EFFECTS OF WEANING, FASTING AND B-VITAMIN INJECTIONS ON PLASMA B-VITAMIN CONCENTRATIONS IN BEEF CALVES

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Story in Brief

Twelve 364 lb beef calves were weaned, limit-fed and fasted for three days during the 20-day period after weaning, and then inoculated intranasally with live attenuated infectious bovine rhinotracheitis on day 20. Six of these calves were injected intramuscularly with water soluble vitamins every 2 days from day 9 to day 34. Plasma ascorbic acid and pantothenic acid were depressed by weaning and restricted feeding; folic acid and B₁₂ were not. Plasma concentrations of vitamin C, folic acid, pantothenic acid and vitamin B₁₂ were increased by injections by day 20. Plasma vitamin C, pantothenic acid and B₁₂ but not folic acid, were markedly reduced 5 days after infectious bovine rhinotracheitis-infection in both control and vitamin-injected calves, but rebounded by day 34 with recovery from infectious bovine rhinotracheitis. Results indicate that stress of weaning and infection can alter blood levels of some vitamins. Plasma vitamin concentrations were consistently high for calves injected with vitamins on the day of inoculation, perhaps reflecting altered tissue release or metabolism and excretion. Whether supplements will reduce severity of infections is not yet known.

(Key Words: B-Vitamins, Stress, Cattle, Disease, IBR.)

Introduction

Requirements for vitamin B₆, folic acid and pantothenic acid are increased by up to 20-fold by stress or injury in humans. Supplementation with these vitamins plus vitamin C and B₆ have helped to overcome stress and cortisol-induced immunosuppression in man and animals.

Infection with infectious bovine rhinotracheitis vaccine (IBRV) was used as a model for respiratory disease, because IBRV infection predisposes cattle to bacterial pneumonia. Stress on our calves, from weaning, injections, restricted feeding and a 3-day fast, probably was less severe in magnitude than shipped

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calves normally endure. Plasma levels of vitamin B6, folic acid, vitamin B12, pantothenic acid and ascorbic acid were measured in these calves to investigate the effects of vitamin injections, restricted feeding and fasting, and IBRV infection.

Materials and Methods

Twelve 6 to 8 month old steer calves (Hereford X Angus) from one herd which had been wintered on dormant native range were weaned on April 23, 1991. They were trucked 10 miles, weighed on arrival and housed individually in metabolism stalls. The calves were assigned randomly to two treatments (Control, +Vit). The six control calves received sterile saline injections while the six +Vit calves received injections of B-vitamins and vitamin C (Table 1). Vitamins were injected every 2nd day from 9 days after arrival until day 34.

On arrival, each calf was fed 2 lb of native prairie grass hay (4% crude protein). From day 0 to day 11, and day 15 to day 22, calves were offered native prairie grass hay once daily at 1% of body weight. Alfalfa hay (21%

Table 1. B-vitamin requirements and levels supplied by injection.

<table>
<thead>
<tr>
<th>Vitamin</th>
<th>Estimated Requirement&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Stress factor&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Dosage&lt;sup&gt;c&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thiamin</td>
<td>6.76 mg</td>
<td></td>
<td>13.5 mg</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>16.91 mg</td>
<td></td>
<td>33.8 mg</td>
</tr>
<tr>
<td>Niacin</td>
<td>67.64 mg</td>
<td></td>
<td>135.0 mg</td>
</tr>
<tr>
<td>Folic acid</td>
<td>2.09 mg</td>
<td>15</td>
<td>60.0 mg</td>
</tr>
<tr>
<td>Pantothenic acid</td>
<td>54.1 mg</td>
<td>2</td>
<td>216.0 mg</td>
</tr>
<tr>
<td>Vitamin B&lt;sub&gt;6&lt;/sub&gt;</td>
<td>6.76 mg</td>
<td>8</td>
<td>108.0 mg</td>
</tr>
<tr>
<td>Vitamin B&lt;sub&gt;12&lt;/sub&gt;</td>
<td>67.64 ug</td>
<td>2</td>
<td>270.0 ug</td>
</tr>
<tr>
<td>Vitamin C</td>
<td>Unknown</td>
<td></td>
<td>1000.0 mg</td>
</tr>
</tbody>
</table>

<sup>a</sup> Daily B-vitamin requirements for a 420 lb calf were estimated on a metabolic body weight basis as equivalent to 3.56 times the requirements for a 75 lb pig (NRC, 1990).

<sup>b</sup> The estimated daily requirement was multiplied by factors of 2 for pantothenic acid and vitamin B<sub>12</sub>, 15 for folic acid, and 8 for vitamin B<sub>6</sub> to account for the increased requirements of these specific B-vitamins during stress or "moderate" injury (Mueller and Thomas, 1975).

<sup>c</sup> Twice the daily estimated requirement was supplied by injection every 2 days, with one 3 ml dose containing the B-vitamins and one 4 ml dose containing the vitamin C.
crude protein) was substituted for 50% of the prairie hay intermittently and for the final week of the study. Calves were fasted on days 12, 13 and 14. On day 22, feeding rate was increased to 1.5% of weight so that calves would maintain body weight.

On day 20, the 12 calves each were inoculated intranasally with $10^7$TCID$_{50}$ IBR virus (vaccine strain). The calves had received 6 injections of B-vitamins or sterile saline by day 20. Blood plasma samples were taken on 8 different dates as shown in the figures. Day 5 provided samples before vitamin injections began. Day 12 and 15 samples were taken at the beginning and end of the fast. Day 20 samples were taken immediately before IBRV inoculation. Day 25 samples corresponded with peak IBRV infection (symptoms were evident in most calves only between days 24 and 26). Day 34 samples were taken at the termination of vitamin injections. Day 36 and 38 samples were taken to provide further information on blood vitamin concentrations after cessation of vitamin injections. Blood samples were taken immediately before vitamin injections were given.

Blood was obtained by jugular venipuncture into tubes containing sodium heparin and tubes containing potassium EDTA. Vitamin C was analyzed by a standard colorimetric method. Folic acid and vitamin B$_{12}$ were analyzed by a radioassay method (Quantaphase Radioassay, Bio-Rad Clinical Division, Hercules, CA). Plasma pantothenic acid was measured using an indirect ELISA assay (Song et al., 1990). Plasma vitamin data were analyzed to test effects of vitamin treatment and collection day.

Results and Discussion

Details of body weight changes, feed intake, IBRV symptoms and immunological parameters were reported previously (Dubeski et al., 1992). Calves had a mean weight of 364 lb. The restricted feeding/fasting regimen resulted in an average shrink of 13.3% over a period of 16 days. Some of the weight loss during fasting was recovered immediately when feed and water were provided; shrink in the 20 days prior to IBRV infection averaged 7.7%. IBRV caused a mild respiratory infection in all calves, with symptoms (body temperature, nasal secretions) peaking between 4 and 6 days after inoculation.

Plasma vitamin C concentrations were similar for the two treatment groups before vitamin injections began (Figure 1). On all figures, stars indicate times that plasma concentrations differed significantly between vitamin injected and control calves. The decline in plasma vitamin C in control calves between days 5 and 12 may be due either to the negative energy balance or to reduced stress from blood sampling. Stress due to handling and blood sampling elevates blood vitamin C levels due to cortisol release from the adrenal glands. As animals became accustomed to handling, blood vitamin C concentrations should reflect nutritional status more accurately.
Plasma vitamin C in +Vit calves was not immediately increased by the injection of 1000 mg vitamin C every 2 days starting on day 9. This may represent tissue repletion because vitamin C is several times higher in many tissues than in plasma. At the end of the fast period (day 15), plasma vitamin C tended to be higher in +Vit calves (P<.09), and it was much higher on day 20. We suspect that the high day 20 level in +Vit calves was related to tissue release from stress, because blood samples were taken immediately following nasal sampling with swabs and tampons for the first time. In both groups of calves, plasma vitamin C had declined by day 25, the time of peak IBRV infection, although levels still tended to be higher in +Vit calves. In other species including man, vitamin C content of plasma and leukocytes is reduced by disease possibly reflecting an increased requirement during an immune response.

Folic acid concentrations are shown in Figure 2. Vitamin injections increased mean plasma folic acid rapidly (day 12) and levels remained elevated throughout the study. The folic acid dose was 60 mg/day, estimated to be 15 times the daily requirement. Girard et al. (1989) injected 4 month old heifers with up to 20 mg folic acid in a single injection but did not increase serum folate above 14.8 ng/ml; possibly his levels injected were too low.

Plasma folic acid was not sensitive to feed intake, although in other research at OSU, plasma folic acid was much higher in grain-fed cattle than in

Figure 1. Impact of stress and vitamin injections on vitamin C concentrations in blood plasma of steers.
calves. The extremely high plasma folic acid concentration on day 20 is difficult to explain. We observed relatively high folic acid concentrations in stressed calves in an earlier study (6.4 to 14.5 ng/mL) suggesting that folic acid status is not seriously reduced by stress and reduced feed intake in cattle.

Plasma vitamin B$_{12}$ results are shown in Figure 3. Plasma concentrations were increased by injections. In our study, plasma B$_{12}$ increased for control calves during restricted feeding and fasting, reaching maximum concentrations on day 20. Tissue vitamin B$_{12}$ stores in cattle, similar to humans, may be a thousand times greater than the daily requirement. IBRV infection reduced plasma vitamin B$_{12}$ in both control and +Vit cattle, (day 25 vs day 20). Plasma B$_{12}$ dropped 31% in control calves, possibly due to increased requirements. The peak in B$_{12}$ on day 20 may be due to gradually increasing vitamin status. The decrease in plasma B$_{12}$ (and similarly in other B-vitamins) on day 25 indicates a drastic change in vitamin metabolism or distribution due to IBRV.

Plasma pantothenic acid concentrations appeared to be sensitive to restricted intake and fasting and IBRV infection, but responded slowly to B-vitamin injection (Figure 4). Mean plasma pantothenic acid was relatively high (.190 mM) on day 5 in both control and +Vit calves. Plasma pantothenate was lower on day 25 (the point of most severe IBR infection) than at any other time,
Figure 3. Impact of stress and vitamin injections on vitamin $\text{B}_{12}$ concentrations in blood plasma of steers.

Figure 4. Impact of stress and vitamin injections on pantothenic acid concentrations in blood plasma of steers.
and rebounded on day 34, due to either increased feed intake or recovery from IBRV or both. However, injections of pantothenate did not elevate plasma concentration except on day 20.

In conclusion, a very mild infection depressed plasma concentrations of vitamin C, vitamin B₁₂ and pantothenic acid but failed to affect folic acid, at 5 days post-infection. Restricted intake and a 3-day fast decreased plasma levels of vitamin C and pantothenate but increased plasma B₁₂; these stresses did not affect plasma folic acid concentration. Plasma vitamin analyses should prove useful to detect specific deficiencies and to measure vitamin status responses to stress and to supplements.

Literature Cited


