

FCEB Proj. #28, Nelson et al.

## **Florida Cattle Enhancement Grant**

### **Final Project Report**

**September 1, 2017**

UF Project agreement number: AGR00007489

FCEB project number: 28

**Project Title:** Effects of dietary vitamins A, D and E sources fed peri-partum on body stores of vitamins A, D, and E and performance of beef cows and calves.

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### **Matching Funds:**

As stated in the proposal, DSM Nutrition provided matching support for 50% of the direct costs of the project. The funds from DSM will be used to complete planned activities of the project which have not been completed yet.

**Project objectives:** Determine the effects of level and source of dietary vitamin A, D, and E fed to Florida beef cows during the peri-partum period on:

- Body stores of vitamins A, D and E in cows and calves,
- Immune status of cows and calves, and

- Reproductive status of cows.

### **Background:**

The fat-soluble vitamins (A, D &E) each have important and unique roles in immunity, lactation, and reproductive function of cattle<sup>1,2</sup>. Cattle receive adequate supplies of vitamins A and E when grazing fresh, good-quality pastures, and vitamin D from sun exposure. Therefore, it is often assumed that Florida cattle receive adequate supplies of each vitamin throughout most of the year, as compared to cattle in more northern regions. However, low-quality forages that Florida cows consume during the winter months or periods of drought are poor sources of vitamins A and E. Vitamin D synthesis in the skin from sun exposure also decreases during winter months.

Our preliminary data indicated striking deficiencies of vitamins D and E in Florida beef calves born to cows fed according to typical industry standards during the winter months. The concentration of beta-carotene, the precursor to vitamin A that also has critical functions in immunity and reproduction independent of vitamin A, remains unknown. As winter forages and stored forages are low in beta-carotene compared to fresh pasture we hypothesize that Florida cows and calves also have low beta-carotene status in the winter months. We hypothesized that increased supplementation of sources of vitamins A, D and E to beef cows during late gestation and early lactation would improve fat-soluble vitamin status of the cow and calf, thereby improving the overall calf crop through decreasing still-births and incidence of disease, along with improved reproductive performance of the cow.

**Approach:**

**Animals and treatments:** Sixty cross-bred cows located at the North Florida Research and Education Center were used for the study. Cows were assigned to one of three treatments at 250 d of gestation:

- **Control:** Supplement formulated to meet average daily intake of current National Research Council requirements of vitamins A, D, and E for beef cattle. The NRC rates meet the minimal requirement for cattle but are not believed to be adequate for optimal performance.
- **Vit1:** Supplement formulated to provide average daily intake of 100,000 IU of vitamin A as retinyl-palmitate, 40,000 IU vitamin D, and 500 IU of vitamin E. These rates are similar to typical rates fed to dairy cows and have benefits for health and reproduction.
- **Vit2:** Supplement formulated to provide an average daily intake of 100,000 IU equivalents of vitamin A as  $\beta$ -carotene, 40,000 IU equivalents of vitamin D as 25-hydroxyvitamin D, and 1,000 IU of vitamin E. The beta-carotene and 25-hydroxyvitamin D supplements are alternative sources that may provide added benefits compared to vitamin A and vitamin D alone.

Treatments were randomly assigned to individual cows (20 cows/treatment) with cows blocked by parity. The treatments were provided 3 days per week (M, W, and F) as a top-dress and fed from 250 d of gestation to 60 days after calving.

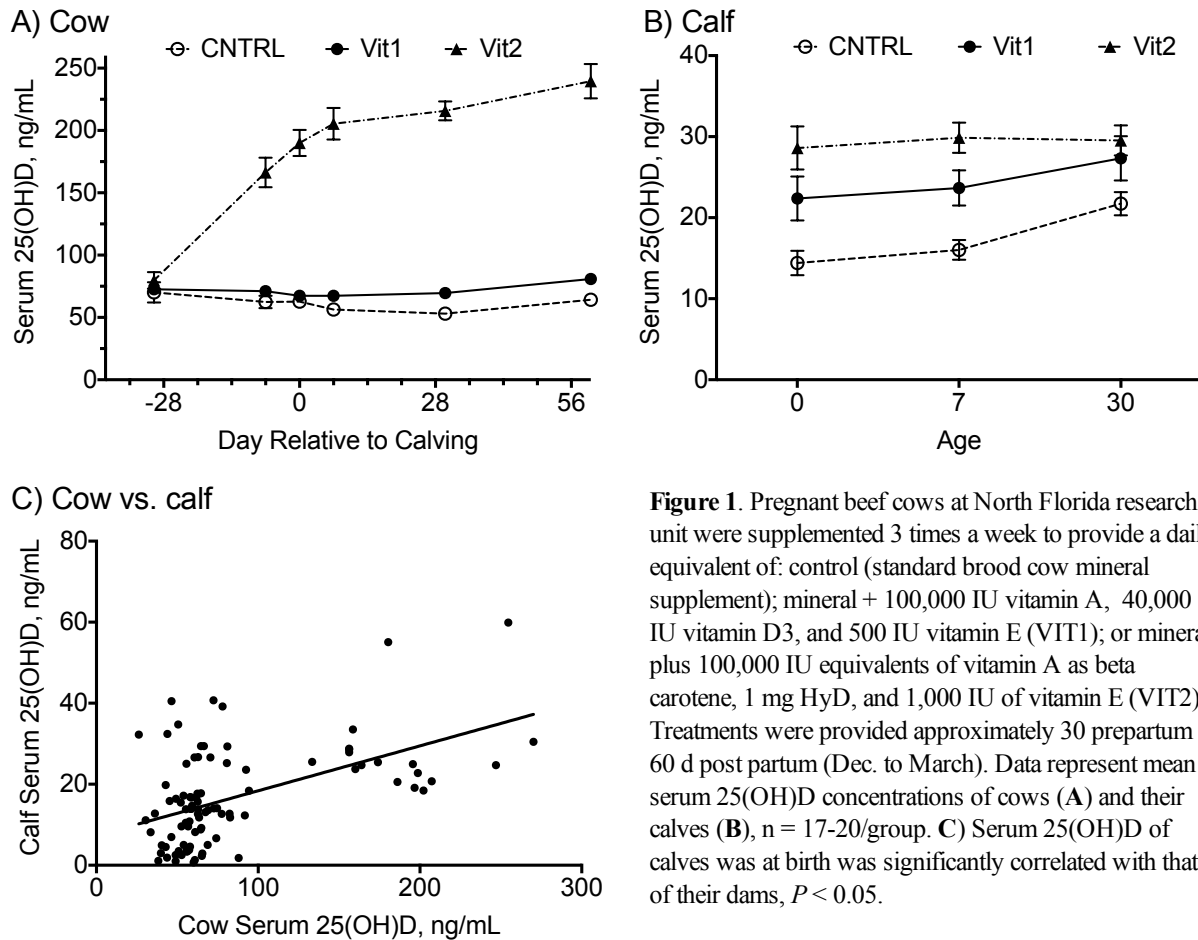
**Measurements:** Blood was sampled from all cows at 0 and 21 d relative to start of treatments and 0, 7 30 and 60 d relative to calving. Blood was sampled from calves at birth, 7 and 30 d of age. Body weights also will be collected at time of blood sample collection. Colostrum was sampled from all cows at time of calving. Liver biopsies were performed on 6 cows/treatment at -30 d, -7 d, and 30 d relative to calving and 5 calves per treatment at 7 d and 30 d of age.

Concentrations of 25-hydroxyvitamin D in serum samples of cows and calves were measured as an indicator of vitamin D status. Concentrations of retinol in liver and  $\alpha$ -tocopherol in serum will be measured as indicators of vitamin A and vitamin E status. Immune status of cows and calves was assessed by neutrophil oxidative burst capacity and blood leukocyte profiling of CD11b, CD14, CD21, and CD62L protein abundance by flow cytometry. Concentrations of IgG1, IgG2, and IgA in serum of cows and calves and colostrum samples also will be measured as an indicator of immune status. The time to return of estrus in cows will be measured as an indicator of reproductive health of cows by measuring concentrations of progesterone and estrogen in serum sampled weekly from 30 to 72 d post partum.

### **Current results:**

Analysis of the effects of the vitamin treatments on concentrations of 25-hydroxyvitamin D in serum of cows and calves has been completed (Figure 1). Cows fed Vit1 and Vit2 treatments, along with calves born to those cows, had elevated concentrations of 25-hydroxyvitamin D in serum compared with Control cows and calves. Notably, the average serum 25-hydroxyvitamin D concentrations of Control calves was

below the 20 ng/mL target threshold for cattle, confirming previous data that current winter vitamin nutrition practices for beef cows in Florida are not adequate.



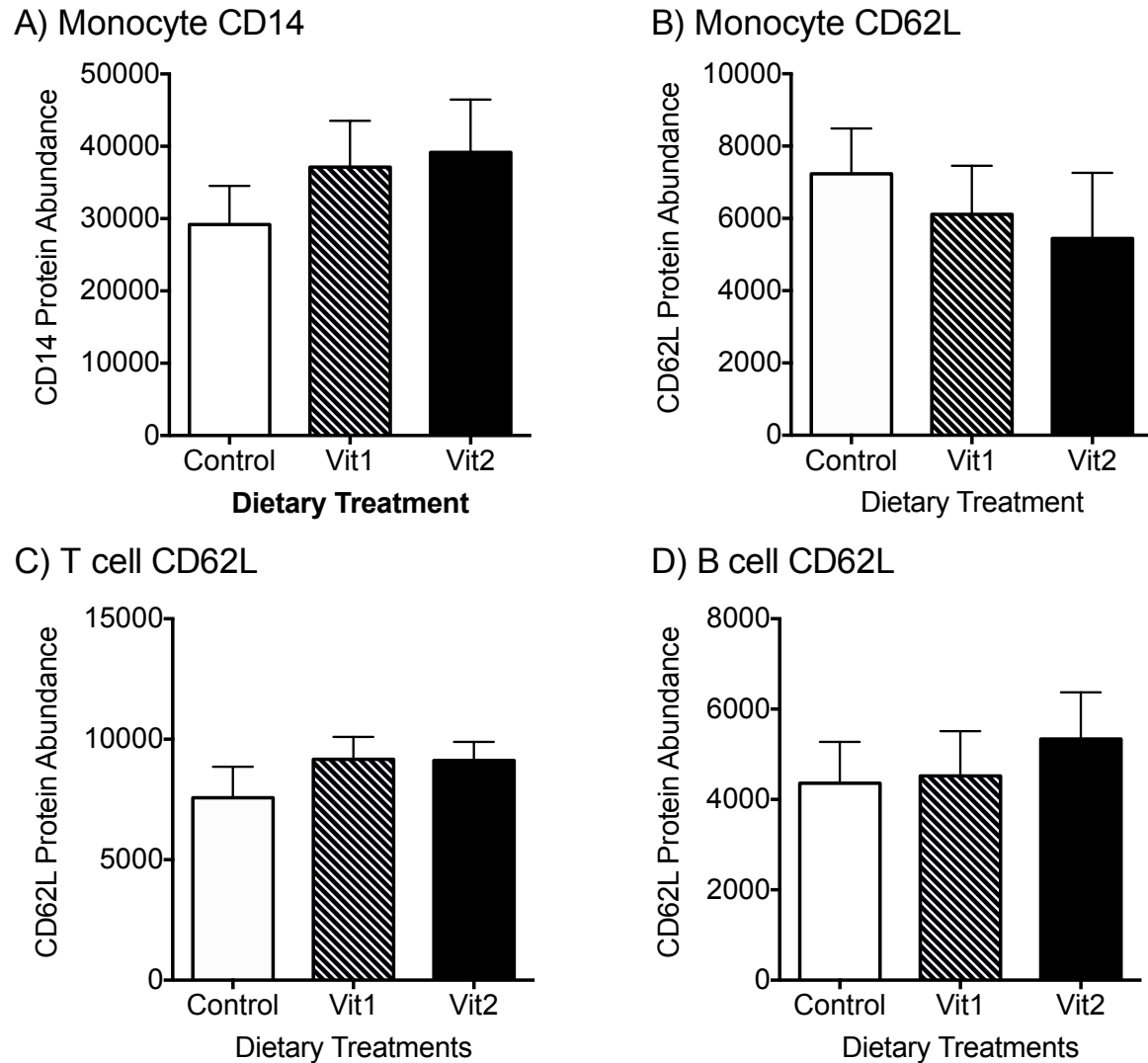
**Figure 1.** Pregnant beef cows at North Florida research unit were supplemented 3 times a week to provide a daily equivalent of: control (standard brood cow mineral supplement); mineral + 100,000 IU vitamin A, 40,000 IU vitamin D<sub>3</sub>, and 500 IU vitamin E (VIT1); or mineral plus 100,000 IU equivalents of vitamin A as beta carotene, 1 mg HyD, and 1,000 IU of vitamin E (VIT2). Treatments were provided approximately 30 prepartum to 60 d post partum (Dec. to March). Data represent mean serum 25(OH)D concentrations of cows (A) and their calves (B), n = 17-20/group. C) Serum 25(OH)D of calves was at birth was significantly correlated with that of their dams,  $P < 0.05$ .

Supplementation of cows with an equivalent of 40,000 IU of vitamin D<sub>3</sub> per day (provided in Vit1 treatment) improved vitamin D status of calves, and is an affordable (less than a tenth of a penny per day) to achieve adequate vitamin D status of cows and newborn calves in the winter months. Supplementation of cows with 1 mg 25-hydroxyvitamin D<sub>3</sub> per day dramatically increased concentrations of 25-hydroxyvitamin

D<sub>3</sub> in cows over the peri-parturient period, which has been shown to have positive benefits for health and production of dairy cows. The effects of treatments on vitamin A and vitamin E status of cows and calves will be completed using matching funds from DSM Nutrition.

The effects of treatments on blood leukocyte profiles has been completed. We did not observe effects of treatments on percentages of lymphocytes, monocytes, or neutrophils in cows, but we did observe that calves fed the Vit1 and Vit2 treatments had elevated proportions of cells that expressed the CD14 protein, a protein critical for detection of bacterial pathogens (Figure 2). In contrast, expression of CD62L, an adhesion protein, was decreased on monocytes and increased on T cells and B cells. Overall the data indicate potential improvement for recruitment of T cells and B cells to sites of infection, and potential improvement in ability of immune cells to recognize bacterial pathogens. The effects of treatments on calf serum IgG concentrations, and cow reproductive health (return to cyclicity and pregnancy rate) are still under investigation.

In conclusion, the current results support the approach of increasing dietary vitamin D supplementation of Florida beef cows during gestation in winter months from the current practice of 4,000 IU/d to 40,000 IU/d for the purpose of achieving adequate vitamin D status in their calves. Overall, the practice of increasing vitamins A, D and E in winter mineral supplements for gestating beef cows modulates immune status of their calves. Continued research with a sufficient number of cows is warranted to determine the effects of increasing supplemental vitamins A, D, and E in cow mineral supplements on health and performance of beef calves born in the winter.



**Figure 2.** Data represent mean  $\pm$  95% CI expression of CD14 protein on monocytes (A) and CD62L protein on monocytes (B), T cells (C), and B cells (D) sampled from blood of 7 d old calves born to cows fed the Control, Vit1, or Vit2 treatments.

<b>BUDGET FOR FLORIDA CATTLE ENHANCEMENT FUND- BUDGET JUSTIFICATION</b>			
<b>PROJECT TITLE: Chemically treating forages with alkali may improve digestibility and enhance beef cattle performance - Project # P0038505 (FCEB# 29)</b>			
DETAILED LINE ITEM DESCRIPTION	TOTAL	EXPLANATION/JUSTIFICATION OF DELIVERABLE	COMPLETION DATE
Animal per diem	\$ 6,900.00	Use of experimental cows and calves reagents for analyses	6/1/2027
Vitamin assays	\$ 5,068.00	Measurement of vitamins in serum	8/1/2017
Immune assays	\$ 10,077.00	Measurement of immune status indicators	9/1/2017
Progesterone and Estrogen assays	\$ 4,500.00	Measurement of reproductive status of cows	9/1/2017
Materials and Supplies	\$ 1,591.00	Laboratory supplies needed to perform the project	9/1/2017
Project Support and Tuition	\$ 20,210.41	Support to perform the project	9/1/2017
Indirect Cost	\$ 5,656.43		N/A
<b>GRAND TOTAL: (equal to percentage of completion)</b>	<b>\$ 54,002.84</b>		