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# TRACE MINERALS FOR THE PERFORMANCE HORSE: KNOWN BIOCHEMICAL ROLES AND ESTIMATES OF REQUIREMENTS

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In the absence of conclusive experimental data concerning the requirements for any nutrient, the nutritionist is faced with the dilemma of ignoring that nutrient or compiling information which enables an educated guess at the intake of a nutrient adequate to support a specific physiological function. The lack of conclusive experimentally derived data concerning the trace mineral requirements of all classes of horses is astonishing but the lack of data concerning the trace mineral requirements of the athletic horse is staggering. The literature concerning trace minerals and the horse is beginning to allow educated assessment of the requirements for growing horses and brood mares for selenium, zinc, copper, manganese, iron and more recently iodine and chromium. These minerals all play crucial roles in the physiological response to exercise, in energy metabolism and in tissue conservation during a period of insult brought about by exercise. Surely the performance horse has distinct needs for these as well as other nutrients in order to perform an athletic task effectively. The following is a discussion of the roles of certain trace minerals in exercise and where possible an estimation of the trace mineral needs of the athletic horse.

## Iron

It is probably fitting that a discussion of trace mineral requirements of the athletic horse begin with a discussion of iron. Iron is, at least from the standpoint of the layman, the first trace mineral that is considered in terms of supplementation. A recent survey conducted at a California race track indicated that a large majority of trainers had their horses on some type of iron supplement (Carlson, 1994). This concern with iron stems from the well known function of iron as part of the heme molecule. This is but one of the important functions of iron but is the basis of the interest in iron and the performance horse. The ancient Greeks recognized iron deficiency anemia and were said to have consumed water in which an iron sword had been placed in order to correct the symptoms of iron deficiency. As early as 1886 it was shown that pure crystals of horse hemoglobin contained 0.33 % iron. Cytochromes, catalases and peroxidases are other iron containing compounds and were discovered as early as the late 1800's. The cytochromes, like hemoglobin, are involved in oxygen utilization via electron transport.



Hemoglobin requires iron for its synthesis and is the component of the red blood cell which allows oxygen to be carried to the tissues. It appears that the cytochromes and enzymes associated with tissue metabolism as well as myoglobin take precedence in the supply of iron in the body and as such the first deficiency symptom associated with iron is anemia. The anemia associated with iron deficiency is hypochromic, microcytic anemia. There are few instances when practical diets would result in iron deficiency anemia. In the previously mentioned study of Carlson, horses which received supplemental iron had iron levels which fell into the normal range for iron in adult horses. Very few of the horses examined had any evidence of anemia and those with resting hematocrits below 34% (defined as anemia) showed no evidence of impaired iron status. This scenario is frequently the case and furthermore it is rare that a horse with lowered hematocrit responds to supplemental iron with a concomitant increase in hematocrit. Clinically significant anemia in the athletic horse is indeed a rare entity. One of the significant aspects of iron metabolism which buffers the horse and other animals from iron deficiency anemia is the ability of the body to conserve iron. Approximately 67% of the iron in the body is contained in the red blood cells in the form of hemoglobin. Red blood cells are formed in a process known as erythropoiesis and remain in the circulation for about 150 days. When these red cells "die," iron contained in the heme molecule is recycled and utilized to synthesize new heme molecules and red blood cells. As such, there is rarely net loss of iron from the body. It is interesting to note that exceptions to this are severely parasitized horses, horses with gastric ulceration that leads to blood loss and perhaps in horses which suffer from severe EIPH (exercise induced pulmonary hemorrhage). A recent study indicated that in excess of 80% of the horses on the race track had some degree of gastric ulceration or at least some erosion of the gastric mucosa. Whether this problem has resulted in net blood and therefore iron loss is still subject to some speculation. The fact that iron recycling is effective and iron deficiencies are rare is apparent from the lack of effectiveness of some of the iron tonics in increasing packed cell volume and hemoglobin concentrations in the blood of horses in training. More times than not, low hematocrits are an indicator of infection, low grade systemic disease or even perhaps marginal B-Vitamin status brought about by stressed large colon or cecal microbial populations and not a deficient dietary intake of iron.

Iron appears to be efficiently absorbed at low levels of intake. Iron absorption is decreased as iron intake increases and with the presence of high concentrations of copper, zinc, manganese, cadmium and cobalt. Additionally, due to the effective iron conservation mechanism, net iron excretion is low and that iron appearing in the feces appears to result primarily from unabsorbed dietary iron rather than from net endogenous fecal losses. Work conducted by Lawrence (1987) in Florida has failed to show an increase in hemoglobin, packed call volume or serum iron when ponies were supplemented with high levels of iron.

There appears to be some iron lost in the sweat. Meyer (1986) estimates that the concentration of iron in horse sweat is about 21 mg/l. Horses in intense exercise may lose as much as 25 - 30 liters of sweat per day and this sweat loss would represent a

net iron loss in the sweat of 500 mg/day (25 l X 21 mg/l). With these sweat losses and the possibility of greater synthesis of hemoglobin and myoglobin (muscle iron containing compound) in the athletic horse there may be reason to believe that the athletic horse has greater requirements for iron than does the mature sedentary horse. Practical dietary constituents contain from 40-50 ppm iron for the cereals, 100-150 ppm iron for the oilseed meals and from 200-1000 ppm iron for many of the forages. Care must be taken in sampling of pastures to avoid soil contamination. Soil in forage samples results in abnormally high iron values and may lead to errors in assessing the iron status of the diet. Another factor to consider in assessing the iron concentration in a manufactured feed is the amount and source of any dicalcium phosphate in the diet. Rations containing much dical, especially supplements or mixing pellets, may contain as much as 1000 ppm iron. It is probable that most of this is iron oxide which is poorly absorbed. Questions still remain, however, as to the impact of this iron on the efficiency of absorption of other minerals.

The National Research Council, Nutrient Requirements of Horses (NRC, 1989) lists the iron requirement of the performance horse as 40 mg/kg of dry matter (ppm). Assuming a dry matter intake of roughly 12 kg for the horse at high work intensities this would result in an iron requirement of roughly 500 mg/day. For horses in intense work that are losing a great deal of sweat this may only be enough iron to replace that iron lost in sweat and therefore may be a bit low in terms of meeting the horses' iron requirement. There is no reason to believe that the requirement is any higher than 40 ppm for the horse at light to moderate work intensity. Meyer (1986) suggest that the iron requirement of the 500 kg horse is 500, 600 and 1200 mg/day for light, moderate and heavy exercise. Unfortunately, it will be a long time before the people doing the feeding are convinced that some of the expensive hematinics don't really make their horses run faster and jump higher. Hopefully at the levels at which iron is fed it is fairly innocuous. It is just possible that in attempts to stimulate the "blood picture" some horsemen are unwittingly causing interactive deficiencies of other minerals.

## Manganese

The best known function of manganese is its role in bone formation. Manganese is needed in several of the steps necessary in the formation of chondroitin sulfate and in the synthesis of the organic matrix of bone which is predominantly a mucopolysaccharide ground substance. Manganese is required in the synthesis of chondroitin sulfate (ring any bells?, this is one of the new wave of supplements for the performance horse) and appears to be required in the enzymes galactotransferase and polymerase required in the synthesis of chondroitin sulfate from beta-D-glucuronide and 1,3-N-acetyl-D-galactosamine. Manganese deficiency is expressed as a disorganization of the cells making up the epiphyseal plate, a narrowing of the epiphyseal plate and a reduction of blood vessel migration into the growth plate. Additionally the cartilage of the growth plate in manganese deficient animals contains lower levels of

chondroitin sulfate than do the growth plates of normal animals. With respect to this function of manganese it stands to reason that two and three year old horses in which skeletal maturation is not complete may have a slightly higher requirement for manganese than do older animals.

Manganese is also known to be an activator of several enzymes such as arginase, thiaminase, carnosinase and deoxyribonuclease. Manganese is required for oxidative phosphorylation in the mitochondria and as such is very involved in regeneration of ATP from ADP, a reaction critical for the maintenance of adequate concentrations of the high energy phosphagens in the athletic horse. It is unlikely that a Mn deficiency would be rate limiting in the production of ATP but this demonstrates the crucial role that trace minerals play as co-factors and components of various of the enzymes. Manganese is also known to be involved in fatty acid synthesis and in amino acid metabolism either as a co-enzyme or as an activator of enzymes . Manganese also is an effective chelating agent allowing for more efficient and rapid transfer of amino acids. Because of the role that Mn plays in energy metabolism through various roles in lipid metabolism and carbohydrate metabolism it is of interest in the formulation of performance horse diets. In most areas of the world manganese deficiency does not appear to be of concern in horses grazing fresh forage. Mn concentrations in forage range from 50 - 300 ppm and these levels should meet the requirements of most horses. However, cereal grains contain less Mn and may range in Mn concentration from 5 ppm (corn) to 15 ppm (barley). In the athletic horse on high grain diets and consuming grass hay of variable quality it is probable that some supplemental manganese may be required. It is our practice to supplement performance horse diets with 80-100 ppm manganese with about 1/3 of the supplemental Mn coming from chelated manganese and 2/3 from manganous oxide. High concentrations of calcium and phosphorus in feeds or forages may inhibit manganese absorption and this should be taken into account in the formulation of feeds for all classes of horses. It is interesting to note that some of the forages I have sampled from around the world that are the highest in manganese are also the highest in calcium. Some forages in Ireland for instance may have from 200-300 ppm Mn and 1% calcium. Unlike in the US when these calcium concentrations are found in mixed grass/legume swards, these calcium concentrations are common in pure swards of ryegrass. Whether there is a significance to this is still under investigation.

There is little, <0.2 mg/l , manganese in horse sweat and as such one would expect manganese losses in sweat to be minimal. We currently would think that 350 mg/day of Mn should be adequate for horses at low work intensities and think that horses' requirement for this mineral should be met with 500 mg/day for horses at moderate to high work intensities.

## Selenium

Selenium is recognized as an essential nutrient as well as an environmental toxicant. The selenium requirements listed for domestic livestock may be as much a political compromise as an accurate assessment of the physiological needs for the mineral. Selenium is best known as an essential component of the selenium dependent enzyme glutathione peroxidase and functions as part of the cellular antioxidant defense system. Selenium functions much as does vitamin-E in a role of biological antioxidant and indeed there are a number of deficiency symptoms of selenium that may be partially corrected by vitamin-E and vice-versa. The antioxidant defense system allows for the trapping of free radicals and superoxides which cause oxidative damage to lipid membranes. Simply put, glutathione peroxidase converts reduced glutathione to oxidized glutathione and destroys peroxides by converting them to harmless alcohols. This conversion of the peroxides prevents them from reacting with lipid membranes and causing loss of membrane integrity.

It follows that since exercise results in increased oxygen delivery to the tissues and oxidation of energy substrate resulting in the generation of reactive oxygen by-products, peroxides, the selenium requirement for the athletic horse should be increased. Shellow (1985) did a trial with sedentary geldings fed a basal diet and three levels of supplemental selenium, 0.05, 0.1, and 0.2 ppm supplemental selenium to assess the impact of feeding supplemental selenium on plasma and whole blood selenium and on glutathione peroxidase which has been thought to be an accurate indicator of selenium status. They reported that there was a linear increase in plasma selenium over time until a level of 0.14 ppm was reached in the plasma and then selenium plateaued regardless of the amount of selenium that was fed. There was no difference in peak plasma selenium values between 0.1 and 0.2 ppm supplemental selenium. When the basal selenium level of the diet, 0.06 ppm was added to the supplemented selenium, this experiment suggested that a requirement of 0.16 ppm in the total diet was appropriate. Shellow (1985) was unable to find a relationship between dietary selenium level and glutathione peroxidase activity and suggested that plasma selenium may be a better indicator of selenium status than is glutathione peroxidase. It is interesting to note that in this experiment there was a significant difference in plasma and whole blood selenium due to the relatively high concentration of selenium associated with the red blood cells. Whole blood selenium is higher than is plasma selenium and it is important that the distinction be made in the interpretation of blood test results.

Stowe (1993 ) has suggested that the appropriate concentration of selenium in the total diets of horses is 0.3 ppm. This would mean that if a concentrate mix was 50% of the diet and the forage component of the diet was 0.06 ppm se, the grain mix would need to be roughly 0.6 ppm. There have been few studies dealing directly with the relationship of selenium requirements to exercise. Shelle (1983) investigated the effect of supplemental selenium on plasma selenium and on glutathione peroxidase in Arabian and crossbred horses subjected to a conditioning program. He reported that conditioning increased erythrocyte glutathione peroxidase activity and suggested that

horses at high work intensities may have higher requirements for selenium than the 0.1 ppm requirement suggested by the NRC. Supplementing horses with 1-2 mg of selenium and 1000 mg of vitamin E has long been thought to prevent exertional rhabdomyolysis in some horses. It is probably done in response to the "leakage " of muscle enzymes into the plasma from the muscle. Appearance of high levels of LDH, SGOT and CPK in the plasma has been indicative of muscle damage and the thought was that the muscle membrane was subjected to reactive peroxides resulting in damage to the membranes and leakage of the enzymes into the circulation. Unfortunately not all horses have responded to supplementation with selenium and vitamin E and ceased to tie-up. It is still a good idea to evaluate the diets of horses that tie-up and rectify selenium and vitamin E deficiencies should there be any. More recently, another interesting role of selenium has been revealed. Selenium appears to be part of the enzyme Type I iodothyronine deiodinase. This is a selenoenzyme which catalyzes the conversion of thyroxine (T4) to triiodothyronine (T3) which is the tissue active form of the thyroid hormone. It is interesting to speculate whether some of the clinical manifestations of hypothyroidism may in fact be secondary to selenium deficiency.

Our current recommendations for selenium in the performance horse range from 2.5 to 3.5 mg/day depending on work intensity, weight, and other environmental factors. We DO NOT recommend selenium and Vitamin-E injections as there have been numerous reports of anaphylactic reactions to the injection of some of the commercial preparations of these nutrients. In most cases the horses affected by the anaphylaxis died and since dietary selenium and vitamin E are well absorbed and result in increased plasma levels of these two nutrients it is not worth the risk to use the injection.

## Iodine

Much like selenium, iodine has suffered in relative obscurity since the margin between adequate and toxic has been over-stressed. The only known function of iodine is as part of the thyroid hormones thyroxin and triiodothyronine. The classical deficiency symptom for iodine is a hypoiodine goiter and the classic toxicity is a hyperiodine goiter. As such the only real way to assess the adequacy of the diet is to evaluate the ration. Thyroxin and the tissue active form of the hormone, triiodothyronine (T3), serve a multitude of metabolic and regulatory roles. The thyroid hormones affect all of the organ systems, muscle metabolism, the nervous system, respiration and the cardiovascular system. Thyroxin also controls growth rate and cell division, metabolic rate and oxidative metabolism.

In the athletic horse, perhaps the most important role of thyroxin is control of basal metabolic rate and cellular energy metabolism. Thyroid hormone stimulates respiration in the mitochondria resulting in increased oxygen consumption and energy production. There has been a great deal of interest in the use of supplemental, synthetic thyroxin in the performance horse. This use of Thyrol-L is predicated on what appears to be a fairly common hypothyroxemia among performance horses. Unfortunately,

no one has done the research to ascertain whether this apparent lack of thyroactive hormones is due to a lack of iodine in the diet, a lack of selenium for the conversion of T4 to T3 or to dysfunction of the thyroid gland. Additionally there are some known goiterigenic compounds such as kale, cabbage, Brassica, and perhaps Kentucky 31 tall fescue which interfere with iodine uptake and thyroid function.

Current NRC recommendations for iodine are for 0.1 ppm (mg/kg of dry matter). At this level of intake a dry matter intake of 10 kg per day would result in only 1 mg of iodine intake. This is a little less than half of our current recommendations for iodine. We would propose that the 500 kg horse at light work requires 1.75 mg of iodine, at moderate work 2.5 mg and in intense training 2.75-3.0 mg/day of iodine. There is still a great deal of work to be done in understanding the relationship between dietary iodine and thyroid function but I suspect it will be the next trace mineral topic that is in vogue.

## Chromium

The recognized function of chromium is as a component of glucose tolerance factor (GTF). GTF is thought to potentiate the action of insulin in Cr-deficient tissue. Insulin has anabolic characteristics as it promotes glucose uptake by the cell, stimulates amino acid synthesis and inhibits tissue lipase. Because of its role in carbohydrate, lipid and protein metabolism and in the clearance of blood glucose it is interesting to consider the requirements of the athletic horse for chromium. Chromium excretion is greater in athletic than in sedentary humans and the chromium requirement is increased by physical activity. Chromium supplementation has increased lean body mass in humans and pigs and has resulted in a partitioning effect on nutrients which favors tissue anabolism and muscle protein accretion. In calves chromium excretion is greater during stress and chromium supplementation has resulted in a stimulation of the immune system and less mortality and morbidity in shipped feedlot cattle. Certainly this aspect of chromium is interesting when we consider the performance horse and the kind of stresses that they may see. There has been a great deal of research on chromium in humans, rats and pigs owing to the crucial role insulin status plays in diabetes. There is very little research work that has been done in other species of animals and only a few reports in horses.

Pagan *et al.,* (1995) reported that supplementing performance horses with 5 mg/day of chromium in the form of chromium yeast had a beneficial effect on the response of horses to exercise stress. Horses were subjected to a standard exercise test on a high speed treadmill and blood and heart rate were monitored. Horses in that experiment receiving chromium cleared blood glucose following a meal more quickly than control horses, showed lower peak insulin values and lower cortisol levels. Chromium supplemented horses also had higher triglyceride values during exercise indicating perhaps more efficient fat mobilization. There was no difference in the heart rate in response to exercise between the two groups but peak lactic acid concentrations in

the chromium supplemented group of horses were significantly lower than for the controls.

Work on chromium continues in our laboratories at Kentucky Equine Research. There is gathering evidence from the field that chromium supplementation may have a positive impact on horses known to have chronic problems with exertional rhabdomyolysis. One of the features of horses that routinely tie-up is a problem with normal glucose and glycogen metabolism. It may be that the role of chromium in GTF assists in clearing glucose and may make glycogen utilization and storage more efficient. Trainers that have had horses on Metaboleeze (the KER chromium supplement) have commented that horses appear less nervous and more work tolerant than non-supplemented horses. Current work with chromium is progressing in the areas of the laminitic horse and in other conditions thought to be related to insulin insensitivity.

The good news is that everyone pretty well realizes that chromium is an essential nutrient for man and animals; the bad news is that chromium is not GRAS listed (generally recognized as safe) by the FDA. As such, it is currently not legal to add chromium yeast to commercial feeds as a source of organic chromium in the US. If a horseman wants to supplement with chromium he may do so in one of two ways: 1) he may buy chromium at a human health food store or 2) he may get a prescription from a veterinarian for Metaboleeze from KER. We have set the chromium requirement for the performance horse at 5 mg/day but know that a great deal more research needs to be done to titrate this requirement and elucidate further the beneficial effects of chromium in the diets of horses in training.

## Zinc and copper

There has been a great deal of attention given to the requirements of the young horse and the broodmare for copper and zinc. The role of copper in the copper dependent enzyme lysyl oxidase and that enzyme's role in the formation and maturation of cartilage has obviously stimulated interest in copper as it relates to developmental orthopedic disease in young horses. Additionally, zinc has long been known to play a role in the maintenance of epithelial integrity and in keratogenesis. The use of copper and zinc supplementation in modern horse feeds for all classes of horses stems from the possible role of these two nutrients in reducing physitis, osteochondrosis, wobbler syndrome and other manifestations of developmental orthopedic disease.

It is interesting to note that in addition to its role in cartilage and bone metabolism, copper is also involved in hemoglobin formation and in nerve conductivity and coordination. Copper is also involved in other enzymes such as tyrosinase and cytochrome oxidase to name but a few of the more well known of the enzymes containing a copper moiety or which need copper as a co-factor. There does not really seem to be a down side to the inclusion of copper at the levels we now use. Currently we would recommend the performance horse receive 131, 170 and 187

mg/day of copper for light, moderate and heavy work respectively. We also would typically use bioplexes (chelates) to provide 30% of the supplemental copper in most of our rations. These allowances are not a great deal more than those of the NRC for the performance horse. Current NRC recommendations are for 10 ppm copper or 10 mg/kg of dry matter intake. We would think that with calcium intakes being higher due to the use of legume hays and with typical feeds ranging from 0.6 - 1.0 % calcium, there is justification for higher copper inclusion rates. There is a small loss of copper in the sweat of horses and probably is no greater than 4 mg/l of sweat. Still this may result in sweat losses of 80-100 mg of copper per day.

Zinc is involved as a co-factor in a multitude of enzyme systems. Alcohol dehydrogenase, carbonic anhydrase and carboxypeptidase are but a few of the Zn requiring enzymes. In addition to the role as enzyme activator/co-factor, there are in excess of 200 zinc containing proteins. 50 to 60 % of the zinc in the body is stored in the muscle. Zinc deficiency has resulted in reduced insulin levels and reduced glucose tolerance along with increased insulin resistance especially in the peripheral tissues. There has been a noted decrease in glucose utilization in zinc deficient animals with a concomitant increase in fat catabolism. There is a lowering of resting R.Q. in zinc deficiency which would be characteristic of increased fat catabolism. In terms of bone metabolism zinc deficiency directly inhibits the effectiveness of somatomedin in stimulating cartilage growth and the classical symptom of zinc deficiency is disrupted keratogenesis.

There is a significant amount of zinc that is lost in the sweat. Meyer (1986) has determined the loss of zinc in the sweat to be 20-21 mg/l of sweat. Obviously in the hard working horse this zinc loss can be substantial. High levels of calcium and copper can reduce zinc absorption so the kind of hay being fed may have an impact on the amount of zinc required by the horse. Increased levels of protein or increased protein intake has been shown to reduce zinc absorption and increase zinc excretion. These classic and well documented physiological and biochemical functions of zinc along with the relatively high zinc losses in the sweat have resulted in the development of more liberal allowances for zinc than the NRC proposes. We would currently recommend the zinc intake of the performance horse in moderate and heavy work needs to be 500 mg/day and think that 400 mg/day is probably adequate for the horse at light work intensities. Regarding copper, it is our general practice to use bioplexes as 30% of the supplemental zinc in the ration. Another note of interest: when the efficacy of hoof supplements is considered, the inclusion of zinc-methionine along with biotin and 3 grams of methionine appears to result in greater growth of the stratum germinativum and the tubular horn of the hoof wall than does the feeding of biotin alone.

The trace mineral nutrition of the performance horse remains an educated guess. It is encouraging to see more emphasis on trace mineral nutrition in the horse and particularly in the athletic horse. Hopefully over the next several years we will be able to more thoroughly elucidate the trace mineral requirements of the horse and accurately

describe their metabolic functions. Until then hopefully the requirements that NRC and KER have derived will be of some use in preventing deficiency and enhancing the horses' ability to cope with the stress of exercise.

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