Vitamin Supplementation for Lactating Dairy Cows: Industry Perspective

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Summary

In the current 2001 7th revised Dairy NRC, supplemental vitamin requirements for lactating dairy cows are listed for vitamin A, Vitamin D, and Vitamin E (75,000; 20,000; and 545 IU per head per day, respectively). In addition, production responses to 20 mg supplemental biotin and 50 grams of rumen-protected choline were discussed in detail. Since that time, most research advances have been made in transition cows, with vitamin requirements for lactating cows in "set it and forget it" mode, especially with cows entering the lactation in good health status. However, supply disruptions in late 2017/early 2018 brought vitamin discussions and supplementation strategies to the forefront. Technological advances include: herd status auditing, new product forms, and new understanding of modes of action beyond classical deficiency signs.

Introduction

In late 2017, several factors resulted in an unprecedented drop in global vitamin supply and subsequent rise in prices. This was coupled with shortages and outages in some specialty markets, such as certain vitamin forms for liquid feeds. In the case of vitamin A (retinyl acetate) and vitamin D3, prices reached 3 to 10 times greater than previous typical levels and local availabilities were widely affected (Figure 1). These price increases and shortages resulted in many discussions and strategies at all levels of the ruminant feed supply chain. Should cost of a nutrient be a factor in determining biological adequacy? It certainly was in early 2018, as nutritionists and producers scrambled to re-evaluate vitamin supplementation strategies, based on actual or perceived shortages and as an attempt to control input costs.

Vitamin A and Beta-carotene

Vitamin A is needed for eyesight, growth, reproduction, and maintenance of epithelial tissues. The activity of vitamin A is measured in retinol equivalents (1 IU of vitamin A equals 0.3 μg of all-*trans* retinol), and the most prevalent supplemental form is the retinyl acetate ester, usually encapsulated in a "cross-linked" gelatin beadlet for feed storage stability. Signs of vitamin A deficiency include: abortion, retained placenta, reduced immune function, and calf morbidity and mortality (NRC, 2001). As reviewed by Weiss (2018a), adjustments in the NRC vitamin A requirement should be made (upward) for ration forage comprising less than 60% of the ration, and for milk production > 75 lb, or for storage losses, which can amount to 9%/month in high-stress premixes containing inorganic trace minerals and choline (Shurson, 2011). Dietary beta-carotene (**BC**) is the major precursor of vitamin A with an activity of 400 IU per milligram for ruminants. Dietary BC is

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absorbed with fat and converted to retinol by intestinal enzymes. In ruminants, intact BC is also absorbed and stored directly, without immediate conversion to retinol. Guernsey and Jersey cattle convert less BC to retinol in the enterocyte, resulting in higher circulating levels and more excretion in milk than in Holstein cattle.

BC also functions separately from vitamin A as an antioxidant and can directly enhance immunity with possible reproductive and mammary benefits (Chew, 1993). The National Research Council (NRC, 2001) concluded that, although available data were insufficient to establish a BC requirement for dairy cattle, additional dietary vitamin A should be considered with low forage diets, high corn silage diets, diets with low quality forages, and situations with high pathogen loads or reduced immunocompetence.

Beta-carotene status

Responses to BC supplementation have been inconsistent in part due to the wide variation in serum BC status (Weiss, 1998; deOndarza et al, 2009). Most BC is found in vegetative plants and concentrations decrease with plant maturity. Most grains and fermented feeds contain minimal levels of BC because of heat damage and breakdown during storage (Pickworth et al., 2012). A serum BC level of 3.0 μg/ml has been suggested as the level at which supplementation is beneficial (Frye et al., 1991). A large proportion of serum samples from the 1996 NAHMS study of U.S. dairy herds (NAHMS, 1996) contained less than 3.0 μg/ml BC. LeBlanc et al. (2004) found mean serum BC concentration of 1828 samples from peripartum (+/- 1 wk) Holstein cows from 20 Canadian herds to be 1.12 μg/ml (SD = 0.78). Stage of lactation greatly affects serum BC levels (Kawashima, 2009a), with the lowest occurring immediately pre-calving (Figure 2).

Although the mode of action is not well understood (improved antioxidant/immune status?), some studies have found supplemental BC to positively effect milk yield. Heat-stressed cows supplemented with 400 mg BC increased cumulative milk yield by 11% (Arechiga et al., 1998). Oldham et al. (1991) supplemented 300 mg BC and increased milk yield by 6.4% with this difference approaching significance. However, others have not seen production responses with supplemental BC (Bindas et al., 1984; Rakes et al., 1985; Wang et al., 1988b).

Immune function

Chew et al. (1982) reported that cows with lower plasma vitamin A, BC, and total retinol equivalents had more mastitis. Chew (1983) supplemented 300 mg BC and 53 KIU vitamin A, or 80 KIU vitamin A, or 53 KIU vitamin A, or no supplement from 30 days before calving to 70 DIM. In this study, BC had a positive effect on immune response. Rakes et al. (1985) supplemented 300 mg BC and numerically lowered SCC content of milk, and Wang et al. (1988b) required fewer clinical mastitis treatments in cows supplemented with 300 mg BC.

Other researchers have not found indications that BC improved immune function. Oldham et al. (1991) did not reduce the incidence of mastitis with supplemental BC. Bindas et al. (1984) found that supplementing 600 mg of BC per day had no effect on SCC. LeBlanc et al. (2004) could not relate serum BC concentrations with either retained placenta or mastitis. However, they did find that when there was a 100 ng/ml increase in serum retinol concentration during the last week prior to calving, there was a 60% reduction in clinical mastitis in early lactation.

Reproduction

Dietary BC levels have been linked to fertility as evidenced by higher concentrations of BC in the ovary, particularly the corpus luteum (Chew et al., 1984). Schweigert (2003) postulated that BC is converted to retinol specifically in the uterus and ovaries. Graves-Hoagland et al. (1988) found plasma BC to be positively related to progesterone production by corpus luteum cells. Cows that ovulated during the first follicular wave postpartum had a higher mean plasma BC concentration than anovulatory cows three weeks prepartum (Kawashima et al., 2009a). In a follow-up study, Kawashima et al (2009b) supplemented BC during the close-up period (500 mg/day or 2000 mg/day in two different experiments) and increased the number of ovulating cows at the first follicular wave postpartum. Pregnancy rate at 120 days postpartum in heat-stressed cows supplemented with 400 mg BC/day for > 90 days was increased (35.4 vs. 21.1%; Arechiga et al., 1998). Rakes et al. (1985) found that supplementing 300 mg of BC for the first 100 DIM reduced days to first estrus and reduced cervix diameters at 21 and 28 DIM ($P < 0.05$). Lotthammer (1978, 1979) found that supplemental BC improved conception rates, uterine involution, and ovulation and reduced incidence of cystic ovaries and early embryonic death. Others have seen no positive reproductive responses to BC supplementation in dairy cattle (Bindas et al., 1984; Marcek et al., 1985; Wang et al., 1988a; Wang et al., 1988b) possibly due to season or initial BC status (Weiss, 1998). Greenburg et al. (1986) concluded that BC did not improve reproduction in beef heifers.

Colostrum quality, Beta-carotene, and calf issues

Calves are born with minimal vitamin A liver stores, making ingestion of colostrum with high vitamin A and BC concentrations imperative, as both have proven to be important for proper immune function. Kehoe et al. (2007) found that the BC concentration of colostrum from cows sampled across Pennsylvania varied from 0.1 to 3.4 µg/g. Torsein et al. (2011) found that calves born with serum levels below 0.25 ug/ml (up to 40% of the calves in high-mortality herds) were 5.3 times more likely to die than calves with higher serum BC levels.

Supplementing 1 g/day of BC increased BC concentration in colostrum compared to control (3.1 vs 1.44 mg/L, respectively; Kaewlamun et al., 2011; Table 1). Concentration of colostral BC was also increased in cows supplemented with 800 mg BC during the close-up period (Prom et al., 2016). The number of calves with detectable BC concentrations was higher for calves receiving maternal colostrum from dams supplemented with BC, compared to calves born from control fed dams. Recently, Aragona et al. (2017a) fed 700 mg BC/cow/day for 4 wk prepartum to determine effects on colostrum quality and calf performance. Colostral IgG concentration increased (82.7 vs 57.6 g/L for BC and control fed cows, respectively), although colostrum yield was reduced in BC-supplemented cows. Calves born from cows supplemented with BC gained 0.44 g/g DMI compared to calves born from cows not supplemented with BC that gained 0.32 g/g DMI ($P = 0.03$).

Evaluating Beta-carotene status and targeting supplementation

Because the actual BC content of diets varies and BC status was usually unknown in previous research, it can be difficult to evaluate BC supplementation strategies. Mean serum BC can now be assessed on the farm using the iEx™ system, a single step denaturation and BC extraction into organic solvent followed by BC measurement using iCheck® (BioAnalyt GmbH, Germany), a portable spectrophotometer (Schweigert et al., 2007). Routine BC measurements can be used to evaluate herd status and to recommend specific supplementation strategies in the field.

Vitamin D

Vitamin D requirements listed in NRC 2001 are listed as 18,000 to 25,000 IU/hd/day; levels adequate to prevent rickets and to assist in preventing milk fever. Hymoller et al. (2010) measured vitamin D synthesis in the skin of cows exposed to 5.4 hr of Danish sun/day, finding that exposure alone was adequate to support 20 to 25 ng 25-OH-D3/ml of serum, or just below adequacy. Since that time, researchers have focused on extra-rachitic responses, such as immunity (Lippolis et al., 2011) and gene expression (Viera-Neto et al., 2017). Nelson et al. (2016) surveyed 702 serum samples from US dairy herds, finding that most herds supplementing 30 to 50 KIU D3 maintained serum levels of 25-OH-D3 above the desired 30 ng/ml cutoff, with a mean observation of 69 ng/ml. However, one herd supplementing at 20 KIU saw 22% of the samples below 30 ng/ml.

25-OH-D3 approved for bovines

The 25-hydroxylated form of D3 (HyD®) was approved for bovine feeding in the US in October, 2018. Research with monogastrics in the US and with dairy cows in Europe and New Zealand has shown advantages in supporting skeletal health and peripartum calcium metabolism, along with many vitamin D-dependent reactions in immune tissue, muscle cell differentiation, and other extra-rachitic modes of action. Recent studies demonstrate that 25-OH-D3 positively influences calcium nutrition during different stages of the production cycle. The combination of 25-OH-D3 and diets with negative DCAD have been shown to reduce the nadir of Ca in plasma immediately post calving (Wilkens et al., 2012) and to reduce the incidence of diseases linked with sub-clinical hypocalcemia (Martinez et al., 2018ab). There have also been investigations into the use of 25-OH-D3 during lactation, with studies demonstrating an increase in absorption efficiency of both Ca and P, as well as a reduction in bone degradation and increase in bone formation (McGrath et al., 2012; Oehlschlager et al., 2014). Furthermore, a recent study also demonstrated a link between 25-OH-D3 and energy metabolism (Rodney et al., 2017). Viera-Neto et al. (2017) fed either 1 or 3 mg of D3 or 25-OH-D3 to pregnant, lactating Holstein cows. On day 21, researchers challenged cows with an intramammary *Streptococcus uberis* dose, and then measured rectal temperatures, mastitis severity, and several gene markers in milk somatic cells for 96 hours post-challenge. The severity of mastitis was decreased at 60 and 72 h post-challenge for 3 mg 25-OH-D3 vs. 1 mg D3 (Figure 3). These effects were explained in part by increasing availability of 25-OH-D3 for synthesis of 1,25-OH-D3 by 1α-hydroxylases in immune cells of infected glands.

Vitamin E

In transition cows, LeBlanc et al. (2004) found that, compared to healthy cows, cows that later contracted mastitis had lower serum alpha-tocopherol, retinol, and BC levels in the late dry period. Further, Goff et al. (2002) found that the metabolic draw of colostrum synthesis lowered serum status of all 3 vitamins versus mastectomized cows. Researchers at the University of Florida found that 2000 IU supplemental vitamin E had beneficial effects on milk production in heat-stressed multiparous cows, but no beneficial effects in first-lactation heifers (Staples et al., 2016). European researchers (Pottier et al., 2006) investigated the effect of very high (12,000 IU/hd/day) levels of vitamin E upon experimentally-induced milk fat depression. In their study, cows were fed 1.86 kg/day of extruded linseed plus 190 g/hd/day of linseed oil. The vitamin E treatment elevated milk from 3.29 to 3.88 % and decreased *trans*-10 C18:1 by 47%.

B Vitamins

With a few exceptions, B vitamins are thought to be in adequate supply from microbial synthesis. However, several researchers have challenged whether microbial supply has kept pace with increasing production in modern dairy cows (Shaver and Bal, 2000), or whether rumen-protected forms might do a better job of reaching the small intestine. Weiss (2017) pointed out that since 1990, the average Holstein synthesizes about 33% more milk and components today, but has only increased DMI by 15% - a possible imbalance between supply and need. Our understanding of B vitamin synthesis, rumen destruction, and kinetics has advanced (Castagnino et al., 2017), and several encapsulated combination B vitamin products have been tested with good results in North America (Sacadura et al., 2008; Morrison et al., 2018).

Biotin

As one of the exceptions, use of supplemental biotin (usually 20 mg/hd/day) is widespread in the US and Canada. In its unprotected (straight vitamin) form, biotin bypasses most rumen destruction and enters the bloodstream as-is (Zimmerly and Weiss, 2001). Two recent meta-analyses were published in 2011: Chen et al. (2011) found from their review that milk yield was increased by 1.66 kg when biotin was included, and Lean and Rabiee (2011) found a milk yield increase of 1.29 kg in their analysis. In addition, Lean and Rabiee (2011) evaluated effects on hoof health, concluding that although studies were lacking in consistency of hoof measurements, overall biotin had a consistently positive effect on hoof health.

Niacin

Schwab et al. (2005) analyzed niacin lactation studies with dairy cows, finding that 12 grams supplemental increased 3.5 of FCM by 0.5 kg/day in a meta-analysis. Cost of supplemental niacin in mid-2019 would be about 10 cents/hd/day, so economic returns would be marginally positive. Morey et al. (2011) investigated a rumen-protected niacin source with early lactation cows, finding that 9.6 g niacin in protected form reduced NEFA. Aragona et al. (2017b) supplemented 16, 32, or 48 g of niacin to prefresh cows from -28 days to calving, and measured colostrum IgG concentration and yield, and calf performance. They found that supplemental niacin resulted in a linear increase in colostral IgG concentration up to the highest niacin level, and that calves born to dams receiving 32 g/day of supplemental niacin had ADG greater than the controls and other treatment levels.

Rumen-Protected Choline

Choline is usually considered in the water-soluble vitamin category, although requirements are in gram amounts rather than milligrams. Unprotected dietary choline is rapidly degraded in the rumen, but serves important functions in fat transport when absorbed. Rumen protected choline has been widely researched in transition and early lactation. Weiss (2018b) summarized 6 studies where 50 g of rumen protected choline was fed, noting a significant milk increase of 2.3 kg for the first 2 months of the lactation.

Conclusions

Research has shown that current recommended supplemental levels of vitamins A, D, and E are adequate to support excellent health and production in lactating dairy cows. Along with biotin, rumen-protected choline, and BC, these 6 vitamins are those most widely supplemented for dairy cows. Advances have been made in vitamin status auditing with several cowside assays available for serum retinol, alpha-tocopherol and BC, which can be used to tailor recommendations to ration and herd status. Our understanding of vitamin adequacy includes scientific bases for more than classical deficiency symptoms, including immune status, colostrum production, calf health, or storage losses. Vitamins not considered essential for adequacy but which are widely used for economic benefits also include biotin and rumen-protected choline. Future advances will include a better understanding of vitamin responses, the role of antioxidants in disease prevention, and rumen-protected forms of vitamins possibly marginal or limiting in high-producing dairy cows.

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Table 1. Colostrum responses to beta-carotene supplementation.

Study	Beta-carotene (mg/day)	IgG, mg/ml	BRIX%	Colostrum BC, mg/L	Calf Serum BC, ug/ml	Calf Serum IgG, mg/L
Kaewlamun et al. (2011)	1000/14 days			↑215% ($P < 0.01$)		
Prom et al. (2014)	800/21 days	↑3.4% (NS[1])	↑2.0% (NS)	↑239% ($P < 0.01$)	↑566% ($P < 0.05$)	--
Aragona et al. (2017a)	700/28 days	↑ 43% ($P < 0.05$)	--	--	NS	↑41% ($P < 0.01$)

[1] NS = Not significant.

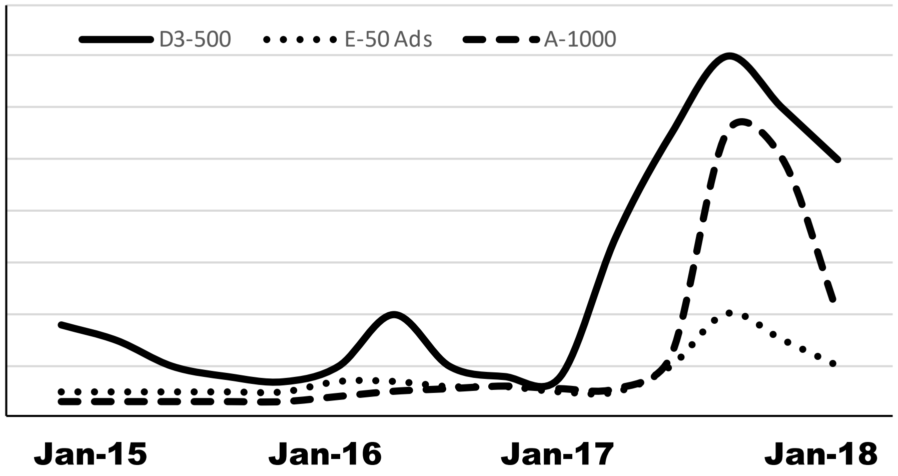

Figure 1. Vitamin prices in North America, 2015-2018 (DSM, 2019, personal communication).

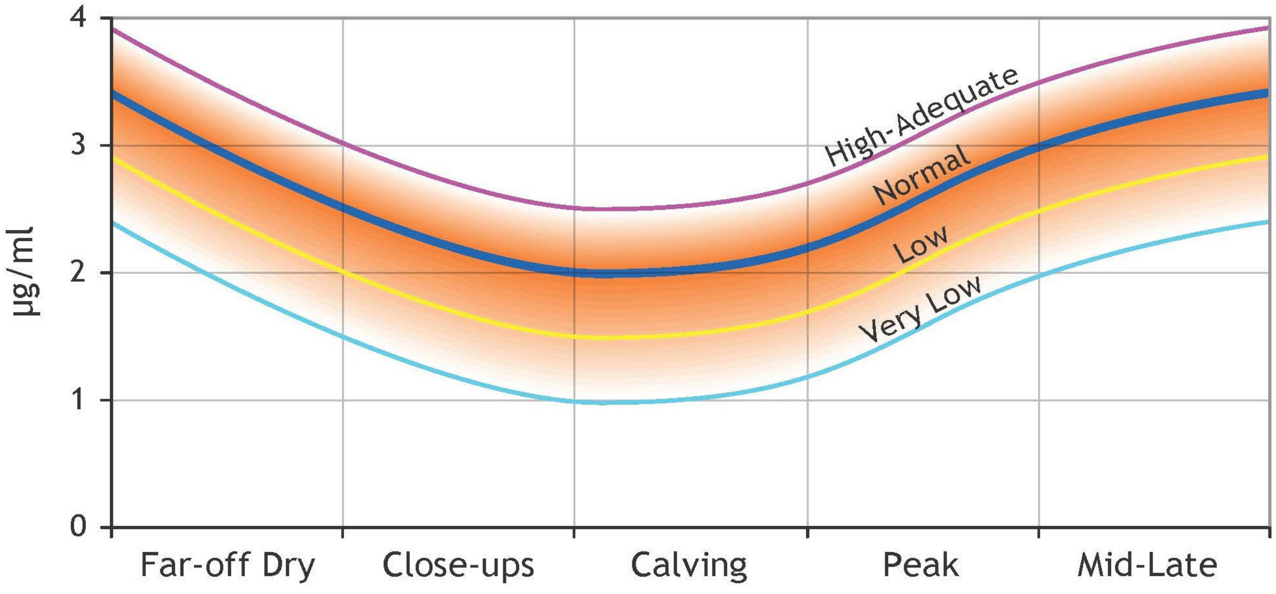

Figure 2. Herd whole blood β-carotene means from North American dairy herds, 2018 (DSM).

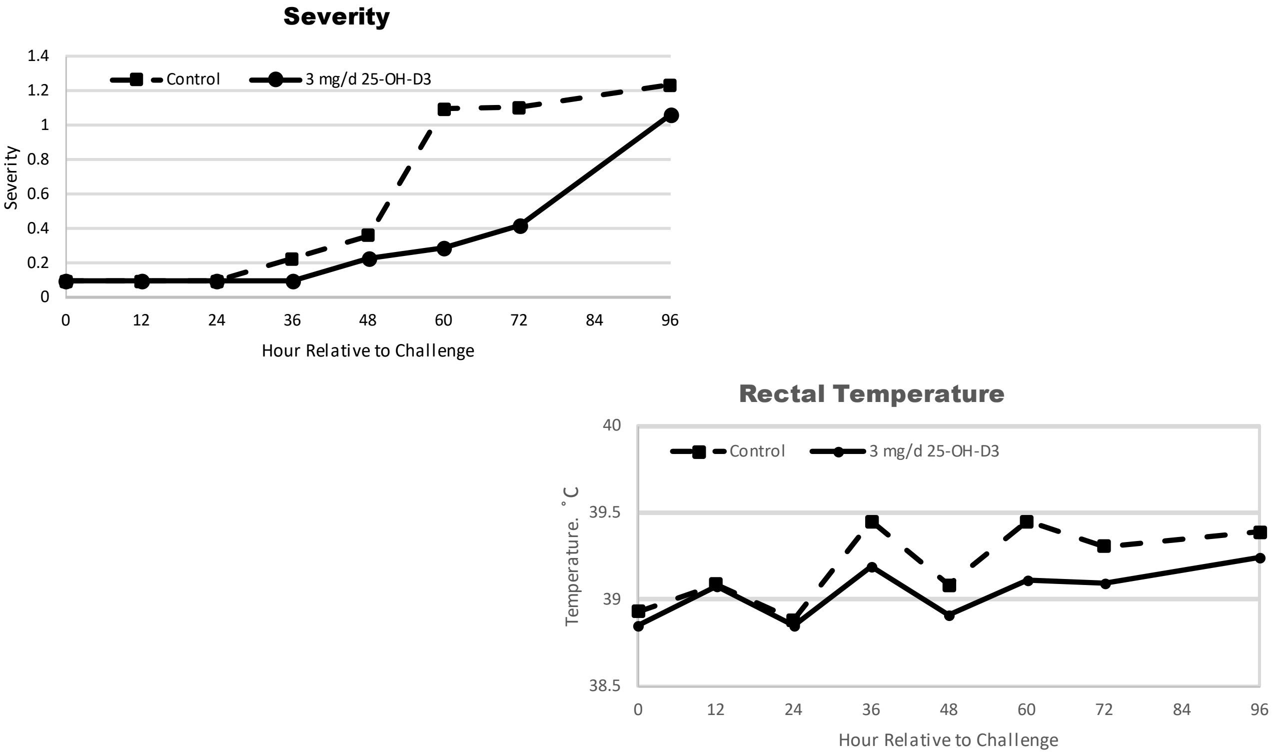

Figure 3. Effects of Vitamin D source on mastitis severity. 30 cows fed 1 mg vitamin D3 (Cholecalciferol) or 3 mg 25-hydroxyvitamin D3. Challenged with intramammary *Streptococcus uberis* at 21 day of treatment. (Viera-Neto et al., 2017).