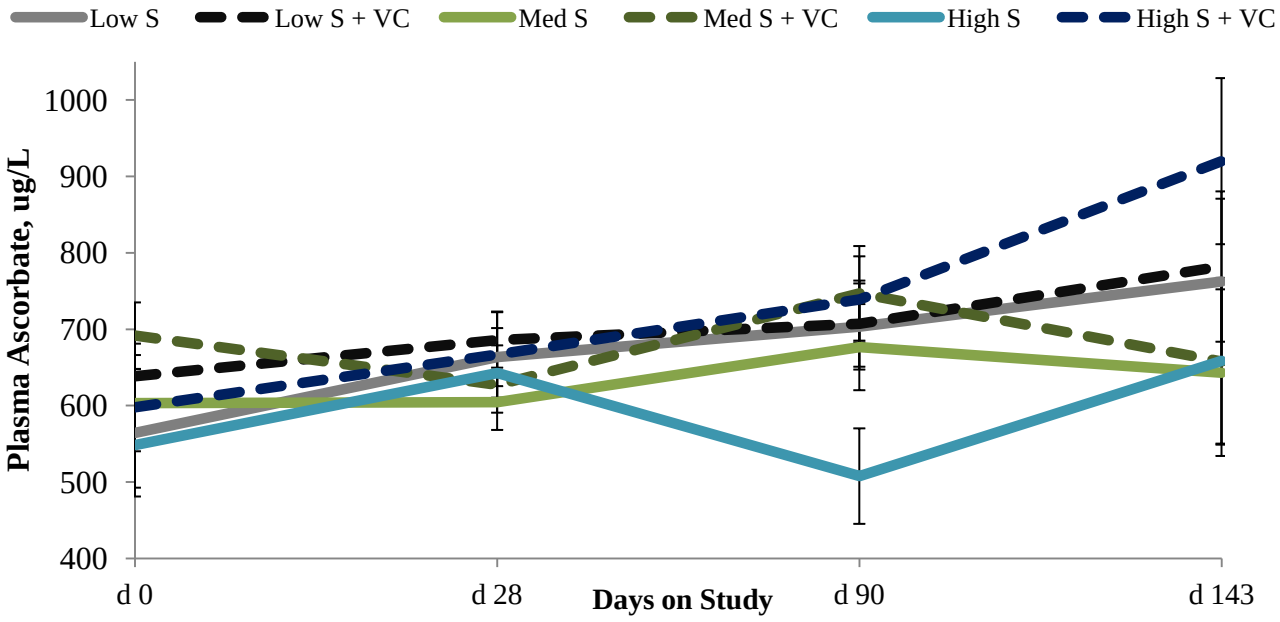
Diet Sulfur	Low		Medium		High			
Diet Vit C	-	+	-	+	-	+	SEM	Sig[1-3]
Item								
Sulfhemoglobin, mg/dl	31.5	36.6	42.0	49.4	51.0	60.7	3.77	A**B**C†D*
Total hemoglobin, g/dl	13.6	13.8	13.7	13.5	13.8	13.1	0.52	
Sulfhemoglobin[4], %	0.24	0.31	0.38	0.37	0.36	0.46	0.05	B**

[1]Sig: Significance of Contrast Statement
[2]Contrast Statements: A= Vitamin C vs. No Vitamin C; B = Linear effect of Sulfur; C = Vitamin C within High Sulfur; D = Vitamin C within Sulfur; E = Vitamin C within Corn Diet
[3]†($P \leq 0.10$);*($P \leq 0.05$);**($P < 0.01$)
[4]Sulfhemoglobin as a percentage of total hemoglobin



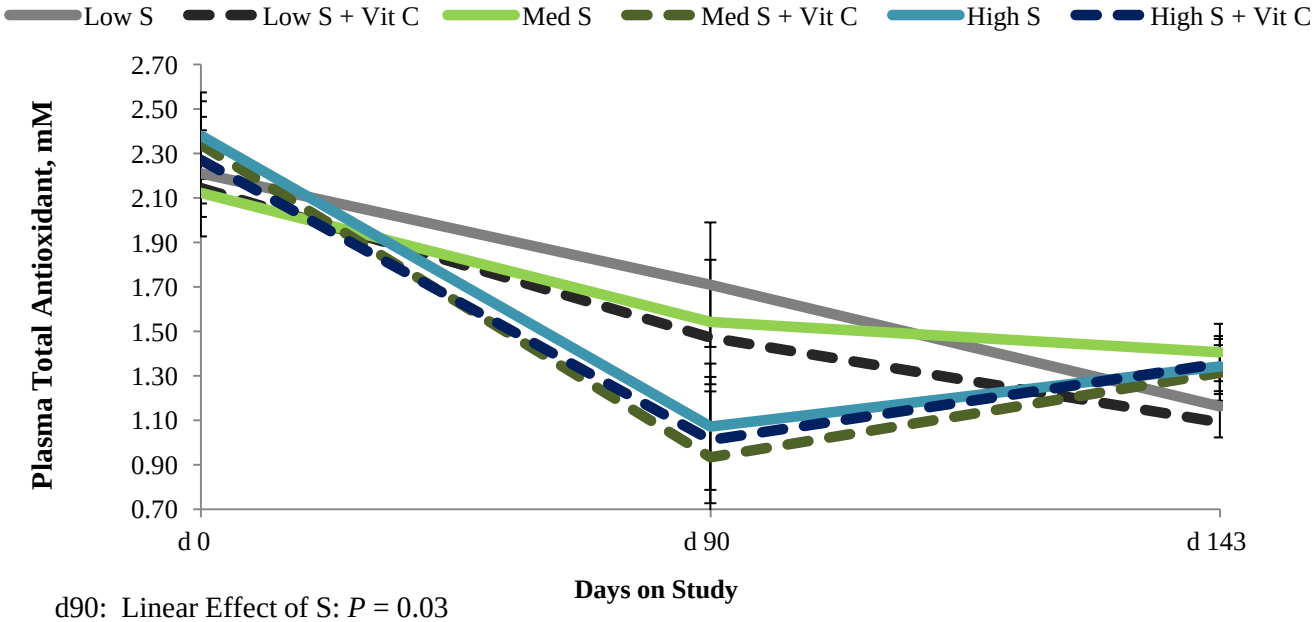
Treatment, Day, Trt*Day: P = 0.001
Linear effect of S: P = 0.001

Figure 2. Plasma Vitamin C



Treatment: *P* = 0.08
Vit C vs No Vit C: *P* = 0.04; Vit C within High S: *P* = 0.01; Vit C within Sulfur: *P* = 0.02

Figure 3. Plasma Total Antioxidant Capacity



d90: Linear Effect of S: *P* = 0.03
d143: Linear Effect of S: *P* = 0.05