

# Determination of Appropriate Vitamin E Supplementation Levels and Administration Times to Ensure Adequate Muscle Tissue $\alpha$ -tocopherol Concentrations in Cattle Destined for the Nolan Ryan Tender-Aged Beef (NRTAB) Program.

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## SUMMARY

Retail caselife for beef products has been a major concern for various segments of the beef industry. Several studies have shown that dietary Vitamin E supplementation prior to slaughter has provided a vital antioxidant to help increase retail meat case life in beef products. This study demonstrated that  $\alpha$ -tocopherol concentrations in the *rectus capiti dorsalis major* (RCDM) muscle increased as more Vitamin E was fed to cattle, and determined that the recommended 500 IU/head/day for the last 100 days prior to slaughter was sufficient to maintain caselife for periods longer than two days in a retail meat case.

**Key Words:**  $\alpha$ -tocopherol, Beef, Vitamin E

## INTRODUCTION

Previous research has established that Vitamin E supplementation substantially reduces rates of lipid and muscle carrier-protein (myoglobin) oxidation that, in turn, slows the rate of development of rancid, off-flavor conditions in beef displayed in retail meat cases and enhances retail display color desirability and stability (Faustman, et al., 1998). Research at the University of Wisconsin suggests that muscle  $\alpha$ -tocopherol concentrations must be between 3.0-3.7  $\mu\text{g/g}$  wet weight to effectively

produce these positive case life attributes in fresh beef.

The Nolan Ryan Tender Aged Beef (NRTAB) program markets cattle in Texas and requires in their USDA-certified program that cattle be fed 500 IU Vitamin E/head/day for 100 days prior to slaughter. Recently, concerns have developed relative to the dose and duration of Vitamin E supplementation that the cattle being fed to meet NRTAB specifications, (1) increased supplementation beyond the recommended level and (2) that it may be important to standardize the amount of time for which NRTAB cattle receive Vitamin E supplementation.

The purpose of this study was to (1) establish the total quantity of Vitamin E supplementation necessary for cattle, destined for the NRTAB program, to achieve a minimum muscle  $\alpha$ -tocopherol concentration of 3.2  $\mu\text{g/g}$  and (2) to establish the time required, at differing levels of Vitamin E supplementation, to achieve the same minimum muscle  $\alpha$ -tocopherol concentration in NRTAB cattle fed in south Texas.

## MATERIALS AND METHODS

To address the original objectives of this study, vitamin E concentrations in feed and fed cattle muscle tissue were monitored as feedlot cattle were "ramped-up" to supranutritional dietary concentrations of Vitamin E supplementation. Feedlot cattle were from south Texas and while in the feedlot were to have been fed a minimal to no amount of Vitamin E prior to the ramp-up phase. Feedlot cattle at Perry Feeders (Pleasanton, Texas) were ramped up 7 days to a targeted supplementation level of 1,000 International Units (IU) of Vitamin E (as  $\alpha$ -tocopheryl acetate) for approximately 160 days. Vitamin E was incorporated into the diet as a pre-mixed supplement; supplementation occurred at a rate of 5.6% of the total finishing ration.

## Feed Sampling

Duplicate feed samples ( $n = 5$ ) from 5 different pens were collected

immediately following feed delivery to the feed bunks on day 0, and every 25 days thereafter. Using a 5-lb. bucket, random samples were collected from the feed bunk, thoroughly mixed, and two random samples (1 pound) were pulled from the bucket. Collected samples were frozen (28°F) and shipped overnight to the University of Florida-Gainesville where  $\alpha$ -tocopheryl acetate concentration was determined via High Pressure Liquid Chromatography (HPLC) as described by Njeru et al. (1995) and Wilkinson (2000).

## Tissue Sampling

Within 30 minutes post-mortem, duplicate muscle tissue samples (RCDM) were excised from the same side of ten randomly selected carcasses from various lots of cattle coming from Perry Feeders. Samples were collected at day 0, and every 25 days thereafter (from cattle on the study ready for slaughter) following initiation of the ramp-up study, coinciding with the collection of feed samples. Immediately following excision, the collected samples were placed in Whirl-pak bags, labeled, frozen (21°F), and shipped overnight to the University of Florida-Gainesville for muscle  $\alpha$ -tocopherol concentration determination via HPLC (Njeru *et al.*, 1995; Wilkinson, 2000).

## Statistical Analysis

Data were analyzed using regression procedures of SAS (1999) to predict the number of days it would take in order to achieve an  $\alpha$ -tocopherol concentration of 3.2  $\mu\text{g/g}$  muscle tissue while feeding 1,000 IU Vitamin E-head<sup>-1</sup>·day<sup>-1</sup>. A regression line also was developed for the purpose of demonstrating the homogeneity of  $\alpha$ -tocopherol concentrations in neck muscle tissue over a 125-day period and to show linear trends. Means and standard deviations were calculated using the GLM procedure of SAS (1999) and when appropriate, means were separated using Tukey's HSD method.

## RESULTS AND DISCUSSION

### Feed Samples

Feed assays of  $\alpha$ -tocopheryl acetate concentrations averaged 12.24 IU/lb for the basal feed samples and 37.7, 45.7, 38.3, and 41.5 IU/lb on days 26, 46, 74, and 100 of high E supplementation, respectively. Calculated  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  of Vitamin E by sampling date are shown in Table 1. Daily Vitamin E intake was much higher than the industry average of 50 IU  $\alpha$ -tocopheryl acetate per head/day prior to day 0 (Table 1). Basal feed samples indicated that cattle received approximately 43,500 IU of Vitamin E/head over an average of 136 days of feeding prior to initiation of the ramp-up period. Total Vitamin E intake for cattle following 100 days of high Vitamin E supplementation over the ramp-up period averaged 125,370  $\text{IU}\cdot\text{head}^{-1}$ . The average Vitamin E intake for each collection time and the total amount consumed over time is shown in Table 1 and illustrated in Figure 1.

### Tissue Samples

Tissue samples collected at day 0 were obtained from carcasses of cattle that had averaged 136 days on feed (DOF), and based upon the initial feed analysis, consumed approximately 43,500 IU of Vitamin E over that time period. Tissue levels of  $\alpha$ -tocopherol from carcasses samples at day 0 averaged 3.7  $\mu\text{g/g}$ , which were higher than results observed by Westcott *et al.* (2000), who reported muscle tissue concentrations of 3.1  $\mu\text{g/g}$  of  $\alpha$ -tocopherol when cattle receive 50,000 IU of Vitamin E per 100 days. The significance of the rise in muscle  $\alpha$ -tocopherol concentrations between samples collected at day 0 and samples collected at day 25 suggested that between 43,459 and 66,091  $\text{IU}\cdot\text{head}^{-1}$  must be fed over 100 days in order to increase muscle  $\alpha$ -tocopherol concentrations to 3.2  $\mu\text{g/g}$  to achieve the desired shelf-life effects needed for beef retail display. The present study suggested that dietary Vitamin E supplementation over an extended period of time prior to the ramp-up phase to a (higher Vitamin E

supplementation) may result in higher  $\alpha$ -tocopherol concentrations in muscle tissue compared to those achieved in cattle previously receiving fresh forage for a period of time (Faustman, *et al.*, 1998) prior to ramp-up phase of high dietary Vitamin E supplementation.

Concentrations of  $\alpha$ -tocopherol in the RCDM averaged 3.7, 6.6, 6.8, 6.4, 8.9, and 9.3  $\mu\text{g/g}$  muscle tissue on days 0, 25, 46, 74, 100, and 132 days, respectively. The 132-day muscle tissue samples were collected to determine the point of maximum  $\alpha$ -tocopherol accretion. Concentrations of  $\alpha$ -tocopherol in the RCDM muscle increased ( $P<0.05$ ) over time between days 0 to 26, and between days 74 to 100 (Figure 2). Because initial muscle  $\alpha$ -tocopherol concentrations exceeded the recommended concentration for improved shelf life (3.2-3.7  $\mu\text{g/g}$  tissue) prior to ramp-up phase to high (1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$ ) Vitamin E supplementation (Figure 1), the recommended time that cattle in the NRTAB Program should receive 1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  to attain the minimum level of  $\alpha$ -tocopherol required to insure maximum shelf-life in the RCDM muscle was not able to be determined.

Using a regression procedure in SAS (1999) with a minimum  $\alpha$ -tocopherol concentration level of 3.2  $\mu\text{g/g}$  tissue as the target RCDM concentration to insure maximum shelf life, a minimum of 59 days of supplementation with 1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  would be required for NRTAB Program cattle to elevate RCDM muscle Vitamin E concentrations to a minimum level of 3.2  $\mu\text{g/g}$  of  $\alpha$ -tocopherol/g. Assuming  $\alpha$ -tocopherol concentration was 0  $\mu\text{g/g}$  in the muscle, this predicted number of days would give the required minimum rise in  $\alpha$ -tocopherol concentration in the muscle if cattle could not be fed 1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  for 100 days prior to slaughter. These results were similar to those of Sanders *et al.* (1997), who observed elevated levels of  $\alpha$ -tocopherol in *longissimus dorsi* muscle tissue following 1,000 and 2,000

$\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  Vitamin E for 100 days. Roeber *et al.* (2001) conducted a similar study while feeding 1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  for 100 days and sampled inside round steaks, T-bone steaks (strip loin and tenderloin separate), top sirloin steaks, and ground beef for product performance during retail display;  $\alpha$ -tocopherol concentrations in each sample were as follows: 5.4, 4.2, 6.8, and 7.2  $\mu\text{g/g}$ , respectively. Moreover, after feeding 500  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  for a minimum 100 days, Zerby *et al.* (1999) observed elevated levels of  $\alpha$ -tocopherol concentrations in *biceps femoris* (3.9  $\mu\text{g/g}$ ), *triceps brachii* (4.0  $\mu\text{g/g}$ ), *semitendinosus* (3.7  $\mu\text{g/g}$ ), *semimembranosus* (3.1  $\mu\text{g/g}$ ), *longissimus dorsi* (3.2  $\mu\text{g/g}$ ), *psaos major* (3.4  $\mu\text{g/g}$ ), and ground beef (4.7  $\mu\text{g/g}$ ). Consequently,  $\alpha$ -tocopherol accumulation is dependent on muscle type (Arnold *et al.*, 1993a, 1993b; Roeber *et al.* 2001); comparisons of  $\alpha$ -tocopherol concentrations in muscle tissue in this study with those of Sanders *et al.* (1997) should be made with caution.

## CONCLUSIONS

Supplementation of  $\alpha$ -tocopheryl acetate at a targeted level of 1,000  $\text{IU}\cdot\text{head}^{-1}\cdot\text{day}^{-1}$  for 59 days is an effective way of feeding dietary Vitamin E to attain the recommended level of 3.2-3.7  $\mu\text{g/g}$  tissue of  $\alpha$ -tocopherol in muscle tissue, and thus to improve the display life for beef products. Considering that cattle received total supplementation Vitamin E of 43,459  $\text{IU}\cdot\text{head}^{-1}$  before the initiation of this study, total intake after 26 days of Vitamin E supplementation during ramp-up in this study resulted in total dietary supplementation of 66,091  $\text{IU}\cdot\text{head}^{-1}$ . Since 90% of the cattle achieved the recommended level of 3.2-3.7  $\mu\text{g/g}$  tissue of  $\alpha$ -tocopherol by initiation of this study (day 0), NRTAB Program cattle should receive an estimated total of 43,459 and 66,091  $\text{IU}\cdot\text{head}^{-1}$  for 100 days to achieve the desired shelf-life effects.

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Table 1: Feedlot performance, Vitamin E intake levels, and RCDM  $\alpha$ -tocopherol concentrations before and after Vitamin E supplementation

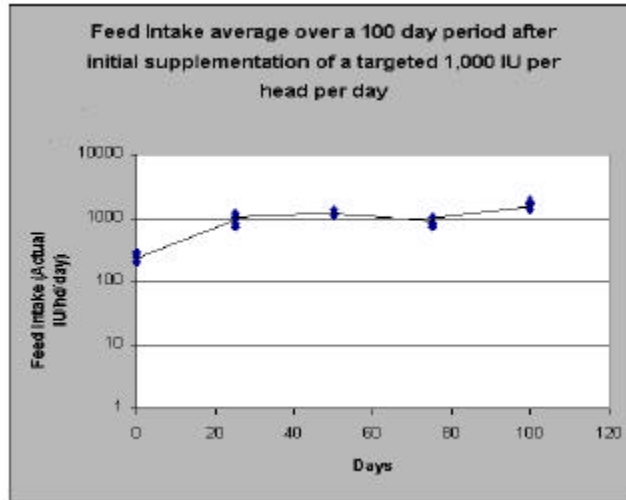
Characteristic	Sampling Intervals (days) with Means and Standard Deviations				
	0 <sup>a</sup>	26	46	74	100
Average wgt, lbs	1084.0 ±212.2	1214.4 ±127.5	1078.8 ±91.6	932.8 ±228.4	1076.20±122.6
Days on feed <sup>b</sup>	136 ±32.3	162 ±21.1	149±19.8	125±29.3	128±11.9
Feed Consumption, lbs/d	20.8 ±3.7	20.9 ±6.7	22.7 ±1.6	24.5 ±12.0	--- <sup>c</sup>
Vitamin E Feed Assays, IU/lb (N=25)	12.2	37.7	45.7	38.3	41.5
Calculated Vitamin E intake, IU/hd/d	319.5±41.6	802.5±194.9	1051.3±45.8	938.1±142.4	--- <sup>c</sup>
Total Vitamin E intake, IU/head	43,459	66,091	81,252	104,636	125,370
RCDM. Conc. $\alpha$ -toc. $\mu$ g/g (N=50)	3.7 <sup>x</sup> ±1.0	6.6 <sup>y</sup> ±1.2	6.8 <sup>y</sup> ±1.4	6.7 <sup>y</sup> ±1.0	9.3 <sup>z</sup> ±1.6

<sup>a</sup>Feed samples were taken prior to the initial supplementation of 1,000 IU  $\alpha$ -tocopheryl acetate per animal per day.

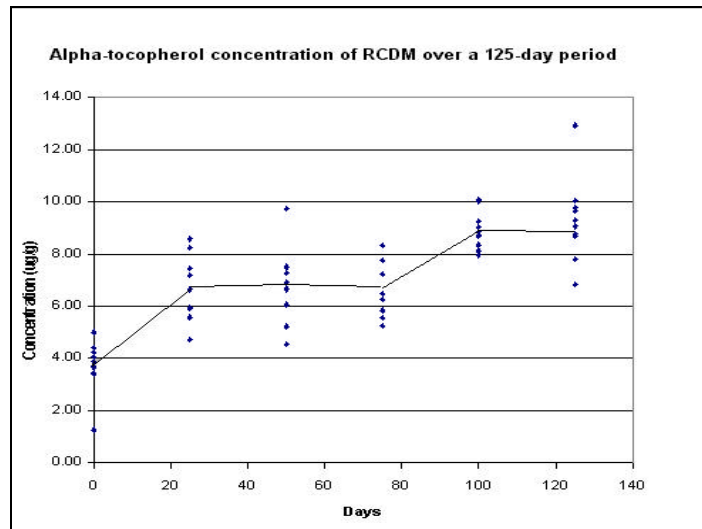
<sup>b</sup>Cattle had already been on some supplementation prior to 1,000 IU supplementation, and different pens were sampled from during each sampling period.

<sup>c</sup>Data provided for feed consumption was roughly estimated and not presented.

<sup>x,y,z</sup> Means with different letters are significantly different.



**Figure 1:** Day 0 is prior to any supplementation of the targeted 1,000 IU of Vitamin E  $\cdot$ head<sup>-1</sup>  $\cdot$ day<sup>-1</sup>, and the days thereafter represent the actual intake of Vitamin E over a 100-day period. Day 132 samples were not collected because of time constraints.



**Figure 2:** Concentrations of  $\alpha$ -tocopherol averaged 3.7- $\mu$ g/g-muscle tissue prior to initial supplementation, then steadily increased and then peaked, with still a larger increase at 100 to 125 days.