

# **Effect of Feeding 2,500, 50,000 or 100,000 IU of Vitamin D<sub>3</sub> Daily on Feedlot Performance, Carcass Merit, and Plasma and Tissue Metabolite Concentrations**

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## **Story in Brief**

One hundred eighty yearling steers (initial BW =  $357 \pm 28$  kg) were used in a randomized complete block design to determine the effects of supplementing vitamin D<sub>3</sub> throughout the finishing phase on feedlot performance, carcass merit, and plasma and tissue metabolite concentrations. Vitamin D<sub>3</sub> was supplemented at 2,500 (control), 50,000 or 100,000 IU·steer<sup>-1</sup>·day<sup>-1</sup> over the entire 175-d (avg) finishing phase. Dry matter intake did not differ among treatments; calculated vitamin D<sub>3</sub> intakes based on lab assay were 3,607, 27,506 and 70,075 IU·steer<sup>-1</sup>·d<sup>-1</sup>. Final BW and ADG did not differ among treatments. Vitamin D<sub>3</sub> supplementation did not affect hot carcass weight, carcass characteristics or lean and skeletal maturity. Plasma D<sub>3</sub> (5.3, 17.3,  $30.9 \pm 0.8$  ng/mL) and 25-hydroxyvitamin D<sub>3</sub> (68.1, 97.0, and  $117.0 \pm 3.5$  ng/mL) concentrations increased ( $P < 0.001$ ) as level of vitamin D<sub>3</sub> supplementation increased, whereas plasma 1,25-dihydroxyvitamin D<sub>3</sub> level numerically ( $P = .13$ ) increased. Liver and muscle levels of vitamin D<sub>3</sub> and metabolites did not differ among treatments. Vitamin D<sub>3</sub> can be supplemented over the entire finishing period without negatively affecting feedlot performance and carcass characteristics. In addition, numerical increases in metabolite concentrations of tissue and plasma samples indicate that calcium concentrations within the body can be safely increased with low levels of vitamin D<sub>3</sub> supplementation over extended periods of time.

Key Words: Beef, Calcium, Tenderness, Vitamin D

## **Introduction**

Supplementing vitamin D<sub>3</sub> to beef cattle with the intent of increasing beef tenderness has been debated within the literature despite considerable investigation. The hypothesis has been that increased dietary vitamin D<sub>3</sub> increases the amount of calcium within the body. Upon harvest, the increased calcium is then available to more fully activate the calpain proteolytic system, increasing beef tenderness. Some data has suggested that high doses of vitamin D<sub>3</sub> (5 to 8 million IU·animal<sup>-1</sup>·d<sup>-1</sup>) fed for short periods of time (5 to 10 d) before slaughter improved Warner-Bratzler shear force of specific cooked beef cuts (Swanek et al., 1999; Montgomery et al., 2000, 2002). However, vitamin D<sub>3</sub> supplementation at high levels results in lower DMI, which may subsequently cause lower final live BW and hot carcass weight (HCW) (Karges et al., 1999 and 2001; Scanga et al., 2001). Additionally, high levels of vitamin D<sub>3</sub> supplementation increase the concentration of vitamin D<sub>3</sub> (or its metabolites) within the meat to potentially toxic levels (Montgomery et al., 2000, 2002; Foote et al., 2004). We hypothesized that vitamin D<sub>3</sub> continuously fed at lower levels (50,000 and 100,000 IU animal<sup>-1</sup>·d<sup>-1</sup>) would improve tenderness without negative effects on intake, BW and HCW.

## **Materials and Methods**

**Animals.** A total of 180 yearling steers (initial BW =  $357 \pm 28$  kg) were received in three loads at the Willard Sparks Beef Research Center in May, 2003. On arrival (d 0), steers were individually weighed and identified with an individual ear tag. Based on initial BW, steers were stratified into three groups (60 animals per group) by weight and randomly assigned within weight block to 6 pens of 10 steers each. One third of the pens in each block (n=2) were randomly assigned to one of three treatments: 2,500 IU (control), 50,000 IU, or 100,000 IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup>. To decrease bias of cattle origin, cattle originating from a different source were equally distributed among pens and treatment groups.

**Processing.** Steers were processed (d 1) the day after arrival. Individual weights were recorded and each steer received the following: vaccination with Titanium 5 L5<sup>TM1</sup> and Vision 7 with SPUR<sup>TM2</sup> (2 mL each, sub-Q; Intervet Inc., Millsboro, DE); treatment with anthelmintics for internal and external parasites (7 mL sub-Q; Ivomec-Plus<sup>TM3</sup>, Merial Limited, Iselin, NJ); and implantation with Revalor-S<sup>TM4</sup> (20 mg trenbolone acetate, 4 mg estradiol; Intervet, Inc.). Steers were reimplanted with Revalor-S on d 70. Subsequent BW (unshrunk) were taken on d 35, 70, 105, 141, and 176. Additionally, steers in the heavy block (n = 60) had blood samples drawn on d 176 by venous puncture from the jugular into sterile 10 mL BD Vacutainer® [Beckton Dickinson & Co., Franklin Lakes, NJ] tubes containing sodium heparin. Plasma was then collected and frozen at -20°C for later analyses by USDA-ARS National Animal Disease Center.

**Diet.** All steers were stepped up with 4 adaptation diets (55, 70, 80, and 87% DM of concentrate for 8, 6, 7, and 6 d, respectively) to a final finishing diet consisting of (DM basis) 80.7% rolled corn, 8.0% ground alfalfa hay, 3.0% fat, and 8.3% pelleted vitamin D<sub>3</sub> supplement (Table 1). The OSU vitamin premix consisted of 19 parts fine ground corn to 1 part vitamin D<sub>3</sub>-500 [Roche Vitamins, Nutley, NJ] to dilute the pure vitamin D<sub>3</sub> to levels that would allow for the mixing of supplemental vitamin D<sub>3</sub> directly into the pelleted supplement.

**Table 1. Vitamin D<sub>3</sub> supplement ingredients (% DM)<sup>a</sup> by treatment level**

Supplement	2,500 IU	50,000 IU	100,000 IU
Soybean meal	23.45	23.45	23.45
Cottonseed meal	23.71	23.71	23.71
Wheat midds	23.05	22.83	22.61
Limestone 38%	16.22	16.22	16.22
Salt	3.79	3.79	3.79
Vitamin A – 30,000 IU	.13	.13	.13
Vitamin E – 50%	.08	.08	.08
Rumensin – 80b	.21	.21	.21
Zinc Sulfate	.05	.05	.05

Manganous oxide	.05	.05	.05
Copper sulfate	.01	.01	.01
Selenium – 600	.09	.09	.09
Urea	9.03	9.03	9.03
Tylan –40b	.12	.12	.12
OSU vitamin premix	.01	.23	.46

<sup>a</sup>Rumensin provided at the rate to supply 0.37 g/kg and Tylan provided at the rate to supply 0.11 g/kg

<sup>b</sup>Elanco Animal Health, Greenfield, IN

The listed treatment levels of 2,500 (control); 50,000 and 100,000 IU vitamin D<sub>3</sub> were established target levels of intake. Samples were taken from each batch of supplement and composited by month. Assay of the supplements (Dr. Jonathan Wilson, Nutritional Products, Inc., Parsippany, NJ) showed lower than expected levels of vitamin D<sub>3</sub>. Calculated average intake of vitamin D<sub>3</sub>, based on assay results, are shown in Table 2.

**Table 2. Calculated average daily intake of assayed vitamin D3 by target treatment**

Treatment	Assayed D <sub>3</sub> level, IU/kg	Total D <sub>3</sub> intake, IU/d
2,500 IU	4,748	3,607
50,000 IU	37,397	27,506
100,000 IU	88,998	70,075

**Slaughter.** Steers were determined to be at optimum finish by visual appraisal and were harvested based on weight block. The heavy and intermediate blocks were harvested together on d 146, while the light block was harvested on day 181. All groups were harvested at Tyson Fresh Meats in Emporia, Kansas. Oklahoma State University personnel accompanied cattle to the plant to collect HCW, REA, marbling score, fat thickness, KPH estimates, lean and skeletal maturity, and USDA quality and yield grade on all harvest groups. Longissimus steak samples, as well as kidney and liver tissue samples, were collected from the heavy block of steers (n = 60) and frozen for later analysis. Analyses of steak, kidney, liver, and plasma samples were conducted by USDA-ARS National Animal Disease Center.

**Statistical Analysis.** Performance and carcass data were analyzed as a randomized complete block design with pen serving as the experimental unit. The PROC MIXED procedure of SAS was used to determine means and standard errors of means, with treatment level of vitamin D<sub>3</sub> and block as fixed effects. For tissue and plasma samples, individual animal was considered the experimental unit. The PROC MIXED procedure of SAS was used, but with load and treatment considered as main effects, and pen and the load\*pen interaction considered as random effects.

## Results and Discussion

**Feedlot Performance.** Beginning and interim BW, average daily gain (ADG), feed efficiency, and DMI are reported in Table 3. At the initiation of the experiment BW did not differ among treatments. Final BW for steers fed 100,000 IU of vitamin D<sub>3</sub> was 11 kg greater compared with control steers. However, final BW did not differ among treatments. By decreasing the level of vitamin D<sub>3</sub> supplementation from previously reported levels, we were able to eliminate decreases in final body weight observed by Karges et al. (2001), Scanga et al. (2001), and Montgomery et al. (2002).

**Table 3. Effect of vitamin D3 supplementation on feedlot performance**

Item	2,500 IU	50,000 IU	100,000 IU	SEM
Pens	6	6	6	-
Inwt, kg	356	357	356	.47
Final BW, kg	593	589	602	4.28
Carcass adj. BW, kg <sup>a</sup>	610	618	616	3.49
DMI d 0 – finish, kg/d	9.41	9.50	9.63	.18
ADG d 0 – finish, kg/d	1.49	1.53	1.56	.03
Gain:Feed d 0 – finish, kg/kg	.158	.162	.162	.002
Carcass adj. ADG, kg/d	1.56	1.60	1.60	.02
Carcass adj. gain:feed, kg/kg	.174	.181	.180	.003

<sup>a</sup>Carcass adj. BW calculated by dividing HCW by average dressing percent of each block

Average daily gain was calculated by weigh period and by overall time on feed, based on a 4% pencil shrink applied to interim and final BW. In contrast to Scanga et al. (2001) and Montgomery et al (2002), no difference in ADG among treatments was observed. Scanga et al. (2001) reported cattle that received greater than  $10 \times 10^6$  IU of vitamin D<sub>3</sub> over an 8-d period had lower ( $P < .05$ ) ADG than negative control cattle, and cattle that received  $10 \times 10^6$  IU of vitamin D<sub>3</sub> or less over the same 8-d period had intermediate ADG that did not differ from control cattle or cattle receiving the higher dose of supplementation. Montgomery et al. (2002) observed similar findings and reported vitamin D<sub>3</sub> treatment linearly decreased ( $P < .01$ ) ADG across the last 21 d of feeding with supplementation rates of 5 and  $7.5 \times 10^6$  IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup>, resulting in negative ADG that differed ( $P = .02$ ) from those of steers treated with  $1 \times 10^6$  IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup>.

No difference in DMI was observed among treatments during any period. This agrees with Montgomery et al. (2002) who reported no difference in daily feed intake with vitamin D<sub>3</sub>

supplementation. However, Montgomery et al. (2002) did report a vitamin D<sub>3</sub> supplementation x day interaction (P<.002) when feed intake was measured during a 9-d supplementation period; supplementing steers with 2.5, 5, or 7.5 x10<sup>6</sup> vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup> decreased feed intake during d 7 and 8 compared with that of control steers (P<.05). Similarly, Scanga et al. (2001) reported that following d 2 of supplementation with vitamin D<sub>3</sub>, the appetite of cattle receiving more than 1 x 10<sup>6</sup> IU vitamin D<sub>3</sub>/d declined. Karges et al. (2001) also reported numerically lower DMI for steers supplemented with vitamin D<sub>3</sub>. In the present study, no differences in efficiency (ADG:DMI) were observed when vitamin D<sub>3</sub> levels of 2,500, 50,000, and 100,000 IU/hd/d were fed over the entire finishing period.

**Carcass Merit.** Effects of vitamin D<sub>3</sub> supplementation on carcass characteristics are shown in Table 4. Supplementation did not affect carcass yield, quality, or maturity traits as expected since no difference in feedlot performance was observed. Montgomery et al. (2002) reported that hot carcass weight and dressing percentage were not affected by vitamin D<sub>3</sub> supplementation for 9 d despite supplementation effects on ADG and feed intake.

**Table 4. Effect of vitamin D<sub>3</sub> supplementation on carcass characteristics**

Item	2,500 IU	50,000 IU	100,000 IU	SEM
Hot carcass weight, kg	384	389	388	2.19
12th rib fat thickness, cm	1.14	1.17	1.17	.04
Longissimus muscle area, cm <sup>2</sup>	87.92	86.18	87.40	1.33
Kidney, pelvic, and heart fat, %	2.2	2.2	2.3	.10
Marbling <sup>a</sup>	380	376	380	7.04
Lean maturity <sup>b</sup>	172	180	174	3.08
Skeletal maturity <sup>b</sup>	159	158	156	3.69
USDA yield grade	2.3	2.4	2.2	.08
USDA quality grade <sup>c</sup>	2.7	2.6	2.7	.05

<sup>a</sup>Marbling score: 300 = slight, 400 = small<sup>00</sup>

<sup>b</sup>Maturity score: 100 = A, 200 = B

<sup>c</sup>USDA quality grade: 3 = select, 2 = choice

**Plasma and Tissue Concentrations.** As shown in Table 5, vitamin D<sub>3</sub> supplementation at 50,000 and 100,000 IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup> significantly increased (P<.05) plasma vitamin D<sub>3</sub> and 25-hydroxyvitamin D<sub>3</sub> concentrations. Plasma concentrations of vitamin D<sub>3</sub> and 25-hydroxyvitamin D<sub>3</sub> were 5.8– and 1.7– fold greater in cattle fed 100,000 IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup> compared with steers fed the control diet (2,500 IU vitamin D<sub>3</sub>·steer<sup>-1</sup>·d<sup>-1</sup>). Additionally, vitamin D<sub>3</sub> and 25-hydroxyvitamin D<sub>3</sub> concentrations in kidney tissue increased (P<.05) with increasing

level of supplementation. Concentrations of the biologically active form of vitamin D<sub>3</sub>, 1,25-dihydroxyvitamin D<sub>3</sub>, were numerically greatest in plasma, liver tissue, and muscle tissue.

**Table 5. Effect of treatment on vitamin D<sub>3</sub> and metabolite concentrations in plasma and tissue**

	2,500 IU	50,000 IU	100,000 IU	SEM	P>F
Plasma	60	60	60		
<sup>a</sup> D <sub>3</sub> (ng/g)	5.32 <sup>a</sup>	17.25 <sup>b</sup>	30.19 <sup>c</sup>	.82	<.0001
<sup>b</sup> 25D <sub>3</sub> (ng/g)	68.08 <sup>a</sup>	96.96 <sup>b</sup>	116.86 <sup>c</sup>	3.54	<.0001
<sup>c</sup> 1,25D <sub>3</sub> (pg/g)	53.81	46.63	60.15	4.68	.13
Ca (mg%)	8.92	9.06	9.24	.15	.30
Mg (mg%)	1.69	1.68	1.73	.07	.89
Liver	44	44	41		
<sup>a</sup> D <sub>3</sub> (ng/g)	38.11	45.93	47.11	3.79	.12
<sup>b</sup> 25D <sub>3</sub> (ng/g)	6.95	8.52	13.58	2.87	.35
<sup>c</sup> 1,25D <sub>3</sub> (pg/g)	138.20	97.28	134.05	18.29	.22
Muscle	60	59	60		
<sup>a</sup> D <sub>3</sub> (ng/g)	15.70	16.41	15.90	.89	.85
<sup>b</sup> 25D <sub>3</sub> (ng/g)	1.44 <sup>a</sup>	1.80 <sup>ab</sup>	2.27 <sup>b</sup>	.18	.10
<sup>c</sup> 1,25D <sub>3</sub> (pg/g)	51.07	61.02	69.33	6.71	.19
Kidney	57	57	41		
<sup>a</sup> D <sub>3</sub> (ng/g)	5.51 <sup>a</sup>	25.05 <sup>b</sup>	39.71 <sup>c</sup>	3.75	<.0001
<sup>b</sup> 25D <sub>3</sub> (ng/g)	6.50 <sup>a</sup>	9.28 <sup>b</sup>	11.07 <sup>b</sup>	0.54	<.0001
<sup>c</sup> 1,25D <sub>3</sub> (pg/g)	126.95	156.17	131.82	37.28	.83

<sup>a</sup>Vitamin D<sub>3</sub>.

<sup>b</sup>25-hydroxyvitamin D<sub>3</sub>.

<sup>c</sup>1,25-dihydroxyvitamin D<sub>3</sub>.

<sup>abc</sup>Means with different subscripts differ,  $P < 0.05$

Calcium concentrations in plasma numerically increased as level of vitamin D<sub>3</sub> supplementation increased. This supports previous work by Karges et al. (1999), Montgomery et al (1999), and Swanek et al (1999) who reported increased plasma calcium concentrations with increased vitamin D<sub>3</sub> supplementation. Karges et al. (2001) reported blood plasma calcium concentrations were significantly greater ( $P < .03$ ) for animals supplemented with  $6 \times 10^6$  IU of vitamin D<sub>3</sub> daily for 4 or 6 d before harvest, with cattle supplemented for 6 d having greater plasma calcium concentrations than those supplemented for 4 d.

### **Implications**

Low levels (50,000 to 100,000 IU·steer<sup>-1</sup>·d<sup>-1</sup>) of vitamin D<sub>3</sub> can be supplemented throughout the finishing period to increase plasma calcium concentrations, as well as concentrations of vitamin D<sub>3</sub> and 25-hydroxyvitamin D<sub>3</sub> in plasma and kidney, without affecting feedlot performance or carcass merit. Based on the numerical increase of vitamin D<sub>3</sub> and its metabolites in other tissue samples with increasing level of supplementation, more data are needed to determine at what level, if any, vitamin D<sub>3</sub> can have a positive effect on beef tenderness when fed over the entire finishing period.

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