

## **RELATIONSHIP OF MINERAL AND VITAMIN SUPPLEMENTATION WITH MASTITIS AND MILK QUALITY**

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Several trace minerals and vitamins have a direct effect on the function of the immune system and therefore could have a direct effect on the ability of a cow to combat mastitis. Smith et al. (1984) was the first to show a direct link between mineral and vitamin (selenium and vitamin E) nutrition and mastitis. Since that paper was published other research has shown relationships between vitamin A and B-carotene and mastitis. A limited amount of published data is available on the effects of copper and zinc on mastitis. The purpose of this paper is to briefly review clinical data on the effects of minerals and vitamins on mastitis, discuss how the newly published nutrient guidelines (NRC, 2001) pertain to mastitis, and present nutritional guidelines with respect to improved mammary gland health.

### Trace Minerals and Vitamins Involved with Immune Function

Every essential nutrient of a cow is probably related to immune function. However, this paper will discuss only those trace minerals and vitamins that are known to directly influence immune function. Most of the trace minerals and vitamins that affect immune function are part of the cellular antioxidant system (Table 1). Known antioxidant pathways suggests that the requirements of antioxidant nutrients are interrelated. A deficiency of one of the antioxidant nutrients may increase the requirement of another nutrient. Vitamin A is not a strong antioxidant but it is extensively involved in many aspects of immune function. Other antioxidant nutrients also may influence immunity independent of their antioxidant roles.

Simply because a nutrient is directly involved with immune function does not mean that supplementing diets with that nutrient will improve immunity and reduce mastitis. Cows need to consume a certain quantity of biologically available minerals and vitamins to maintain optimal status. A cow is in optimal status when she has adequate amounts of trace minerals and vitamins for maximal production and to maintain good health. When cows are below optimal status, supplementation of a biologically available form of the nutrient should elicit a positive response but once optimal status is obtained, no additional positive responses would be expected when the nutrient is supplemented. Excessive supplementation may decrease immune function and increase health problems. The likelihood that a cow will respond to mineral or vitamin supplementation is a function of the amount of nutrient the cow would consume with an unsupplemented diet. Selenium in soil (and the resulting crops) is low in the major dairy areas of the U.S. which means that selenium supplementation often elicits a positive response. Many feeds, however, contain substantial concentrations of manganese so a response to supplemental manganese is much less likely.

Table 1. Antioxidant systems of mammalian cells.

Component (location in cell)	Nutrients Involved	Function
Superoxide dismutase (cytosol)	Copper and zinc	An enzyme that converts superoxide to hydrogen peroxide
Superoxide dismutase (mitochondria)	Manganese and zinc	An enzyme that converts superoxide to hydrogen peroxide
Ceruloplasmin	Copper	An antioxidant protein, may prevent copper from participating in oxidation reactions
Glutathione peroxidase (cytosol)	Selenium	An enzyme that converts hydrogen peroxide to water
Catalase (cytosol)	Iron	An enzyme (primarily in liver) that converts hydrogen peroxide to water
$\alpha$ -tocopherol (membranes)	Vitamin E	Breaks fatty acid peroxidation chain reactions
$\beta$ -carotene (membranes)	$\beta$ -carotene	Prevents initiation of fatty acid peroxidation chain reactions

### Brief Review of Clinical Data

Numerous experiments have been conducted examining the influence of nutrients on immune function in vitro and in vivo; several of which used cattle. Data from these experiments are useful in understanding mechanisms and to suggest possible links to health disorders; however, altered immune status does not necessarily mean altered health of a cow. Epidemiological studies also can suggest relationships between nutrient status and mammary health, but results are equivocal. Clinical trials are needed to determine whether a nutrient affects health. A well-designed clinical trial has enough animals for statistical validity and uses accepted disease measurement protocols. Clinical trials measuring the impact of nutrition on mammary gland health (e.g., intramammary gland infections, clinical mastitis, etc.) are extremely limited.

*B-carotene and Vitamin A.* Two clinical trials to study the effects of vitamin A and (or) B-carotene on mammary gland health of dairy cows were conducted at Washington State University. In one study, cows were fed 53,000 IU of supplemental vitamin A/day, 173,000 IU/d or 53,000 IU/day plus 300 mg of supplemental B-carotene/day starting 3 wk before dry-off and continuing through the dry period (Dahlquist and Chew, 1985). Cows fed the vitamin A plus B-carotene treatment had fewer new infections during the early dry period (27% of previously uninfected quarters) than cows fed the other treatments (49% for low vitamin A and 50% for the high vitamin A treatments). In another trial the same treatments were used but supplementation started 3 wk before calving and continued through 10 wk of lactation (Chew and Johnston, 1985). The vitamin A plus B-carotene treatment reduced mean SCC during wk 2 to 8 of lactation (85,000 cells/ml compared with 225,000 and 125,000/ml for low and high vitamin A treatments). Oldham et al. (1991) conducted a study using similar treatments (50,000 IU supplemental vitamin A/day, 170,000 IU/day, and 50,000 IU/day plus 300 mg of B-carotene/day)

starting 2 wk before dry-off and continuing until 6 wk of lactation. In that study, treatment did not affect new infections during the dry period (mean = 11.3% of quarters), new infections at calving (mean = 6.4%), total new infections during the experiment (27.5%), and cases of clinical mastitis (9.4% of quarters). A likely reason for the difference between the Washington State studies and Oldham et al. is a difference in vitamin A and B-carotene status of the control cows. At the start of the Dahlquist and Chew (1985) experiment, mean plasma concentration of B-carotene was 2.5 mg/L, but in the Oldham et al. study, the mean concentration was 10 mg/L. Jukola et al. (1996) suggested that plasma concentrations of B-carotene in dairy cows should be >3 mg/L to optimize udder health.

*Selenium and Vitamin E.* Smith et al. (1984) reported that dry cows fed approximately 1000 IU of supplemental vitamin E/day during the 60 day dry period and (or) injected with approximately 50 mg of selenium 21 day before expected calving had fewer cases of clinical mastitis with reduced duration compared with cows not fed vitamin E or injected with selenium. Malbe et al. (1995) reported that supplementation of a very low selenium diet with 0.2 ppm of selenium from selenite or selenized yeast reduced SCC in lactating dairy cows. In a study from New Zealand (Wichtel et al., 1994) mean SCC over an entire lactation was reduced or tended to be reduced when cows were supplemented with 6 to 12 mg of selenium/day, however, no difference in clinical mastitis was found between treatments. Erskine et al (1989) reported that feeding cows 2 mg of supplemental selenium/day starting 3 months before calving and throughout lactation reduced the severity and duration of mastitis caused by experimentally challenging cows with *Escherichia coli*. Control cows in that study were fed a diet with 0.04 ppm Se. In a similar study, Erskine et al. (1990) found no effects of supplemental selenium on mastitis when cows were experimentally challenged with *Staphylococcus aureus*. Weiss et al. (1997) reported that feeding 1000 IU/d of supplemental vitamin E during a 60 day dry period to cows with low selenium status reduced clinical mastitis during the first week of lactation compared with feeding 100 IU/day. In that same study, feeding 4000 IU of supplemental vitamin E/day during the last 14 days of the dry period further reduced clinical mastitis and reduced new infections at calving. Clinical mastitis incidence during the first week of lactation was 25.0, 16.7, and 3.6% of quarters for cows fed 100 IU/day, 1000 IU/day, or 4000 IU/day during the first 46 days of dry period followed by 4000 IU/day during the last 14 days of the dry period. An important feature of that study was that cows were fed a low selenium diet (0.1 ppm) and had marginally low plasma selenium concentrations (0.048 mg/L).

*Copper.* No peer-reviewed publications were found from clinical trials on the effects of copper on mammary gland health, but two reports that have not been peer-reviewed are available. Harmon and Torre (1994) used heifers that received no supplemental dietary copper since weaning and fed them a diet with 0 or 20 ppm supplemental copper (from copper sulfate) from 84 days before calving through 105 days of lactation. At calving, heifers fed supplemental copper had fewer quarters infected than unsupplemented animals. Following an *E. coli* endotoxin challenge, cows supplemented with copper had lower clinical score and fewer abnormal quarters than unsupplemented cows. Supplementation had no effect on the response to a *S. aureus* challenge. Scaletti et al. (2000) fed heifers a diet with about 7 ppm total copper or the same diet plus 20 ppm supplemental copper (from copper sulfate). Copper supplementation

reduced the severity of mastitis following experimental challenge with *E. coli*. Based on liver copper concentrations, the unsupplemented group was copper deficient.

*Zinc.* Peer-reviewed clinical data are also extremely limited on the effects of dietary zinc on mammary gland health. Several studies (mostly non-peer reviewed) have examined the effect of supplemental zinc from zinc methionine on SCC. Supplementation was typically about 360 mg of zinc/day and in most studies the control and treatment diets were not equal in zinc concentrations (any effect could be caused by source or amount of supplemental zinc). In most studies zinc methionine supplementation statistically or numerically reduced SCC (see review by Kellogg, 1990). Only one study on zinc was found in which incidence of mastitis and infection rates were measured. Whitaker et al. (1997) compared the effects of providing supplemental zinc from a mixture of zinc proteinate (250 mg of zinc/day) and inorganic zinc (140 mg/day) or from all inorganic sources (390 mg of zinc/day). Diets contained approximately 50 ppm total zinc (about 25 ppm supplemental and 25 ppm from basal diet). Source of zinc had no effect on infection rate, new infections, clinical mastitis and SCC. More experiments similar to Whitaker et al. are needed to determine whether increasing zinc intake of cows is directly related to mammary gland health.

*Manganese.* No clinical studies have been published on the effect of manganese on mammary gland health.

#### NRC 2001 Requirements for Trace Minerals and Vitamins

In 2001, the NRC released the latest version of the Nutrient Requirements of Dairy Cattle. For some trace nutrients, immune function and clinical mammary gland health data were used to establish requirements. The requirements for trace minerals and vitamins published in that book should be the starting point when formulating diets. No currently available data suggests that supplementation above NRC (2001) requirements will improve mammary gland health except in specific, limited situations. The NRC requirements assume that bioavailability of minerals and vitamins are normal, but if excessive antagonists are consumed, NRC requirements may not be adequate. For example, diets (or drinking water) that provide excessive amounts of sulfur and molybdenum reduce availability of copper and NRC requirements may not be adequate.

Trace mineral requirements in the NRC are given as milligrams of each mineral that must be absorbed each day, not as dietary concentrations. To convert NRC requirements to dietary concentrations, one must know dry matter intake and the absorption coefficient of the mineral. Intake can be measured on-farm or estimated using equations. Average absorption coefficients for common sources of supplemental minerals are in NRC (2001). For vitamins, NRC recommendations are expressed as IU of supplemental vitamin needed unit of body weight.

*Vitamin A.* The new NRC increased the vitamin A requirement of dry and lactating cows to 110 IU/kg of body weight (50 IU/lb). On a daily basis, an average lactating Holstein cow should consume about 72,000 IU of supplemental vitamin A/day, and an average dry Holstein cow should consume about 77,000 IU/day. Based on clinical data, no improvement in mammary gland health should be expected at supplementation rates greater than these requirements.

*B-carotene.* The NRC did not establish a requirement for B-carotene. Based on clinical data, mammary gland health may be improved with B-carotene supplementation when cows are in low B-carotene status. Concentrations of B-carotene in feeds or plasma are not normally measured in the field, therefore B-carotene status is usually not known. The cost of supplementing 300 mg of B-carotene/day is substantial and should not be done routinely. Diets that contain a large proportion of weathered hay may be low in B-carotene and supplementation might be useful. Diets based on good quality silage or fresh forage probably provide adequate B-carotene and supplementation would not be economical.

*Vitamin E.* The new NRC, based on clinical mammary gland health data, substantially increased the vitamin E requirement. The new requirement for a dry cow is 1.6 IU/kg of body weight (0.7 IU/lb) and 0.8 IU/kg of body weight (0.36 IU/lb) for a lactating cow. This equates to about 500 IU of supplemental vitamin E/day for a lactating Holstein cow and 1000 IU/day for a dry Holstein cow. Because fresh forage usually has a very high concentration of vitamin E, cows that consume a substantial amount of pasture or green chop do not need as much supplemental vitamin E. The NRC suggest that grazing cows be supplemented with approximately 160 IU/day (lactating cows) or 330 IU/day (dry cows). Limited data suggest that supplementation of vitamin E above NRC might be necessary to optimize mammary gland health when periparturient cows are in marginal to poor selenium status.

*Selenium.* The concentration of supplemental selenium in dairy diets is regulated by the U.S. FDA at 0.3 ppm and the new NRC set the selenium requirement at 0.3 ppm for dry and lactating cows. No clinical data are available suggesting that more than 0.3 ppm supplemental selenium will improve mammary gland health.

*Copper.* The new NRC increased the copper requirement of lactating and dry cows. Assuming normal bioavailability and typical ingredients an average dry Holstein cow needs to consume 170 to 180 mg of copper/day. An average Holstein cow producing 50 or 100 lbs of milk needs to consume 225 mg or 300 mg of copper/day, respectively. These recommendations assume normal dietary concentrations of sulfur and molybdenum. When diets or drinking water contain excessive concentrations of sulfur and molybdenum these copper recommendations will not be adequate. Clinical data showing positive responses to copper supplementation at 20 ppm (total diet copper was approximately 25 to 27 ppm) used control diets that were below NRC requirements. An improvement in mammary gland health to copper supplementation would be less likely if control cows were fed copper at NRC recommendations. Relative to requirements, copper is the most toxic mineral routinely supplemented (recommended safe concentration is only four to five times the requirement); excessive supplementation of copper should be avoided.

*Zinc.* Assuming normal bioavailability and common ingredients the new NRC zinc requirement is about 300 mg/day for an average Holstein dry cow, about 900 mg/day for a cow producing 50 lbs of milk, and 1400 mg/day for a cow producing 100 lbs of milk. Because clinical data are lacking, it is not known whether these concentrations are adequate for optimal mammary gland health. Cows can tolerate relatively high dietary concentrations of zinc (at least ten times requirements without adverse effects), however excessive zinc supplementation can cause a

secondary copper deficiency. Because of the interaction between zinc and copper, dietary zinc should not exceed five times the concentration of dietary copper.

*Manganese.* The new NRC requirements for manganese are about 240, 280, and 350 mg/day for a dry cow, a cow producing 50 lbs of milk, and a cow producing 100 lbs of milk, respectively. These values are substantially lower than previous recommendations. No data are available on the effects of manganese on mammary gland health. Manganese has a high safety factor (approximately 50 times requirement).

### Recommended Dietary Concentrations

The approach followed by NRC of expressing requirements in either milligram or IU/day is scientifically correct and accounts for important sources of variation (i.e., variation in intake and bioavailability). However, the NRC was designed to evaluate the nutritional adequacy of diets rather than formulating diets. The NRC values should be considered minimum requirements--they do not include any safety margins. Because of variation in intake, environment and feed composition, diets should be formulated to provide more than recommended amounts of some minerals to ensure all cows consume adequate amounts of nutrients. The guidelines in Table 2 are based on NRC requirements plus some adjustments for some expected variation.

Table 2. Suggested dietary concentrations (dry matter basis) of trace nutrients<sup>1</sup>.

	Nonlactating cows			Lactating cow (milk yield)	
	Dry	Pre-fresh	Fresh cow	50 lbs	100 lbs
Est. intake, lbs/day	30	22	30	44	58
Vitamin A, IU/lb	3300	4500	3300	1850	1500
Vitamin E, IU/lb	35	50	25	12	10
Selenium, ppm	0.3	0.3	0.3	0.3	0.3
Copper, ppm	20	20	15 - 20	15 - 20	15 - 20
Manganese, ppm	30 - 50	40 - 50	40 - 50	30 - 40	30 - 40
Zinc, ppm	40 - 60	50 - 70	60 - 80	50 - 70	60 - 80

<sup>1</sup>Values in the table are for a Holstein cow with an average body weight for various stages of lactation and gestation. Pre-fresh is for cows in the last 2 wk of gestation. Fresh is for cows in the first 3 wk of lactation.

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