

# Trace Minerals in Production and Reproduction in Dairy Cows

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## ■ Take Home Messages

- Fertility in dairy cows has declined significantly in the past 30-50 years
- Factors that control the health of the follicle and oocyte are poorly understood
- Trace minerals have a significant role to play in many aspects of production including fertility
- Improvement in reproductive activity in males and females has been associated with supplementation of minerals, particularly when given in the organic state.

## ■ Introduction

The changing trends in animal production require the owner/manager to become more efficient in all aspects of the production cycle. Major contributors to economic returns include reproduction and nutritional management of the herd. While both of these issues are important in their own right, recent data indicate substantial interrelationships between them. To maintain high milk output, cows should give birth at the shortest possible intervals, which means that there is a very tight schedule, taking account of post-partum uterine involution, anestrus and the duration of pregnancy in the cow. The time of rebreeding may coincide with a period of high milk production and possibly with a period of negative energy balance. These nutritional stresses can affect the reproductive system and may be manifested as delayed return to cyclicity and failure to become pregnant. Successful establishment of pregnancy requires a series of coordinated signals involving the brain, ovary and uterus, all of which may be influenced nutritionally at the macro- or micro-level. Efficient production in domestic animals requires that the essential nutrients in a diet be provided in appropriate amounts and in forms that are most biologically useful.

The relationship between nutrition and reproduction in ruminants is complex and often quite variable. However, nutrient supply is a component of the management system that is under the control of the producer and needs to be carefully evaluated. Nutrient requirements to support follicle growth, ovulation and early pregnancy are extremely low (less than 3 MJ/day) compared with requirements for maintenance and production (60-140 MJ/day). Nevertheless, in the case of lactating cows, inadequate nutrition in the short term, or as a consequence of a prolonged depletion of body reserves during early lactation, can have significant deleterious effects on resumption of ovarian activity post-partum, conception and fertility. In addition, the deleterious effects of nutrition around mating on embryo development are evident. In cattle, the degree of energy deficit during the first few weeks after calving is closely correlated with the interval to first oestrus, and other markers of reproductive success such as conception rate to first service, services per conception and calving to conception interval. In addition differences in negative energy balance (NEB) among cows is large and the NEB is more severe in third parity or older cows than in first parity animals (5).

Dietary factors can affect reproduction by influencing the animal at the level of the hypothalamic-pituitary gland, the ovary and/or the uterus. Ovulation occurs following a clearly defined procedure of follicle growth and oocyte maturation. While the initial stages of follicle growth are independent of gonadotropins, FSH and LH support is required for a follicle to proceed to the ovulatory stage and ultimately to allow resumption of meiosis of the oocyte and ovulation. Between ovulations, there are recurrent waves of follicular growth and regression. The regulation of these waves of growth, and ovulation, is controlled by an interaction between pituitary secretions of LH and FSH, hormones of ovarian origin (the most important of which are estradiol-17 $\beta$ , progesterone, inhibin), and growth factors such as IGF-1 and prostaglandin F2 $\alpha$ . While minor differences exist in follicle turnover characteristics in cattle and sheep, the physiological mechanisms are generally similar. The physiological processes responsible for follicle emergence, dominance, atresia and ovulation are now well understood; what is not so well known is the effect of nutrition and trace mineral supplementation on production and reproduction parameters.

The pattern of folliculogenesis is a series of physiological events involving both germ and somatic cells in the growth, differentiation, maturation and either atresia or ovulation of the dominant follicle (DF) (25). Following parturition, there is a temporary suppression of turnover of DF that normally occurs during other physiological states in cattle such as during the estrous cycle, prior to puberty or during early pregnancy (24). Roche & Diskin (25) indicate that there are 2 endocrine phases to estrous cycle resumption post-partum. The pattern of emergence, selection and dominance of follicles has been established in post-partum dairy (27) and beef suckler cows (20). Because of their role in the endocrine system and in tissue integrity, minerals have a beneficial role to play in resumption of follicular growth and fertility in dairy cows.

## ■ Genetics and Reproduction

As the genetic capacity for milk production has increased in dairy cattle over time, there has been a tendency for fertility to decrease (17;26). There have been trends to feed high levels of crude protein to enhance milk production, but this can be associated with decreased fertility (3). Imbalances in the relative availability of protein and energy may affect efficiency of metabolism and energy status. Some reports indicate the use of lipids that are protected from hydrolysis in the rumen with a view to reducing the post-partum interval. However, these treatments have often been associated with reduced embryo survival, possibly due to excessive estrogen production from the increased follicular growth (15).

Reproductive efficiency and embryo mortality have been widely studied in heifers (23), dairy cows (13) and beef cows (30). Embryo loss rates of the order of 40-50% contribute in a significant fashion to economic inefficiency in cattle production and ways of alleviating this must be pursued with vigor.

## ■ Minerals

Minerals are essential for growth and reproduction and are involved in a large number of digestive, physiological and biosynthetic processes within the body (4). The most obvious function is as components of body organs and tissues and to provide structural support. In addition, they act as electrolytes, as constituents of body fluids and as catalysts in both enzyme and hormone systems. They therefore fulfil several important functions for the maintenance of animal growth and reproduction as well as health status (31).

Proper herd management should be designed to optimize the production of the highest quality product, while minimizing any adverse effects on the health and welfare of the animals (8). In dairy cattle two key goals are adequate nutrition and adequate mammary health so as to produce wholesome milk. Recent data indicate that micronutrient management will enhance the production of good quality milk. The potential for minerals to play a significant role in herd fertility is indisputable. The mineral elements that are of particular importance are categorised into major (calcium, phosphorous, potassium, sodium, chlorine, sulphur and magnesium) and trace elements (iron, iodine, copper, manganese, zinc, cobalt, molybdenum and selenium). The minerals that affect reproduction in cattle are generally found within the trace element group, although deficiencies of calcium and phosphorus can also affect fertility. Organic minerals have a beneficial role to play in resumption of follicular growth and fertility in dairy cows. Replacing sodium selenite with organic selenium resulted in fewer services per conception in a study using 1800 cows (16). The use of organic chromium compared with inorganic forms, has been shown to enhance

the survival of rats and to increase litter size in gilts and sows (14). Reproductive problems are frequently reported in association with trace mineral deficiencies, particularly copper, selenium and manganese.

The key to the effectiveness of a mineral supplement is not necessarily its biological availability, but its biological activity (16). Organic minerals have been shown to have several beneficial effects in ruminant and monogastric animals. There are still discernible differences among chelated minerals, mineral proteins and other organic minerals complexes. Proteinated minerals can possibly improve female reproduction through increased fertilization, lower embryo mortality, improved uterine environment and/or increased intensity of estrous behaviour.

The importance of copper (Cu) as an essential trace element has been recognized for over 70 years, with the early discovery that Cu was necessary for normal haemoglobin synthesis in young rabbits and rats. Since that time, the importance of Cu for normal growth, production and reproductive performance has been established. The biological role of Cu is exerted through a number of Cu-containing proteins including ceruloplasmin and superoxide dismutase (SOD) (22). When Cu is inadequate in animals, physiological and metabolic functions related to the Cu-enzymes may be impaired and, during clinical deficiency, symptoms will appear. Although low Cu content of feedstuffs is a common cause of Cu inadequacy, reducing bioavailability of Cu in ruminants may occur when dietary sulphur, molybdenum, zinc or iron are high (11).

Zinc deficiency in ruminants has been postulated to weaken skin and other stratified epithelia as well as reducing the magnitude of basal metabolic rate following infectious challenge (8). Zinc is a cofactor for many proteins and enzymes involved in the acute phase response to infection and inflammation (21). Because the mammary gland is a skin gland, it is highly likely that zinc will have a positive role in its protection. Skin integrity of the teat has been shown to be especially linked with mastitis prevention. Kellogg (12) reported that chelated zinc decreased somatic cell counts (SCC) by 22-50% in eight trials, depending on the dose of zinc used, and increased milk production. Perhaps cell integrity may have been involved. The keratin lining of the teat canal has been described as a physical and chemical barrier for protection of the mammary gland. Keratin lining may physically trap bacteria and prevent migration into the mammary gland. Most studies in this area have focused on reducing SCC during supplementation with organic zinc as this is more readily available to ruminants than inorganic forms (Harmon, 1998). Supplementation with Bioplex zinc (Alltech) decreased the rate of new intramammary infections (28).

Zinc activates several enzyme systems and is a component of many metallo-enzymes. It plays a vital role in hormone secretion, especially relating to

growth, reproduction, immunocompetence and stress. Zinc is also involved in the generation of keratin and in skin nucleic acid and collagen synthesis. It is essential to the integrity of the immune system, in cation/anion exchange (and therefore water balance) as well as in the maintenance of normal vitamin A concentration in plasma and in ovarian function. Many animals therefore require supplemental Zn in the diet for normal body function because of either low levels in the dietary ingredients or the presence of antagonistic factors which decrease the bioavailability of the element. Antagonism might be due to metal ion interactions such as with Fe or Cu. Source of fibre has also been reported to decrease the availability of Zn (1).

Manganese (Mn) is involved in the activities of several enzyme systems including hydrolases, kinases, decarboxylases and transferases as well as Fe-containing enzymes which require Mn for their activity. It is therefore involved in carbohydrate, lipid and protein metabolism. It is also needed for bone growth and maintenance of connective and skeletal tissue. Mn also plays a role in reproduction and in immunological function. In pigs, Mn deficiency results in abnormal skeletal growth, increased fat deposition, reproductive problems and reduced milk production.

Selenium (Se) is a semi-metal that is very similar to sulphur in its chemical properties. It is an essential constituent of the glutathione enzyme system; and a deficiency of Se will leave cells vulnerable to oxidation and increase the requirement for Vitamin E. It has therefore been usual to supplement the diets of all classes of pigs with inorganic sources of Se, such as sodium selenite. However, recent research suggests that organic selenium is more available and therefore more effective than inorganic forms in meeting requirements (120 to 150% available relative to sodium selenite). In first parity gilts, tissue Se stores were substantially higher following organic Se (Sel-Plex 50) than inorganic Se (18).

## ■ Effects of Organic Minerals on Mammary Gland Health and Reproduction

Traditionally, inorganic salts such as oxides, sulphates and carbonates have been added to the diet to provide the desired amount to meet the requirements of the animal. These are broken down to varying extent during digestion to 'free' ions and are then absorbed. However, they may also complex with other dietary molecules and become difficult to absorb or, if completely complexed, totally unavailable to the animal. Thus, the availability of the element may vary substantially. Because of these uncertainties, the levels provided in the diet are often higher than the minimum required for optimum performance, often resulting in over-supply and unnecessary wastage with obvious environmental impact.

Many salts occur in nature as proteinates or chelates. Chelates may utilize peptide or amino acid uptake pathways rather than normal mineral ion uptake pathways in the small intestine. This prevents competition between minerals for the same uptake mechanisms. Not only is bioavailability therefore higher, but these mineral forms are more readily transported and hence their intestinal absorption is also enhanced. They are more stable and are protected biochemically from the adverse reactions with other dietary nutrients that could reduce their rate of absorption. It is also thought that they can be specifically targeted at certain organs, tissues or functions in the body.

Mastitis is a significant management challenge to dairy producers. Nutritional factors related to immune status include protein/energy nourishment, trace mineral and vitamin nutrition (28). Extensive research indicates that selenium and vitamin E have critical roles in protecting the body from infection (6). Vitamin E is important in protecting the cell membranes from oxidative damage, while selenium is a component in the enzyme glutathione peroxidase, which protects the cells from internal damage when fighting infection. Selenium supplementation increased SCC resistance to intra-mammary infusion of *Escherichia coli*. Erskine et al. (6) demonstrated lower blood selenium concentrations in cows with high SCC compared with cows with low SCC.

Studies at the University of Kentucky have attempted to elucidate the role of Cu status on inflammation and infection of the mammary gland (9). Holstein heifers were assigned to two dietary treatments (+Cu and – Cu) from 84 days prepartum through 105 days of lactation. Liver Cu levels in the +Cu and –Cu cows were 209 and 14 ppm at calving and 474 and 20 ppm at 105 days postpartum. However, plasma Cu in both groups of heifers was within a normal range. The +Cu cows had more uninfected quarters (60%) than the -Cu cows (36%) at calving (Table 1). Several studies have demonstrated a reduction in SCC in dairy cattle that were supplemented with mineral proteinates. It is well established that copper interacts with both iron and zinc as well as with other elements. High levels of either zinc or iron can create copper deficiency and the reverse can also happen. The role of Cu in the immune response is well established. Cattle with no outward signs of Cu deficiency had reduced superoxide dismutase activity and a reduced ability to kill *Staphylococcus aureus*, a primary pathogen of mastitis. Therefore studies have concentrated on using proteinates as a method of reducing or eliminating competition with other elements. The results indicate that Cu proteinates have a higher bioavailability than Cu sulphate. The implications are that Cu proteinate may be absorbed as in the organic form and transported in blood without binding to ceruloplasmin, which is the primary transport system when inorganic Cu is the dietary source.

Harris (1995) reported results of a 90-day field trial in which one group of cows received a total mixed ration (TMR) supplemented with 400 mg Zn per cow per day as Bioplex Zinc and the control group was fed the normal TMR. The mean SCC in the Zn proteinate group decreased 24% and the SCC in the control

group increased 36%. SCC was 57% lower in the supplemented group at trial end. Adjustment for the lower SCC in the Zn proteinate group at initiation of the trial would still show an estimated 30 to 40% lower SCC in the Zn proteinate cows.

**Table 1. Effects of pre-partum treatment with copper treatment on liver Cu concentrations (9).**

	+ Cu	-Cu
Liver Cu @calving, ppm	209	14
Liver Cu @105 days pp, ppm	474	20
Uninfected quarters @ calving	60%	36%

We carried out a series of experiments examining the effects of organic minerals on production and reproduction in dairy cows (2). In Experiment 1, forty nine pregnant Friesian cows were selected 2 weeks prepartum and were paired on the basis of : (i) calving date, (ii) parity and (iii) milk yield and each pair was randomly allocated to either control or supplement with Bioplex (100mg Cu, 300mg Zn, 2mg Se) dairy minerals. Cows were housed and grass silage of approximately 70% DMD was offered ad-lib. Cows were offered 3.2kg of concentrates at each milking. There was no effect of treatment with Bioplex minerals on milk production or composition, but there was a significant reduction in somatic cell counts by 40%. Bioplex Dairy was fed for 4 weeks before the first somatic cell count readings were taken and thus cows were not necessarily balanced for somatic cell counts at the time of first treatment. Cows treated with Bioplex Dairy had a non-significant reduction in days to emergence of the first dominant follicle (7.8 vs 9.3) and to first ovulation (20.4 vs 25.3). Conception rate to first service while satisfactory in both groups, was higher in the treated group (65 vs 58%). Results from this indicate that Bioplex minerals may have a beneficial role in dairy cow reproduction. Mineral status was determined in blood collected from cows at day 0, 35 and 70 of feeding and indicate normal mineral status in this group of cows which suggests that Bioplex minerals have a beneficial effect in cows which are normal for blood minerals concentrations.

Because cows were not balanced for SCC at the start of the experiment a second trial was set up in which 46 Friesian dairy cows were paired on the basis of parity and somatic cell count and milk yield in the current lactation. The cows were assigned at random within pairs to one of the following treatments: 1) Control; 2) 10g Bioplex minerals/De-Odorase (100mg Cu, 300mg Zn, 300mg Mn, 2mg Se, 1.5 g of De-Odorase) for 84 days. There was no significant difference between treatments in milk yield prior to the start of the trial. When original pairing of cows is included in a statistical model and average milk yields are adjusted for differences in yield on day 0 within pairs of cows on each block, mean milk yield is  $1.08 \pm 0.7$  kg/day greater in the cows

treated with the supplement than in the control cows ( $p < 0.06$ ). Total milk yield over the 84 days was higher in the cows treated with the mineral/De-Odorase supplement (Table 2). When the original pairing of the cows is considered in the analysis, the total milk yield from the treated cows was  $285 \pm 101$  kg greater in the 84 days of the trial than in the control cows ( $p < 0.005$ ). The difference in SCC between treated and control animals increased as the trial progressed such that in weeks 9-12 there was a 53% difference in mean and a 38% difference in median SCC (Table 3).

**Table 2. Effect of Bioplex minerals (Cu, Zn, Mn, Se) on milk production and quality following treatment for 12 weeks (2).**

	Control	Bioplex	P value
Number of cows	23	23	
Mean daily yield, kg	21.3+1.2	24.7+1.1	0.05
Yield over 84 days, kg	1786	2074	0.05
Total fat yield, kg	73.3 +4.2	79.3+3.4	0.27
Total protein yield, kg	57.3+2.8	64.5+2.7	0.07
Total lactose yield, kg	82.3+4.8	96.9+4.7	0.03
Median SCC/ml (x 1000)	86	56	

**Table 3. Somatic cell reduction in association with organic minerals.**

Mineral	Dose (mg)	SCC reduction	Reference
Zn	400	-40%	10
Cu Zn Se	100 300 2	-45%	2
Cu Zn Se	100 300 2	-35% wk 0-12 -52% wk 9-12	2

## ■ The 100-Day Contract

The '100-day contract' with the dairy cow begins 30 days before calving and continues for 70 days after calving (29). This includes the final growth of the calf in utero, the birth of a healthy calf, a healthy cow during the period of maximum productivity, controlled loss of body condition and optimizing fertility at first breeding. Markusfeld (19) described a number of metabolic diseases around the time of parturition as parturition disease complex, which are closely interrelated. A cow suffering from milk fever is at increased risk for retained



placenta, left displaced abomasums, and/or ketosis. The incidence of some of these parturient disease complexes was reported for a large herd in New York state (Table 4).

**Table 4. Lactational incidence of postpartum disease complex in dairy cows (7)**

Disorder	Risk (%)	Median day of occurrence
Retained placenta	7.4	1
Metritis	7.6	11
Ovarian cyst	9.1	97
Milk fever	1.6	1
Ketosis	4.6	8
Displaced abomasum	6.3	11
Mastitis	9.7	59

As the median day indicates most of these diseases are likely to occur during the period immediately following calving. However, these disorders have an impact on production and reproduction during the entire lactation. These disorders disrupt the cow's metabolic momentum towards high milk production and also have a negative effect on reproduction. Balancing energy and protein are critical in achieving maximum production and also reproduction. Micronutrients, including minerals and vitamins are important in achieving optimum production. Several minerals and fat-soluble vitamins have been associated with reproductive performance. Of the macro minerals, calcium:phosphorus ratios and total intake of the minerals are important in preventing milk fever at calving. Cows suffering from milk fever are more prone to retained fetal membranes, a prolapsed uterus and metritis. Therefore dry cow nutrition is important in prevention of these disorders and problems. Phosphorus intake should be kept at a ratio of about 1.5: 1 relative to calcium. Two micronutrients associated with enhancing reproductive performance are zinc and selenium. The specific role of zinc in reproduction is not well defined, but animals deficient in Zn have been shown to have lower concentrations of FSH and LH, particularly in males. Zinc supports tissue healing and may be important in the postpartum involution as well as in mammary gland health. Zinc is also a component of enzyme systems that may influence hormone synthesis. Zinc has been shown to decrease the incidence of lameness in dairy cows. Lameness has been associated in a significant increase in days to first service, days open and services per conception (29).

The role of Selenium in reproduction has been more thoroughly established. Supplementation of Se and Vitamin E have been shown to decrease the incidence of retained placenta, metritis and increased the rate of uterine involution. Vitamin E and Se reduce tissue damage and function and maintain

tissue integrity. This could enhance the uterine environment and support better fertility.

## ■ Conclusions

There are real fertility problems in the dairy herd and these have increased as milk production increases. Attempts to rectify the problems have been frustrated by the lack of response to management decisions and to lack of knowledge on the nutritional components that drive reproductive function. In addition it has not been possible to differentiate between factors affecting ovulation and factors contributing to fertility enhancement of the oocyte. We must continue to strive to produce more effective management packages that will support increased milk production without lowering fertility of the herd. Organic minerals have been shown to have beneficial effects under a wide range of applications in ruminants. These have included more efficient production, better quality milk production and improved reproductive efficiency. Further efforts are required to ensure that reproduction can be managed in a more controlled fashion.

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