Influence of supplemental tocopherol level (0, 250 and 500 IU RRR-α-tocopherol/d/steer) and injectable retinol form (retinyl propionate vs retinyl palmitate) on growth performance, carcass characteristics and plasma concentration in calf-fed Holstein steers

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ABSTRACT

The influence of supplemental tocopherol level (0, 250 and 500 IU RRR-α-tocopherol/d/steer) and injectable retinol form (retinyl propionate vs retinyl palmitate) on growth performance, carcass characteristics and plasma tocopherol and retinol concentrations were evaluated in 108 Holstein steers fed a steam flaked corn-based finishing diet during 314-d feeding period. There were no treatment interactions (P > 0.10). During the initial 112-d period, dietary supplemental tocopherol tended to increase ADG (linear, P = 0.07) and DMI (linear, P = 0.06). Overall 314-d ADG, DMI, gain efficiency and carcass characteristics were not affected (P > 0.10) by dietary supplemental tocopherol. Overall DMI tended to be greater (3%, P ≤ 0.10) for steers injected with retinyl palmitate vs retinyl propionate. It is concluded that vitamin E supplementation above basal requirements may enhance growth performance during the initial 112-d phase. However, the overall effect on growth performance and carcass characteristics was not appreciable. Injectable retinol ester form did not affect overall ADG, gain efficiency, or dietary NE. Based on plasma retinol concentrations, the bioavailability of retinyl palmitate is greater than that of retinyl propionate.

1. Introduction

Vitamin requirements (NASEM 2016) are influenced by intrinsic factors including age, weight, health and growth-performance, and by external factor including diet composition (interactions with among dietary components), and feed processing (McDowell 2000). The numerous factors involved make assessments of nutritional requirement difficult. Based on NASEM (2016), the retinol requirement for feedlot cattle is 2200 IU/kg DM. In practice, vitamin A is typically supplemented to meet or exceed that requirement, regardless of the vitamin A equivalent content of the basal diet, per se (Vasconcelos and Galyean 2007). The basis for this practice is not certain, but may be related to the potential for extensive (70–80%) losses related to ruminal degradation (Rode et al. 1990; Weiss et al. 1995). The practical alternative to dietary vitamin A is to provide it in injectable form as retinol esters (Perry et al. 1967). The two injectable ester forms currently commercially available are retinyl propionate and retinyl palmitate. Their comparative suitability as vitamin A sources has not been previously reported. With respect to tocopherol, requirements for feedlot cattle have not been clearly established, but estimates range between 15 and 60 IU/kg diet DM (NASEM 2016). Unlike supplemental retinol, supplemental tocopherol is not appreciably affected by ruminal fermentation (Weiss et al. 1995). Response to vitamin A and E dietary supplementation of feedlot cattle has been variable (Perry et al. 1968; Arnold et al. 1992; Bryant et al. 2010; Burken et al. 2012). Salinas-Chavira et al. (2014) observed that supplementation of a steam flaked corn-based growing-finishing diet fed to Holstein steers with vitamin A and E enhanced (11%) initial 56-d ADG. This effect was due to increased (7%) DMI, as vitamin supplementation did not affect gain efficiency or dietary NE; however, Cano et al. (2017) observed that daily tocopherol supplementation (500 IU/steer) of Holstein steers fed a steam flaked corn-based diet did not affect initial or overall growth-performance. The objective of the current experiment was to further evaluate feedlot growth performance and carcass characteristics of calf-fed Holstein steers as affected by dietary vitamin E supplementation as RRR α-tocopherol, and injectable vitamin A form (retinyl propionate vs retinyl palmitate).

2. Material and methods

All animal care, handling, and sample techniques followed protocols approved by the University of California, Davis, Animal Use and Care Committee.

2.1. Animals, diet, treatments and sampling

One hundred and eight Holstein steers (average initial live weight, 126 ± 8 kg BW) were used in a 314-d growth-
performance trial to examine the effects of dietary supplemental vitamin E level and injectable retinol ester form on characteristics of growth performance, dietary energetics, carcass characteristics, and plasma tocopherol and retinol concentrations in calf-fed Holstein steers fed a conventional steam-flaked corn-based growing-finishing diet. Processing on arrival included branding, vaccination for IBR-PI3 (2 mL, SC, Cattle Master Gold, Zoetis, New York, NY), clostridials and Mannheimia (Pasteurella) haemolytica type A1 (5 mL, SC, One Shot Ultra 8, Zoetis, New York, NY), treatment for internal and external parasites (3 mL SC, Ivomec, Zoetis, New York, NY), and injection with 12 mL Liquamycin (LA-200, Zoetis, New York, NY). Steers were blocked by initial off-truck body weight (BW) into 3 groups and allotted within weight grouping to 18 pens (6 steers/pen). Pens within each weight grouping were then randomly assigned to 1 of 6 treatments in a 2 × 3 factorial arrangement. Calves were weighed at initiation of experiment, on day 112 and at harvest (day 314). Pens were 5.48 × 9.14 m with 26.7 m² of shade, and were equipped with automatic waterers and fence-line feed bunks (4.27 m in length). Steers were implanted with Revalor-S (120 mg trenbolone acetate plus 24 mg estradiol, Merck Animal Health, Summit, NJ) on d 112 and 224 of the study. Composition of the basal experimental diet is shown Table 1. Diets were prepared at approximately weekly intervals and stored in plywood boxes located in front of each pen. Steers were allowed ad libitum access to feed and water. Fresh feed was provided twice daily at 06:00 and 14:00 h, offering approximately 30% of daily consumption in the morning feeding and the remainder in the afternoon feeding. Feed and refusal samples were collected daily for DM analysis, which involved oven drying the samples at 105°C for 24 h. Carcasses were chilled for 24 h, the following measurements were obtained: LM area (cm²) by direct grid reading of the Longissimus muscle (LM) at the 12th rib; subcutaneous fat (cm) over the LM at the 12th rib taken at a location 3/4 the lateral length from the chine bone end (adjusted by eye for unusual fat distribution); kidney, pelvic and heart fat (KPH) as a percentage of HCW; marbling score (USDA 1997; using 3.0 as minimum slight, 4.0 as minimum small, 5.0 as minimum modest, 6.0 as minimum moderate, etc.), and estimated retail yield of boneless, closely trimmed retail cuts from the round, loin, rib and chuck (% of HCW) = 52.56 – 1.95 × subcutaneous fat – 1.06 × KPH + 0.106 × LM area – 0.018 × HCW (Murphey et al. 1960).

2.2. Estimation of dietary NE

Daily energy gain (EG; Mcal/d) was calculated by the equation: 

\[
EG = ADG^{1.097} 0.0557 \sqrt{W^{0.75}}, \]

where W is the mean shrunk BW (kg; NRC 1984). Maintenance energy (EM) was calculated by the equation: 

\[
EM = 0.084W^{0.75} \quad (Garrett \ 1971). \]

Dietary NE\text{\tiny{g}} was derived from NE\text{\tiny{m}} by the equation: 

\[
NE_{\text{\tiny{g}}} = 0.877 NE_{\text{\tiny{m}}} - 0.41 \quad (Zinn \ 1987). \]

Dry matter intake is related to energy requirements and dietary NE\text{\tiny{m}} according to the equation: 

\[
DMI = EG/0.877NE_{\text{\tiny{m}}} - 0.41, \]

and can be resolved for estimation of dietary NE\text{\tiny{m}} by means of the quadratic formula: 

\[
x = (-b - \sqrt{b^2 - 4ac})/2c, \]

where \(a = -\).014EM, \(b = 0.877EM + 0.41DMI + EG, \) and \(c = -0.877DMI \) (Zinn and Shen 1998).

2.3. Carcass data

Hot carcass weights (HCW) were obtained at time of slaughter. After carcasses chilled for 24 h, the following measurements were obtained: LM area (cm²) by direct grid reading of the Longissimus muscle (LM) at the 12th rib; subcutaneous fat (cm) over the LM at the 12th rib taken at a location 3/4 the lateral length from the chine bone end (adjusted by eye for unusual fat distribution); kidney, pelvic and heart fat (KPH) as a percentage of HCW; marbling score (USDA 1997; using 3.0 as minimum slight, 4.0 as minimum small, 5.0 as minimum modest, 6.0 as minimum moderate, etc.), and estimated retail yield of boneless, closely trimmed retail cuts from the round, loin, rib and chuck (% of HCW) = 52.56 – 1.95 × subcutaneous fat – 1.06 × KPH + 0.106 × LM area – 0.018 × HCW (Murphey et al. 1960).

2.4. Statistical design and analysis

For calculating steer performance, initial, interim and final BW were reduced 4% to account for digestive tract fill. Pen was considered as experimental unit. The experimental data were
analyzed as a randomized complete block design experiment with a 2 × 3 factorial arrangement (retinol sources and tocopherol level). The statistically model for the trial was as follows:
\[ Y_{ijk} = \mu + B_i + T_j + E_{ijk} \]
where \( Y_{ijk} \) is the response variable, is the common experimental effect, \( B_i \) is the block (initial weight), \( T_j \) is the treatment effect, and \( E_{ijk} \) is the residual error. Treatments effects were tested for (1) effects of retinol ester form, (2) linear and quadratic components of dietary supplemental tocopherol level, and (3) the interaction of retinol form and dietary supplemental tocopherol (Statistix 10, Analytical Software, Tallahassee, FL). Contrasts were considered significantly when the \( P \) value was ≤ 0.05, and tendencies are identified when the \( P \) value was >0.05 and ≤ 0.10. Blood concentrations of plasma tocopherol and retinol were analyzed using linear mixed model for repeated measures in a completely randomized design with covariance structure: A, CS, AR1, and pen as a random component. Contrasts were considered significant when the \( P \) value was ≤ 0.10.

3. Results and discussion

Based on tabular values for individual dietary ingredients (NRC 2000), the equivalent vitamin A and E concentrations of basal diet (Table 1) were 2420 and 25 IU/kg (DM basis), respectively. The estimated vitamin A concentration was 110% of reported requirement (NASEM 2016). Nevertheless, in a survey of 42 consulting nutritionist representing major feedlot cattle feeding areas of the United States, Vasconcelos and Galyean (2007) observed that nutritionists recommended on average of 5215 × 1000 IU/kg diet DM in addition to whatever amount of vitamin A equivalent is provided by the diet. The requirement for vitamin E has received more limited attention, although it is assumed to fall within the range of 15 and 60 IU/kg diet DM (NASEM 2016). Vasconcelos and Galyean (2007) observed that vitamin E concentration of basal growing-finishing diets averaged 26 IU/kg DM, consistent with that of the present study.

One hundred seven steers completed the study. One steer died during this study (bloat). There were no treatment effects on morbidity (\( P = 0.59 \)) or mortality (\( P = 0.48 \)), averaging 2.8 and 0.9%, respectively. There were no treatment interactions (\( P > 0.10 \)). Main effects of dietary supplementation of tocopherol and injectable retinol ester form on growth performance, dietary energetics and carcass characteristics are shown in Table 2. During the early 112-d period, dietary supplemental tocopherol tended to increase (linear effect, \( P = 0.07 \)) ADG. The magnitude of the response, although small, was attributable to a concomitant tendency for increased (linear effect, \( P = 0.06 \)) DMI. Gain efficiency during this period was not affected (\( P = 0.58 \)). Subsequent and overall 314-d ADG, DMI, gain efficiency, and estimated dietary NE, as well as carcass characteristics were not affected (\( P > 0.10 \)) by dietary supplemental tocopherol.

Based on tabular values for feed ingredients, the basal diet contained 25 IU tocopherol/kg DM, putatively (NASEM 2016) sufficient to meet requirements. Accordingly, enhancements in growth performance in response to dietary supplemental tocopherol would not be expected. The increased DMI and associated increase in ADG with increasing level of dietary tocopherol supplementation during the initial 112 days on feed indicates a potentially beneficial role for dietary tocopherol supplementation of calf-fed Holstein steers during that early growing phase. Likewise, Salinas-Chavira et al. (2014) observed enhanced DMI and ADG in calf-fed Holstein steers fed 250 IU RRR α-tocopherol/d during the initial growing period. In contrast, Cano et al. (2017) did not observe an effect of tocopherol supplementation (0, 250 or 500 IU/d) on feedlot growth performance of calf-fed Holstein steers during an initial 56-d growing phase. Calf-fed Holstein steers entering the feedlot at 125–135 kg initial weight are characteristically 100–110 d of age. Differences in responses to supplementation may reflect the calves initial vitamin E status in consequence to rearing management.

Overall, effects of dietary supplemental tocopherol on feedlot cattle performance have been variable and inconsistent. In numerous studies (Arnold et al. 1992; Garber et al. 1996; Carnegiey et al. 2008) little or no effect of vitamin E supplementation on feedlot cattle performance was observed. Whereas, in other studies vitamin E supplementation improved performance. Hill et al. (1995) observed an increase in ADG (9.8%) and gain efficiency (3.7%) in yearling steers supplemented 1500 IU tocopherol/d during 105 d feeding period. Burken et al. (2012) observed linearly increased ADG, and a tendency for increased DMI and gain efficiency in crossbred cattle supplemented daily with 125, 250 or 500 IU tocopherol during a 97-d finishing period.

There were no treatment effects (\( P > 0.10 \)) of dietary supplementation of tocopherol and injectable retinol ester form on carcass characteristics (Table 3). Liu et al. (1996) did not observe an influence of tocopherol supplementation (0–2000 IU/d) during the finishing phase on carcass characteristics, including quality and yield grades. Likewise, Bloomberg et al. (2011) did not observe an influence of tocopherol supplementation (0, 125, 250 or 500 IU/d) during the final 97 d on carcass characteristics. Secrist et al. (1995) observed that fat thickness, marbling score, and YG were numerically greater with tocopherol supplementation of feedlot diets, although differences were not statistically significant.

Injectable retinol form did not affect initial 112-d DMI. However, subsequent and overall DMI tended (\( P < 0.10 \)) to be greater (3%) for steers injected with retinyl palmitate vs retinyl propionate. The basis for this effect is not certain, but may reflect differences between sources in retinol bioavailability, as at the end of the initial 112-d period plasma retinol was greater (14%, \( P = 0.04 \); Table 4) for retinyl palmitate vs retinyl propionate. There were no treatment effects (\( P > 0.10 \)) on gain efficiency, estimated dietary net energy, or carcass characteristics. Alosilla et al. (2007) compared effects of a variety of dietary supplemental vitamin A sources on carcass performance. However, to our knowledge no information has been previously reported comparing injectable vitamin A ester form on feedlot cattle performance.

Treatment effects on plasma tocopherol and retinol concentrations are shown in Table 4. As expected, dietary supplemental tocopherol increased (linear effect, \( P < 0.01 \)) plasma tocopherol concentrations on d 28 and 112. Injectable retinol form did not affect (\( P > 0.10 \)) plasma tocopherol concentration. However, dietary supplemental tocopherol increased (linear effect, \( P < 0.01 \)) plasma retinol concentration on d 28 following
### Table 2. Main effects of supplemental tocopherol and injectable retinol ester on growth performance and dietary energetics of Holstein steers.

| Item                | Supplemental tocopherol<sup>a</sup> IU/steer/d | P-value | Injectable retinol ester<sup>b</sup> |
|---------------------|-----------------------------------------------|---------|-------------------------------------|
| No. of pens         | 6                                             | 6       | 6                                  |
| No. of animals      | 36                                            | 36      | 36                                 |
| Live BW, Kg<sup>c</sup> |                                               |         |                                    |
| Initial             | 126                                           | 126     | 126                                |
| 112 d               | 261                                           | 262     | 266                                |
| 314 d               | 571                                           | 569     | 587                                |
| ADG, Kg/d           |                                               |         |                                    |
| 1–112 d             | 1.2                                         | 1.22    | 1.25                               |
| 112–314 d           | 1.54                                        | 1.52    | 1.59                               |
| 1–314 d             | 1.42                                        | 1.41    | 1.47                               |
| DMI, Kg/d           |                                               |         |                                    |
| 1–112 d             | 5.10                                         | 5.16    | 5.27                               |
| 112–314 d           | 8.85                                         | 8.99    | 9.04                               |
| 1–314 d             | 7.51                                         | 7.63    | 7.70                               |
| Gain for feed       |                                               |         |                                    |
| 1–112 d             | 0.235                                        | 0.235   | 0.237                              |
| 112–314 d           | 0.175                                        | 0.169   | 0.176                              |
| 1–314 d             | 0.189                                        | 0.184   | 0.190                              |
| Dietary NE<sub>e</sub>, Mcal/kg |                                        |         |                                    |
| 1–112 d             | 1.90                                         | 1.90    | 1.91                               |
| 112–314 d           | 2.27                                        | 2.12    | 2.26                               |
| 1–314 d             | 2.18                                        | 2.14    | 2.20                               |
| Dietary NE<sub>e</sub>, Mcal/Kg |                                        |         |                                    |
| 1–112 d             | 1.26                                         | 1.26    | 1.26                               |
| 112–314 d           | 1.58                                         | 1.45    | 1.57                               |
| 1–314 d             | 1.50                                         | 1.47    | 1.52                               |
| Observed/expected NE |                                             |         |                                    |
| 1–112 d             | 0.86                                         | 0.86    | 0.86                               |
| 112–314 d           | 1.02                                         | 0.96    | 1.02                               |
| 1–314 d             | 0.98                                         | 0.96    | 0.99                               |

<sup>a</sup> alpha-tocopherol (Emcelle Tocopherol Stuart Products Inc. Bedford, TX) was top dressed on feed once daily in the morning feeding.

<sup>b</sup> 500,000 IU of retinol as retinyl propionate (Vita-Jec A&D ‘500’, RXV Products, Kansas City, MO) or retinyl palmitate (Vital E-A+D, Stuart Products, Bedford, TX) were administered subcutaneously on d 1, 112, and 224 of the 314-d study.

<sup>c</sup> Initial weight is the off-truck arrival weight. Interim and final weights were reduced 4% to account digestive tract fill.

### Table 3. Main effects of supplemental tocopherol and injectable retinol ester on carcass characteristics of Holstein steers.

| Item                | Supplemental tocopherol<sup>a</sup> IU/steer/d | P-value | Injectable retinol ester<sup>b</sup> |
|---------------------|-----------------------------------------------|---------|-------------------------------------|
| HCW, Kg             |                                               |         |                                    |
| 350                 | 348                                           | 359     |
| Dressing percentage |                                               |         |                                    |
| 61.3                | 61.0                                          | 61.4    |
| KPH, %<sup>d</sup>  |                                               |         |                                    |
| 2.33                | 2.25                                          | 2.41    |
| Fat thickness, cm   |                                               |         |                                    |
| 0.65                | 0.64                                          | 0.69    |
| LM area, sq cm      |                                               |         |                                    |
| 87.2                | 89.2                                          | 86.8    |
| Marbling score<sup>e</sup> |                                           |         |                                    |
| 4.46                | 4.09                                          | 4.84    |
| Retail yield, %<sup>f</sup>  |                                               |         |                                    |
| 51.6                | 51.9                                          | 51.3    |

<sup>a</sup> alpha-tocopherol (Emcelle Tocopherol Stuart Products Inc. Bedford, TX) was top dressed on feed once daily in the morning feeding.

<sup>b</sup> 500,000 IU of retinol as retinyl propionate (Vita-Jec A&D ‘500’, RXV Products, Kansas City, MO) or retinyl palmitate (Vital E-A+D, Stuart Products, Bedford, TX) were administered subcutaneously on d 1, 112, and 224 of the 314-d study.

<sup>c</sup> Kidney, pelvic, and heart fat as a percentage of carcass weight.

<sup>d</sup> Coded: minimum slight = 3, minimum small = 4, etc.

<sup>e</sup> Estimated proportion of closely trimmed boneless retail cuts from carcass round, loin, rib and chuck (USDA 1997).

### Table 4. Main effects of supplemental tocopherol and injectable retinol ester on plasma tocopherol and retinol of Holstein steers.

| Item                | Supplemental tocopherol<sup>a</sup> IU/steer/d | P-value | Injectable retinol ester<sup>b</sup> |
|---------------------|-----------------------------------------------|---------|-------------------------------------|
| Plasma tocopherol, μg/mL |                                         |         |                                    |
| Day 1               | 1.73                                         | 2.18    | 1.47                               |
| Day 28              | 1.59                                         | 4.90    | 6.62                               |
| Day 112             | 2.62                                         | 6.66    | 8.45                               |
| Plasma retinol, μg/mL |                                         |         |                                    |
| Day 1               | 0.233                                        | 0.275   | 0.217                              |
| Day 28              | 0.175                                        | 0.183   | 0.242                              |
| Day 112             | 0.200                                        | 0.200   | 0.225                              |

<sup>a</sup> alpha-tocopherol (Emcelle Tocopherol Stuart Products Inc. Bedford, TX) was top dressed on feed once daily in the morning feeding.

<sup>b</sup> 500,000 IU of retinol as retinyl propionate (Vita-Jec A&D ‘500’, RXV Products, Kansas City, MO) or retinyl palmitate (Vital E-A+D, Stuart Products, Bedford, TX) were administered subcutaneously.
ing plasma retinol levels with a normal range across a 112-d single injection of 500,000 IU vitamin A is sustainable and its concentration in plasma is indicative that liver vitamin A stores are depleted by the liver (Ross 2006). Nevertheless, plasma retinol concentration is not a reliable indicator of vitamin A status, per se, due to homeostatic regulation of the liver. A single injection of 500,000 IU vitamin A is sufficient for sustaining plasma retinol levels with a normal range across a 112-d interval.

4. Conclusion
Supplementation of vitamin E above basal requirements may enhance growth performance during the initial 112-d feeding phase. However, the overall effect on growth performance and carcass characteristics of cull-fed Holstein steers is not appreciable. Injectable retinol ester form did not affect cattle growth performance. Based on plasma retinol concentrations, the bioavailability of retinyl palmitate may be greater than that of retinyl propionate.

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No potential conflict of interest was reported by the authors.

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