

Chapter 1. Review of Literature

Keeping in mind the objectives of these studies, this review begins with a discussion of the need for supplementation of pasture, including factors such as seasonal variation of pasture, types of supplements available and a discussion of carbohydrate fractions in pasture. Supplements currently available are commonly rich in hydrolyzable carbohydrate, therefore carbohydrate digestion in the horse and its resultant effects on the initiation of the feeding-fasting cycle and carbohydrate status is summarized. These studies have addressed the substitution of fat for hydrolyzable carbohydrate in horse diets, therefore fat nutrition in the horse is reviewed. Finally, considering secondary objectives, this paper addresses milk composition of the mare, growth and bone development (normal and abnormal) of the foal, and the likely influences of carbohydrate and fat supplementation.

Supplementation of pasture

Pastures provide less than optimal nutrition for reproduction and growth of horses. The quality of pasture changes with the season. Low fiber, but high sugar, starch, energy, protein and water content are apparent in the spring and early summer (Blaser et al., 1986). Fiber content increases and energy, protein, phosphorus, and β -carotene contents decrease as plants shed their seeds through late summer, fall and winter. The seasonal variation and resultant inadequacies in pasture affect grazing animals which have pasture as their primary habitat. Therefore, several studies have

been done to assess inadequacies in pasture and further develop recommendations for optimal nutrition of grazing animals. This paper will focus on current recommendations for nutrition of the horse, with some comparisons in other species.

Inadequacies in northern Virginia pasture

In a survey including 12 farms in northern Virginia (Kronfeld et al., 1996), pastures were found marginal or deficient in sodium, phosphorus, zinc, copper and selenium, compared with current recommendations (NRC, 1989). The NRC recommendations may be regarded as mean minimum requirements, so supplementation of pastures is even more necessary to reach allowances or goals for optimal nutrition. Research at the Virginia Tech MARE Center, as summarized below, indicated that supplementation of phosphorus, selenium, zinc and vitamin A enhanced nutrition of mares and growing horses in ways likely to improve health.

Phosphorus. Blood serum analysis indicated phosphorus depletion in weanlings but not in mares fed an all-forage diet of pasture and hay (Greiwe-Crandell et al., 1992). The pasture and hay samples were analyzed to be marginal or deficient in phosphorus. Supplementation with phosphorus in another group of weanlings fed pasture and a pelleted concentrate led to adequate serum phosphorus concentrations. Unsupplemented pregnant mares had hyperphosphatemia and hyperphosphaturia, which were regarded as paradoxical to the low phosphorus in the diet. The hyperphosphaturia was accompanied by calciuria, suggesting mobilization of calcium

and phosphorus from bone in pregnant mares that would ensure a sufficient supply for the fetus.

Selenium. Mares and weanlings fed only pasture and hay showed no clinical signs of selenium deficiency, but blood serum analysis reflected the low concentrations of selenium found in the pasture and hay analyses (Greiwe-Crandell et al., 1992). Selenium supplementation, via a pelleted concentrate, of another group maintained on pasture and hay, effectively maintained serum selenium concentration. Weanlings fed only pasture and hay had more clinically severe cases of an equine herpes virus type 4 infection than supplemented weanlings.

Weaning stress and zinc. The social dislocative stress of weaning may affect appetite, metabolism and immune competence. Serum phosphorus, copper and zinc concentrations declined during weaning in swine (Ullrey et al., 1967). Foals raised on pasture and supplemented with a concentrate coped better with weaning stress, as assessed by behavior and adrenocorticotrophic hormone response tests, than foals raised on pasture and hay only (Hoffman et al., 1995). The pastures and hays were marginal or deficient in phosphorus, zinc and copper. The supplement provided two times more phosphorus, seven times more zinc and five times more copper than the forages. The advantage of feeding the concentrate may have been due to its mineral content. Serum zinc was lower during weaning in foals that had access to only pasture and hay, compared with those that were supplemented.

Vitamin A. Clinical signs of vitamin A deficiency in livestock grazing forages during the winter or in periods of drought have long been documented. One of the

earliest reports of vitamin A deficiency in the equine during a period of drought may be in Jeremiah 14:6, "The wild asses stand on the hilltops and pant for breath like jackals; their eyesight fails them because they have no food." Seasonal differences in vitamin A status of grazing mares was demonstrated using serum retinol concentrations and a relative dose response test (Greiwe-Crandell et al., 1995a). Serum retinol concentrations and the relative dose response in newborn foals were linearly correlated with corresponding data from their dams (Greiwe-Crandell et al., 1996). Supplementation with vitamin A at two times the NRC (1989) recommendation marginally compensated for seasonal differences (Greiwe-Crandell et al., 1995b).

These studies indicate a need for supplementation of northern Virginia pasture with phosphorus, selenium, zinc, copper and vitamin A for the horse.

Types of supplements available

Current research has explored vehicles for supplementation of vitamins and minerals to the horse. Typical carriers include a small amount of concentrated pellets, a fortified treat, or a larger amount of supplement, such as a sweet feed (grain, molasses and soybean meal). Concentrated pellets in small amounts has been used successfully in practice where horses are fed individually, but when horses are fed in groups, the distribution may not be sufficiently even. Data from a farm surveyed by our laboratory indicated that eight weanlings plagued by developmental orthopedic disease were group fed about 450 g of pellets, rich in zinc and marginal in copper and phosphorus, in each of eight buckets (Kronfeld et al., 1996). It could be easily

assumed that some weanlings consumed much more or less than the ideal amount of pellets, and the resultant mineral imbalance may have been the source of their problems. Feeding a fortified treat to each horse by hand would ensure precise distribution, but at farms where a large number of horses are housed, this practice may be too labor intensive. A more common delivery system uses a relatively large amount of supplement, such as a sweet feed. This method is less precise than using a fortified treat, but it is also less labor intensive. Using a larger amount of feed which would be dilute in concentration, compared to potent pellets, may reduce the variation in consumption of supplementary micronutrients.

Seasonal variation in pasture

As cool season grasses grow from leafy to stemmy stages, dry matter yields increase with increases in fiber and lignin, and protein and nonstructural carbohydrate concentration decrease (Figure 1.1, Blaser et al., 1986). These differences correspond with season, with rapid new growth occurring in the spring or during periods after rain, and maturation throughout the summer. As the leafy portion is grazed or cut, regrowth provides additional leafy material until the growing season ends. Pastures analyzed in Virginia increase in nonstructural carbohydrate content, to 20 to 30% DM when rapidly growing, as compared to 5 to 10% DM when mature (D. Wolf, Virginia Tech, personal communication).

Variations in fiber and nonstructural carbohydrate content of pasture may elicit additional needs for optimal nutrition of the horse. Therefore, carbohydrate analysis of

pasture should be completed, and carbohydrate fractions should be considered when assessing intake and needs of the grazing horse.

Carbohydrate fractions in pasture

Accurate, precise and convenient analysis of carbohydrate fractions in plants is important for evaluating the nutrient composition and daily intakes of nutrients of grazing animals. Carbohydrates in feeds and forages include a non-structural fraction: hexoses, disaccharides, oligosaccharides and starches, and soluble dietary fiber, including gums, mucilages and pectins. The structural fraction includes substances found in plant cell walls, including hemicelluloses (complex polymers of arabinose, xylose, mannose and other compounds), cellulose, ligno-cellulose and lignin. Lignins are phenylpropane polymers and form the main noncarbohydrate fraction of plant cell walls. Though not technically classified as carbohydrates, some lignin is bound covalently to cellulose. A scheme of carbohydrates in horse feeds is proposed in Figure 1.2, which includes comparisons of fractions obtained by proximate analysis with fractions digested. The basis for the scheme will be discussed in this section.

Fiber methodology began in 1870, with the Weende method of proximate analysis, which separated carbohydrates into two groups: crude fiber (CF) and nitrogen-free extract (NFE). Crude fiber was determined as the residue remaining after extraction with solvent, dilute acid and alkali, and the subtraction of minerals as ash. Nitrogen was measured in the extract, and the remainder was called NFE:

$$\text{NFE} = 100 - (\text{water} + \text{protein} + \text{fat} + \text{ash} + \text{CF}).$$

The NFE was thought to represent sugars and starches.

Crude fiber was an inaccurate method of analysis and actually contained only 65 to 75% of the original cellulose and lignin from the plant (Hall, 1989). The estimation of NFE by difference included hemicelluloses, some cellulose and lignin, and it accumulated all of the laboratory errors from the other assays.

Proximate analysis of carbohydrates was improved by Van Soest (1963), who divided carbohydrates into three fractions: residues insoluble in neutral detergents, residues insoluble in acid detergent, and non-structural carbohydrate. These fractions provided more physiological groupings of carbohydrates according to their utilization by animals, especially ruminants.

Insoluble fiber (Figure 1.2) includes the plant cell wall components and may be measured as neutral detergent fiber (NDF), which includes hemicelluloses, celluloses, ligno-celluloses and lignins. The NDF method has been criticized for not recovering pectin, which has been regarded by some as part of the cell wall matrix (Van Soest et al., 1991). Acid detergent fiber (ADF) includes celluloses, ligno-celluloses and lignins. Hemicellulose may be estimated as $NDF - ADF$. This estimation of hemicellulose may be too low when pectin is precipitated into the ADF or when biogenic silica, which is soluble in ND reagents and insoluble in AD reagents, is present (Van Soest et al., 1991). Non-structural carbohydrate (NSC) is estimated by difference (NSC “by difference”, Figure 1.2):

$$NSC = 100 - (\text{water} + \text{protein} + \text{fat} + \text{ash} + \text{NDF}).$$

The disadvantage of this estimation, besides the accumulation of laboratory errors from the other assays, is that NSC “by difference” includes soluble fibers, resistant starches and oligosaccharides which are not hydrolyzed by mammalian enzymes. Non-structural carbohydrate may be overestimated in feeds such as citrus, beet pulp and legume forages which contain high concentrations of pectin, which is not recovered in NDF. Non-structural carbohydrates have not been further classified into their various components by ruminant nutritionists because all are readily digested by fermentation in the rumen. The specific nutritive characteristics of NSC fractions for ruminants do not apply to hind gut fermenters, which digest by hydrolysis before fermentation.

Total dietary fiber (TDF) and non-starch polysaccharides (NSP) have been of interest in human nutrition and for animals with hind gut fermentation. Total dietary fiber (Figure 1.2) includes all polysaccharides (cellulose, hemicellulose, pectins, gums and mucilages) resistant to digestion by mammalian enzymes. Most methods include lignin as part of TDF (Van Soest et al., 1991), and some methods include starches resistant to enzyme digestion (Hall, 1989). Another estimation of NSC by difference uses TDF in the equation:

$$\text{NSC} = 100 - (\text{water} + \text{protein} + \text{fat} + \text{ash} + \text{TDF}).$$

This equation may provide a better estimate of hydrolyzable carbohydrate than that estimated with NDF, but this calculation of NSC may include non-hydrolyzable oligosaccharides and resistant starches.

Non-starch polysaccharide (NSP, Figure 1.2) analysis is used in human nutrition and is similar to TDF, including cellulose, hemicellulose, pectins, gums and mucilages,

but not lignin (Englyst et al., 1982). The fraction of NSP may be further divided into soluble and insoluble fractions. Soluble NSP may be estimated as NDF – TDF, when TDF analysis includes lignin (Van Soest et al., 1991), or as NSC minus starch and sugar, though this estimation may be biased by resistant starch (Englyst et al., 1982). For the horse, NSP may not be as useful a measure as TDF, since much larger proportions of lignin are naturally consumed in the equine diet, compared to that of humans. Proponents of NSP tend to neglect the importance of lignin and its binding to cellulose in determining rate of fermentation.

Non-structural carbohydrate may be measured directly (Davis, 1976; Smith, 1981). Carbohydrate fractions may be extracted using hot water (yielding hexoses), then dilute acid (disaccharides) and enzymatic digestion (polysaccharides). The total non-structural fraction is analyzed using colorimetric analysis for glucose using *para*-hydroxybenzoic acid. Direct analysis of NSC, referred to as total nonstructural carbohydrate (TNC, Figure 1.2), yields *all of the hydrolyzable carbohydrates*, namely the hexoses, disaccharides, some oligosaccharides, fructosans, rapidly and slowly hydrolyzable starches. When large amounts of starch and sugar are consumed by horses, a portion may escape hydrolysis in the small intestine and be fermented in the large bowel. Rapid fermentation may increase lactic acid production and increase the risk of digestive disorders (Sprouse et al., 1987; Clarke et al., 1990). Soluble non-starch polysaccharides, such as pectins, arabans and β -glucans, are more slowly fermented than sugars and produce less lactate (Van Soest et al., 1991).

Distinguishing soluble NSP in horse feeds from NSC may help establish “good” fermentation without problems associated with an excess of starch and sugar.

Digestion of carbohydrate in the horse

In most animals, carbohydrate digestion begins with the contact of dietary carbohydrate with salivary amylase. The horse does not produce appreciable amounts of salivary amylase, so carbohydrate digestion begins to some extent in the stomach, where the acidic conditions favor hydrolysis independently of any enzymes (Kronfeld and Van Soest, 1976). The primary site of hydrolytic digestion occurs in the small intestinal lumen with the attack of pancreatic amylase. Amylase hydrolyzes amylose and amylopectin, breaking $\alpha(1\rightarrow4)$ glycosidic bonds [but not $\alpha(1\rightarrow6)$ bonds], yielding, maltose, maltotriose and α -limited dextrans. Amylase may hydrolyze some of the dextrans, but the intestinal epithelium secretes specific dextranses to further hydrolyze the dextrans to glucose. The intestinal epithelium also secretes specific disaccharidases, such as maltase, sucrase and lactase, to hydrolyze dietary disaccharides. Oligosaccharidases hydrolyze some dietary oligosaccharides and the oligosaccharide intermediates produced from the initial hydrolysis of starch (Gray, 1992). The exception is that omnivores and hind gut fermenting herbivores do not secrete oligosaccharidases specific for breaking galacto- and fructo-oligosaccharide bonding. These oligosaccharides, and perhaps others, must be fermented.

Some fermentation occurs in the distal small intestine of the horse (Zentek et al., 1992; Moore-Colyer et al., 1997). It is not well known whether small intestinal fermentation occurs independent of large bowel fermentation, or if fermentation in the small intestine is due to some reflux of large bowel contents. Most fermentation occurs in the large bowel. Resistant starch, some oligosaccharides and soluble dietary fiber are fermented by microorganisms and are further hydrolyzed, yielding glucose, then pyruvate and lactate as intermediate products. Hemicellulose and cellulose are attacked by enzymes secreted by microbes and degraded to oligosaccharides, which are then degraded to glucose and glucose phosphates (Kronfeld and Van Soest, 1976). Pyruvate and lactate are also present in small amounts, but major products include the short chain fatty acids, acetate, propionate and butyrate. Ligno-cellulose may be degraded to cellulose by fungi present in the large bowel, and cellulose is then fermented. Lignin remains undigested and is excreted in feces.

Absorption of sugars occurs in the small intestinal mucosa, either by facilitated diffusion or by Na^+/K^+ active transport. Glucose and galactose may be absorbed by either facilitated diffusion or active transport, but fructose requires active transport.

Blood glucose concentrations are regulated by metabolic and hormonal changes. Glucose, galactose and fructose undergo a series of glycolytic reactions to form glucose phosphates, which are then catabolized through the tricarboxylic acid cycle to form ATP, CO_2 and water, or these sugars are converted to and stored as liver or muscle glycogen, or returned to circulation as free glucose (Kaneko, 1989).

Hormones of the pancreas, anterior pituitary, adrenal cortex and medulla, including insulin, somatostatin, glucagon, adrenocorticotrophic hormone, cortisol and catecholamines, are associated with the metabolism of carbohydrate and the regulation of blood glucose.

Feeding–fasting cycle

Metabolic and hormonal changes associated with the feeding-fasting cycle are noted primarily in meal feeding animals (Figure 1.3). During the fed state, glucose, insulin and somatostatin increase, while free fatty acids, growth hormone and IGF-I, glucagon, and perhaps thyroid hormone, decrease. During fasting, free fatty acids, growth hormone, IGF-I, glucagon and thyroid hormone increase, while glucose, insulin and somatostatin decrease.

Insulin is synthesized as preproinsulin on the rough endoplasmic reticulum in the pancreatic β cells. Preproinsulin is cleaved to proinsulin by microsomal enzymes, and the proinsulin is later cleaved to insulin and C peptide. Insulin release is stimulated by hyperglycemia, amino acids leucine and arginine, vagal stimulation, sulfonylureas, and enteric hormones, including glucagon-like peptide, gastrin inhibitory peptide (GIP), cholecystokinin (CCK), secretin and gastrin (Dupre et al., 1973; Kaneko, 1989). Insulin release is inhibited by hypoglycemia, somatostatin, and the α -adrenergic effect of catecholamines. Insulin acts to promote nutrient storage by enhancing the transport of glucose into muscle and adipose, promoting muscle protein synthesis and the storage

of triglycerides in adipose, stimulating hepatic and muscle glycogen synthesis and storage, and by inhibiting hepatic glycogenolysis, ketogenesis and gluconeogenesis.

Somatostatin is secreted by pancreatic D cells. Somatostatin release is stimulated by many of the same factors which stimulate insulin release, including glucose, arginine and gastrointestinal tract hormones. Somatostatin inhibits the action of growth hormone and restrains the movement of nutrients from the gastrointestinal tract into circulation, prolonging gastric emptying and decreasing gastric acid and gastrin production.

Growth hormone is synthesized by the somatotropic cells of the anterior pituitary. The secretion of growth hormone is regulated by growth hormone releasing hormone and somatostatin. Thyroid hormones, sex steroids and glucocorticoids also influence growth hormone secretion (Mol and Rijnberk, 1989). Growth hormone works by direct or indirect action, mediated by insulin-like growth factor I (IGF-I). Direct action of growth hormone tends to decrease protein catabolism by enhancing lipolysis, and it decreases carbohydrate utilization and restricts glucose transport into cells. Growth hormone, through IGF-I, enhances protein synthesis by increasing amino acid uptake and increases transcription and translation of mRNA.

The liver, osteoblasts and bone marrow stromal cells produce IGF-I, IGF-II and IGF binding proteins. The IGFs act in a paracrine manner on the osteoblasts to stimulate further production of IGFs and associated binding proteins. Insulin-like growth factors act to promote mitogenesis of osteoblast precursors and collagen synthesis (Rosen et al., 1994). Growth hormone, thyroid hormone, parathyroid

hormone, estradiol, vitamin D and other factors may regulate osteoblast synthesis and release of IGFs and IGF binding proteins.

Glucagon is secreted by α cells of the pancreatic islet. Glucagon release is stimulated by amino acids (except leucine), catecholamines, GIP, CCK, gastrin and glucocorticoids. Glucagon acts to provide energy to tissues during the fasted state, by stimulating glycogenolysis, gluconeogenesis and ketogenesis.

In the continuously grazing and browsing horse, the changes associated with the feeding-fasting cycle may not be largely evident. To provide additional energy in order to meet performance demands, humans have introduced starchy cereal grains into the horse diet, commonly supplied as two meals per day. These meals may have increased metabolic and hormonal changes associated with the feeding-fasting cycle, which may not be well tolerated by the horse.

In addition to stimulating the feeding-fasting cycle, supplementation of rapidly growing pasture with starchy cereal grains may provide hydrolyzable carbohydrates in excess of the hydrolytic capacity of the small intestine. Hydrolysis of starch yields glucose, which when present in large amounts may establish the feeding-fasting cycle of metabolic and hormonal changes. When hydrolyzable carbohydrates are consumed in excess, a portion may escape digestion in the small intestine and be rapidly fermented in the large bowel. Rapidly fermentable carbohydrate has been implicated as an etiologic factor in colic, laminitis and developmental orthopedic disease (Sprouse et al., 1987; Clarke et al., 1990; Williams and Pugh, 1993). Supplementation with

dietary fiber as an energy source may buffer seasonal changes in pasture, thus decreasing the risks of disorders associated with hydrolyzable carbohydrate.

Assessing the shifts in glucose and hormone concentrations associated with the feeding-fasting cycle may not be unequivocal, due to circadian variation and homeostatic regulation. Function tests for carbohydrate status may provide a reasonable assessment of glucose regulation and hormonal balance in the horse.

Carbohydrate status in the horse

The assessment of carbohydrate status in the horse is important in order to determine responses to diet or disease.

Glucose tolerance

Carbohydrate status is commonly assessed by means of a glucose tolerance test, the temporal response of blood glucose concentration to an oral or intravenous dose of glucose (Roberts and Hill, 1973; Jacobs and Bolton, 1982). For an oral glucose tolerance test, the initial change in glucose reflects absorption and clearance; blood glucose concentration increases when the rate of glucose entry into the blood exceeds the rate of glucose removal from the blood.

Blood glucose concentrations are regulated by insulin, cortisol and other hormones (Kaneko, 1989). As blood glucose rises, insulin release is stimulated. Insulin facilitates the transfer of glucose into peripheral tissues such as muscle or adipose.

In the horse, peak blood glucose concentrations occur approximately 60 min after the oral glucose dose (Roberts and Hill, 1973; Jacobs and Bolton, 1982; Jeffcott et al., 1986). The subsequent fall in blood glucose is a reflection of clearance rate exceeding that of absorption. Eventually, blood glucose falls below basal concentrations in a hypoglycemic phase characterized by inertia of glucose regulatory mechanisms. In a hypoglycemic state, cortisol, released by the adrenal cortex, stimulates hepatic gluconeogenesis, and blood glucose concentrations return to normal. The entire glucose tolerance test, including the initial rise, fall, hypoglycemic phase and final return to basal concentrations, is complete in 3 to 5 h in the horse, depending on dose and carbohydrate status of the animal.

The oral glucose tolerance test in the horse has been typically used to aid in diagnosis of pancreatic and small intestinal dysfunction (Roberts and Hill, 1973). More recently, as increased risk of laminitis has been associated with hyperglycemia and hyperinsulinemia (Jeffcott et al., 1986; Freestone et al., 1991), glucose tolerance tests have been used as a functional index of the metabolic response of horses to certain feeds (Jacobs and Bolton, 1982; Stull and Rodiek, 1988; Ralston, 1992). A diet with high concentrations of sugar and starch favors a minimum rise in the tolerance curve, while a low or carbohydrate-free diet may elicit a high tolerance curve, similar to that of a diabetic.

The response to an oral glucose challenge was examined in horses grazing fresh pasture, compared to horses in stalls consuming a diet comprised of oat hay, a prepared feed, oat and alfalfa chaff (Jacobs and Bolton, 1982). The pasture fed

horses, compared to the stable fed horses, had higher blood glucose concentrations in response oral glucose. Although dry matter, protein, NDF and ADF were examined as possible contributing factors to the difference in glucose response, NSC concentrations in the diets were not considered. The dietary data presented by Jacobs and Bolton (1982) do not provide sufficient information for the calculation of NSC, however, an estimation by difference from the available data ($\text{NSC} = \text{dry matter} - [\text{protein} + \text{NDF} + 8\% \text{ (assumed for fat and ash)}]$) indicates that the diet fed to the horses in stalls may have had as much as four times more NSC than the pasture. Adaptation of stabled horses to utilize glucose at a faster rate would lower the plasma glucose response to an oral load by increasing the rate of clearance.

Basal plasma glucose concentrations in horses were not influenced by pregnancy or lactation, but the plasma glucose response to an i.v. injection was affected by lactation (Evans, 1971). Marked changes in carbohydrate metabolism, including hyperinsulinemia, enhanced β cell sensitivity, increased degradation of insulin, and insulin resistance were demonstrated in the pregnant mare (Fowden et al., 1984). These changes resulted in exaggerated responses to feeding and fasting in pregnant, as compared to non-pregnant mares.

Glycemic indices

The glycemic index of a feed is an *in vivo* estimate of the digestible carbohydrates in the feed. Although similar in procedure, the glycemic index provides an assessment of the carbohydrate in a feed, while glucose tolerance is a function of the carbohydrate status of the animal. Glycemic index values are normalized to a reference value of

available carbohydrate in a food (e.g. in human nutrition, white bread is the reference food) based on rate of digestion and absorption (Englyst et al., 1996). A feed with high concentrations of sugar and starch would have a high glycemic index, but the glycemic index of a high fiber, low or carbohydrate-free feed would be relatively low.

Blood glucose responses and corresponding insulin concentrations were influenced by differing energy source in diets formulated to be isoenergetic (Stull and Rodiek, 1988). Glucose and insulin response to a meal were largest in horses fed an alfalfa-corn or an all corn diet, lowest in horses fed an all alfalfa diet, and intermediate in horses fed a corn grain-corn oil diet (Stull and Rodiek, 1988). Compared to horses fed alfalfa, horses fed corn tended to have higher glucose ($P < .12$) and insulin ($P < .15$) responses to feeding (Rodiek et al., 1991).

Fat nutrition

The use of diets containing added fat has sparked much interest in the horse industry. Current research has focused on effects of supplemental fat on energy balance, digestibility, behavior, exercise performance, growth and reproduction (Potter et al., 1992).

Digestion of fat in the horse

Depending on dietary intake, fat may be present as triglycerides, fatty acids, phospholipids or cholesterol. The digestion of fat is initiated by the presence of lingual lipase from the mouth and gastric lipase from the stomach. Triglycerides are hydrolyzed to some extent into mono- or diglycerides and free fatty acids by gastric

lipase, although most triglycerides are digested in the small intestine. The primary site of fat digestion in the horse occurs in the small intestinal lumen in the presence of bile acids and pancreatic lipase. Bile acids emulsify the fat to make it soluble. Pancreatic lipase targets α -ester bonds and hydrolyzes triglycerides to form mono- and diglycerides and free fatty acids. The detergent properties bile salts promote the formation of micelles, which are molecular aggregates of monoglycerides with the nonpolar portion directed inward and polar groups projecting outward. Bile salts, cholesterol and fat soluble vitamins are carried in the lipid soluble interior of the micelle.

Some short- and medium-chain fatty acids are directly absorbed into portal blood, but most lipids are absorbed as micelles. Micelles move into the intestinal villi and are absorbed into the intestinal mucosal cell. Within the mucosal cell, monoglycerides and free fatty acids are recombined to form triglycerides or phospholipids. Cholesterol is reesterified. The triglycerides, phospholipids, cholesterol esters and some cholesterol, free fatty acids and fat soluble vitamins combine with protein to form chylomicrons. Chylomicrons are absorbed into the lymphatic system and then into general circulation.

Substitution of energy sources: fat versus hydrolyzable carbohydrate

Some advantages of feeding fat-supplemented diets to horses are associated with the substitution of fat for hydrolyzable carbohydrates. Fat is much more energy dense than carbohydrate (9 kcal/g of fat, 4 kcal/g carbohydrate). Comparing the energy yield between the two substrates, glucose nets 35.5 moles of ATP formed from

ADP and inorganic phosphate during glycolysis and the tricarboxylic acid cycle (Blaxter, 1989). Beta-oxidation of fatty acids yields one Acetyl-CoA and 40 ATP from FADH₂ and NADH per cycle. Twelve ATP are formed from each Acetyl CoA in the tricarboxylic acid cycle, and 2 ATP are required overall, to transport the fatty acid into the mitochondria for β -oxidation. In this manner, 129, 142, 144 and 146 moles of ATP are formed from palmitic, linoleic, oleic and stearic acids, respectively. Net ATP yield from the oxidation of other fatty acids may be calculated using the same means (Blaxter, 1989).

In addition to increased yield of ATP, the direct oxidation of fat, as compared to the direct oxidation of glucose, generates approximately 3% less heat. The generation of 3% less heat may be a small but significant advantage to the performance horse working under heat-stressed conditions (Kronfeld, 1996). This difference in heat production is about 50% when comparing the indirect oxidation of fatty acids via triglycerides to the indirect oxidation of glucose via lactate, glycogen or triglycerides.

Palatability

In three separate palatability studies conducted by Holland et al. (1997), horses largely preferred corn oil over other vegetable and animal fat sources (soybean oil, soy lecithin, hydrolyzed tallow flakes, cottonseed oil, peanut oil).

Behavior

The supplementation of fat in horse diets interests riders and drivers, due to the suggestion that fat reduces “hot” or excitable behavior associated with the feeding of hydrolyzable carbohydrate. Fat supplementation is generally believed to promote

tractability. Horses fed a fat-supplemented diet had less spontaneous activity in the field and less reactive behavior to a novel stimulus, such as the opening of an umbrella or the shaking of a can of coins (Holland et al., 1996). These results provided the first quantitative evidence that dietary fat may reduce activity and reactivity of horses, although these results were consistent with evidence previously reported on the basis of incidental observations (Pagan et al., 1987).

Digestibility

Added fat (corn oil, soy lecithin, corn oil-soy lecithin blend) was more digestible (80 to 90%) than endogenous fat (approximately 50% digestible) in a total mixed ration comprised of hay, grain, soybean meal and molasses (Holland, 1994). Digestibility was lower in animal fat than in vegetable fat (Rich, 1980). Supplementation with soybean oil (170 g oil/kg total feed DM) increased the absorption of phosphorus and the digestibility of ether extract and gross energy (Hollands and Cuddeford, 1992).

Diets supplemented with 7.5% or more fat depressed calcium absorption in poultry (Feede et al., 1960) and sheep (Tillman et al., 1958; White et al., 1958). Calcium availability may be reduced in fat-supplemented diets due to the formation of insoluble calcium soaps (Palmquist et al., 1986). Horses fed fat-supplemented diets containing 10% corn oil or animal fat had a tendency for decreased calcium absorption — 18% calcium absorbed in diets supplemented with animal fat and 24% with corn oil supplementation, versus 28% in controls (Rich, 1980).

Excercise Performance

The strategy of feeding fat for exercise performance is to promote fat adaptation and enhance oxidation of fatty acids. Feeding a fat-supplemented diet may cause a shift from carbohydrate oxidation to fat oxidation during exercise. The result of this shift is the sparing of muscle glycogen (Greiwe et al., 1989). Glucose-6-phosphate accumulates when phosphofructokinase is inhibited by citrate (Randle, 1986). Citrate is produced by fatty acid oxidation, and the accumulation of glucose-6-phosphate is associated with the suppression of glucose and glycogen utilization by the inhibition of hexokinase and phosphorylase.

Fat-supplemented diets have been reported to prevent tying-up and improve the performance of racing sled dogs and horses (Kronfeld et al., 1994). Arabian horses observed in a treadmill laboratory had increased lactate thresholds and higher peak blood lactate concentrations during anaerobic exercise tests when adapted to a fat-supplemented diet (Taylor et al., 1995). The accumulation in blood lactate in response to fat adaptation may be due to reduced activity of pyruvate dehydrogenase (Randle, 1986). The products of fat oxidation, acetyl-CoA and NADH, may directly inhibit pyruvate dehydrogenase, or these products may result in higher ratios of acetyl-CoA/CoA and NADH/NAD⁺, activating pyruvate kinase and converting pyruvate dehydrogenase to its inactive form. Accumulation of lactic acid in the muscle has been generally associated with metabolic fatigue.

Previous work in a treadmill laboratory indicated an increase in fatigue, associated with decreased efficiency of calcium uptake and release by the sarcoplasmic reticulum in horses fed a fat-supplemented diet (Wilson et al., 1994).

This effect may have been due to the type of fat fed, because lecithins included in the ration may have upset the balance of phosphatidylcholine and phosphatidylethanolamine in the cell membrane, thus affecting calcium exchange.

Milk composition

Milk composition of protein, fat and gross energy in light horse breeds from parturition to 4 mo of lactation was described by Ullrey et al. (1966). Compared with bovine milk, equine milk has less fat, more lactose and relatively similar protein concentrations. Current research has examined composition of milk fatty acids, amino acids and vitamin content (Csapó et al., 1995; Csapó-Kiss et al., 1995), effects of lactational stage, age and parity (Doreau et al., 1990; Doreau et al., 1991; Asai et al., 1995), and nutritional influences on milk production and composition (Davison et al., 1991; Doreau et al., 1992).

Milk yield is largely affected by parity and lactational stage. Milk yield was higher for multiparous than primiparous mares (Doreau et al., 1991) and decreased over lactation, with the largest yield occurring around 30 d after foaling (Gibbs et al., 1982). Milk fat, protein, gross energy and calcium decreased and lactose increased over lactation (Ullrey et al., 1966; Doreau et al., 1990). Increasing energy intake in lactating mares decreased milk protein, fat, gross energy and total solids (Pagan and Hintz, 1986), while body condition scores of the mares increased to greater than ideal.

Bovine milk contains primarily saturated fatty acids due to extensive hydrogenation of dietary fatty acids by ruminal microbes prior to absorption (Sutton and

Morant, 1989). The extent of hydrogenation is influenced by the roughage content of the ration. Low roughage diets tended to reduce the proportions of saturated fatty acids and increased proportions of oleic, linoleic and linolenic acids in cows' milk (Sutton and Morant, 1989). Dietary NDF was negatively related to milk production and protein and positively related to fat content (Beauchemin, 1991). Increasing ADF in the hay portion of cattle diets decreased milk yields but did not change milk composition (Alhadhrami and Huber, 1992).

In contrast, little or no hydrogenation of unsaturated fatty acids is expected before absorption in hind gut fermenting animals, therefore milk long chain fatty acid composition should be directly related to fatty acid content of dietary fats in these species. Compared with bovine milk fat, equine milk fat had 2.1 times more dodecanoic acid, 3.1 times more decanoic acid, 4.9 times more linoleic acid, 9.6 times more octanoic acid, and 224 times more linolenic acid (Csapó et al., 1995). Conversely, equine milk fat had .62 times as much myristic acid, .53 times as much palmitic acid, and .2 times as much stearic acid as bovine milk fat (Csapó et al., 1995). Milk of mares fed a diet rich in forage (95% hay, 5% concentrates), compared to milk from mares fed a diet rich in concentrate (50% forage, 50% concentrate), had higher milk fat and protein concentrations, and higher linolenic acid and lower linoleic acid concentrations (Doreau et al., 1992). Thus, the composition of mares' milk may be an assessment of the nutritional status of the mare, and it affects the foal.

Growth of the foal

One of the most extensive collections of growth rate data include weight, height and cannon bone measurements of 1,992 Thoroughbred foals, measured from 1958 to 1976 (Hintz et al., 1979). Commonly known as the Windfields data, after the farm from which it was collected (Windfields Farms, Oshawa, Ontario, Canada), the results have provided a basis for subsequent work. The Windfields data indicated that colts were heavier, taller and had bigger bones than fillies. Foals born in January, February or March were lighter, shorter and had smaller cannon bones than foals born in April, May or June (Hintz et al., 1979). Growth curves from the Windfields data indicated that foals grew most rapidly during the first months of life.

Finnhorse foals born in May tended to be larger than foals born in any other month, with largest differences apparent at ages 6, 12 and 24 mo (Saastamoinen, 1990). Kentucky Thoroughbred foals born in January, February or March maintained a size advantage in their first year of age over foals born in April, May or June (Thompson, 1995). The differences in climate between Kentucky and Canada (Windfields data, Hintz et al., 1979) or Finland (Saastamoinen, 1990) may have influenced growth rates.

Both the Ontario foals and the Kentucky foals exhibited a slump in growth, around 360 to 400 d of age (Figure 1.4). The slump in growth rate may be due to a metabolic change, or a result of seasonal differences in nutrient composition of pasture. Due to the arrangement of the data, it is difficult to determine whether this slump is an effect of age or of season. However, most of the foals would be aged around 360 to 400 d in early spring, which corresponds to the season of rapid pasture

growth. The slump in growth could be easily attributed to the high sugar, starch and water content and overall low nutritional value of spring pasture. Proper supplementation at this time may buffer the changes in pasture and smooth the growth curve.

Creep feeding foals from 10 d to 130 d of age (birth to weaning) increased the rate of skeletal growth, as measured by body weight and wither height (Thompson et al., 1998a). Increases in body weight and wither height were greater for weanlings fed rations containing DE at 150% of requirements (Thompson et al., 1988b). Weanling horses were fed a high concentrate (62%) pelleted diet or a high forage (70% alfalfa) pelleted diet in limited amounts or with ad libitum access to feed (Cymbaluk, 1989). Weanlings with ad libitum access to feed (Cymbaluk, 1989) consumed 35 to 46% more DE than needed for recommended growth and had weight gains 31% higher than reported for normal growth (Hintz et al., 1979). Yearlings had lower weight and heart girth gains when fed restricted levels of concentrate which provided DE and protein that approximated NRC (1978) recommendations, compared to yearlings provided ad libitum access to the same concentrate during two 1.5 h daily feeding periods (Ott and Asquith, 1986). Wither height, body length and hip height did not differ in response to diet. This study indicated that maximum growth rates were not achieved when yearlings were restricted to DE intake at NRC (1978) recommendations (Ott and Asquith, 1986). Foals supplemented with canola oil (10.5% fat in the concentrate), compared to control fed (5% fat) foals, had increased body weight and a tendency for increased heart girth at 7 to 12 mo of age (Saastamoinen et al., 1994).

Growth rates of foals may be closely associated with bone growth and maturation. The selection of horses predisposed to rapid growth has been a goal of many horse owners, in hopes of producing larger and faster racing and sport horses. Genetic selection of horses for rapid growth may be counter-productive, because normal bone growth may be compromised due to metabolic or endocrine imbalances. Developmental disorders of bone have become more prevalent and are linked to chronic lameness and fractures in horses (Krook and Maylin, 1988).

Normal bone growth

Bone templates in the fetus are composed entirely of cartilage. Mature bones develop from cartilage by the process of endochondral ossification. In the fetus, centers of ossification develop at the ends (epiphysis) and centers (diaphysis) of long bones, converting cartilage to bone (Stockwell, 1979; McIlwraith, 1996). Between the epiphysis and diaphysis is the metaphyseal growth cartilage complex, also called the metaphyseal growth plate. After birth, endochondral ossification occurs in articular-epiphyseal and in metaphyseal growth cartilage complexes. The growth cartilage complexes are responsible for the rate of growth and actual bone length (Orth and Cook, 1994).

As the chondrocytes of the growth complexes mature, proliferate and hypertrophy, they produce the extracellular matrix, composed of water, proteoglycans and collagen (Henson et al., 1996). Calcification of the extracellular matrix occurs in the hypertrophic region, in chondrocytes nearest the areas of vascular penetration

(Figure 1.5, Orth and Cook, 1994). Chondrocytes store calcium in the mitochondria in the beginning of the hypertrophic zone and release the calcium in the bottom half of the zone. Extracellular matrix vesicles, rich in mineral, alkaline phosphatase and calcium binding proteins, are found near calcification sites (Wuthier, 1982). The production of alkaline phosphatase increases. This enzyme releases phosphorus into the extracellular matrix. Proteoglycans and collagen are removed, and chondrocytes undergo apoptosis as vessels penetrate the cartilage, following phagocytic endothelial cells (Leach and Lilburn, 1992; Orth and Cook, 1994). Osteoblasts then deposit bone in the calcified cartilage.

The process of bone formation and resorption is influenced by a number of hormones and other factors, including growth hormone, insulin, thyroid hormone, vitamin D, glucocorticoids, parathyroid hormone, calcitonin, androgens and estrogens (Reddi and Sullivan, 1980; Spencer, 1989, Caplan and Boyan, 1994). Autocrine and paracrine effects on bone cells, modulated by growth factors, prostaglandins and immune-cell-derived factors, also regulate bone growth (Thorp, 1995a, 1995b; Goldring and Goldring, 1996). Errors in the process of bone formation and resorption may lead to disorders, such as developmental orthopedic disease or osteoporosis.

Developmental orthopedic disease

Developmental orthopedic disease is a general term than includes all limb problems associated with bone growth disorders in the foal. It includes physitis,

angular and flexural limb deformities, cervical vertebral malformation (wobblers syndrome), osteochondrosis and osteochondritis dissecans.

Physitis is a term used to describe clinical flaring of the metaphyseal cartilage, commonly noted at the distal radius, distal tibia, distal metacarpus and metatarsus, and proximal first phalanx (Stashak, 1987). In the horse with physitis, an error in the conversion of cartilage to bone (see below) leads to retained cartilage. Compressive force on the cartilage as a result of natural activity of the growing horse leads to inflammation and therefore, physitis.

Physitis associated with the retention of cartilage may lead to the collapse of bone trabeculae, causing acquired angular limb deformities (Williams and Pugh, 1993). Some angular limb deformities include a genetic component. Others may be normal variations, in which the bone responds and grows into a normal orientation without intervention. Flexural limb deformities, commonly known as contracted tendons, may be congenital or acquired. Foals may be born with a flexural deformity as a result of uterine position, viral infection, or a plant toxin consumed by the mare. Acquired flexural limb deformities that result from abnormal bone formation have been associated with orthopedic pain (Bramlage, 1987). "Contracted tendons" are probably due to a shortening of the muscle more than the tendon. Affected foals alter their gait by walking on their toes in effort to alleviate joint pain, and eventually the tendon-muscle unit readjusts and shortens to the new position.

Cervical vertebral malformation, which causes one of the wobbler syndromes, is characterized by cartilage defects in the articular cartilage of the vertebrae, which

results in compression of the spinal cord (Reed et al., 1987). Affected animals exhibit irreversible neurologic dysfunction, as biomechanical factors affect subsequent development, causing degenerative joint disease of the vertebral facets and soft tissue thickening of vertebral ligaments (Williams and Pugh, 1993).

Manifestations

Bone is a highly regulated, metabolically active tissue affected by many hormones and growth factors (Spencer, 1989; Caplan and Boyan, 1990; Goldring and Goldring, 1996). A common disorder of bone growth in many species is osteochondrosis, an aberration in endochondral ossification, or the conversion of cartilage to bone (Ekman et al., 1990; Jeffcott, 1991). Osteochondrosis leads to thickening and retention of cartilage. It is prevalent in rapidly growing animals, including horses, pigs, dogs, cattle, sheep, broilers and turkeys (Nakano et al., 1987; Leach and Lilburn, 1992; Jeffcott and Savage, 1996). The disease is probably multi-etiological and includes nutritional, endocrinological, genetic, metabolic and mechanical components. Nutritional and genetic induced effects on endochondral ossification may be mediated by the endocrine system.

Endochondral ossification occurs in articular-epiphyseal and in metaphyseal growth cartilage complexes (Leach and Lilburn, 1992; Henson et al., 1996). The growth complexes have distinct zones: resting, proliferating, and hypertrophic, representing an orderly progression of chondrocytes through steps involving chondrocyte proliferation, extracellular matrix production, chondrocyte hypertrophy,

invasion of blood vessels and eventually, calcification of the matrix (Howlett, 1980; Hunziker et al., 1987; Hunziker, 1994).

The conversion of cartilage to bone is a continuum of delicate changes, morphological and biochemical. Errors in any stage of the process may disrupt the balance between cartilage formation and ossification and lead to the development of osteochondrosis (Jeffcott, 1991). A commonly reported perturbation leading to osteochondrosis is a failure of the chondrocytes to properly hypertrophy and mature (Orth and Cook, 1994; Thorp, 1995a, 1995b).

Dyschondroplasia is an abnormality of growth plate cartilage characterized by a mass of avascular cartilage extending from the epiphyseal growth plate into the metaphysis. Dyschondroplasia represents a disruption of endochondral bone formation, and the term is often used synonymously with osteochondrosis. Osteochondritis dissecans is a result of osteochondrosis or dyschondroplasia, in which cracks form in thickened articular cartilage and extend into the synovial joint. Pressure on the joint may lead to the avulsion of a cartilage flap which may break loose from the articular surface.

Estimates from over ten years ago suggested that tibial dyschondroplasia caused a loss of at least \$200 million to the U.S. poultry industry (Edwards, 1983). Today, this disease has been virtually eliminated by selective culling of affected poultry. The incidence of osteochondrosis is high in swine, but osteochondrotic lesions have not caused detrimental effects on growth rates or economic returns in pigs from birth to market weight (Nakano et al., 1987). In breeding swine, however, 10 to 40%

were culled due to leg weakness; osteochondrosis was considered an important predisposing factor (Nakano et al., 1987). In racing horses, osteochondrosis was reported as the underlying cause in 65% of fractures of the scapula, humerus, carpus and the ends of the metacarpus (Krook and Maylin, 1988).

Articular cartilage in osteochondrotic lesions of horses was biochemically, histochemically and immunohistochemically distinct from normal cartilage (Lillich et al., 1997). Articular cartilage extracts from horses with osteochondrosis had lower proteoglycan, glycosaminoglycan and chondroitin sulfate concentrations.

Endocrine effects

Growth Factors and Growth Hormone. Somatomedins, also called insulin-like growth factors, are important regulators of bone formation, known primarily for mediating the effects of growth hormone. Somatomedin C (insulin-like growth factor I, IGF-I) mediates many of the effects of growth hormone postnatally; IGF-II is less potent than IGF-I and less dependent on growth hormone (Spencer, 1989; Tsukahara and Hall, 1994). Circulating IGFs are released from the liver by growth hormone and act as endocrine regulators of bone growth. Insulin-like growth factors also act in a paracrine manner: they are secreted by the chondrocytes and bone marrow stromal cells and stored in the extracellular matrix with an assortment of IGF binding proteins (Caplan and Boylan, 1994) and act on the osteoblasts to promote further production of IGFs and associated binding proteins. Insulin-like growth factors act to promote mitogenesis of osteoblast precursors and collagen synthesis (Rosen et al., 1994). Growth hormone,

thyroid hormone, parathyroid hormone, estradiol, vitamin D and other factors may regulate the synthesis and release of IGFs and IGF binding proteins.

Fetal sheep infused with IGF-I intravascularly from 120 to 130 d of gestation had increased cross-sectional areas of epiphyses at many sites, associated with increased plasma concentrations of IGF-I and reduced plasma concentrations of IGF-II and insulin (Lok et al., 1996). Insulin-like growth factor-I was shown to be deficient in osteochondrotic cartilage of pigs and chickens (Thorp et al., 1995a, 1995b).

Growth plate chondrocytes also synthesize transforming growth factor- β , which may act as an autocrine or paracrine regulator of endochondral ossification (Tsukahara and Hall, 1994). Transforming growth factor- β has been suggested to induce chondrocyte hypertrophy and to modulate the expression of cartilage matrix proteins and metalloproteases (Caplan and Boyan, 1994; Thorp et al., 1995a). Deficiencies of transforming growth factor- β were evident in porcine and avian osteochondrotic cartilage (Thorp et al., 1995a, 1995b).

Pulsatile patterns of growth hormone secretion were examined in 25 d old male chicks. Those severely afflicted with tibial dyschondroplasia had greater peak heights and growth hormone secretory capacity than normal birds (Vasilatos-Younken and Leach, 1986). In horses, ingestion of a carbohydrate meal was suggested to facilitate growth hormone secretion (Kronfeld et al., 1990).

Insulin and Thyroid Hormone. Insulin has been shown to stimulate cartilage growth and proteoglycan synthesis in vitro (McCumbee and Lebovitz, 1980). Horses with radiographic evidence of osteochondrosis lesions had greater postprandial changes in

glucose and insulin after a starch meal than did non-lesioned horses (Ralston, 1996). Glade et al. (1984) found postprandial increases in insulin and decreases in thyroxine in weanling horses fed a diet containing 160% of NRC energy and protein recommendations.

Tri-iodothyronine has been suggested to have direct effects on epiphyseal cartilage maturation and indirect effects on chondrocyte proliferation, mediated through IGF-I (Root, 1990). Thyroxine may be osteogenic, stimulating the proliferation of resting zone chondrocytes (Spencer, 1989). Hypothyroidism in a foal caused delayed closure of epiphyseal plates, leading to osteochondrosis dissecans and subchondral bone cysts (Vivrette et al., 1984). Hypothyroidism leads to reduced growth hormone secretion; thyroid hormone maintains normal growth hormone secretion through the regulation of hypothalamic secretion of growth hormone releasing hormone (Mol and Rijnberk, 1989).

Calcitonin and Parathyroid Hormone. Calcitonin and parathyroid hormone regulate plasma calcium concentration (Spencer, 1989; Caplan and Boyan, 1990). Calcitonin decreases plasma calcium concentration by direct inhibition of osteoclasts, leading to decreased resorption of bone. Parathyroid hormone tends to increase plasma calcium concentration by stimulating osteoclasts to resorb bone. Disturbances in endochondral ossification induced in Great Dane pups fed calcium at three times the NRC requirement was attributed to a hypercalcitonemia and a hypoparathyroid state (Goedegebuure and Hazewinkel, 1986). Hypercalcitoninism was proposed as an intervening factor in osteochondrosis in the horse, caused by excessive intake of

calcium (Krook and Maylin, 1988). This hypothesis was based on histologic examination of bone.

Glucocorticoids. Prolonged dexamethasone administration in foals interfered with normal growth plate development (Glade et al., 1983). The osteolysis caused by glucocorticoid administration led to osteonecrosis, which is unlike naturally acquired osteochondrosis (Lillich et al., 1997). Decreased bone formation and increased bone resorption in hyperadrenalcorticism has been more commonly associated with osteoporosis than osteochondrosis (Spencer, 1989; Licata, 1992).

Implications. The regulation of bone growth is orchestrated by the actions of many hormones and growth factors (Figure 1.6). Osteochondrosis may be caused by either deficiencies or excesses of endocrine factors. An excess of growth hormone may lead to widening of the epiphyseal cartilage complex. Deficiencies in growth hormone, insulin-like growth factors and transforming growth factor- β may lead to the formation of osteochondrotic tissue. Hyperinsulemia and hypothyroidism may play a role in animals with osteochondrosis, through overstimulation of cartilage growth and proliferation without maturation and conversion to bone.

Nutritional influences

Skeletal development is influenced by many nutrients (Leach and Gay, 1987; Nakano et al., 1987; Jeffcott, 1991), of which some have a narrow tolerance range, as both too little and too much cause bone disease (Kronfeld et al., 1990). In some cases, restricting intake may reduce the incidence of osteochondrosis. However, if the predominant cause is a single nutrient within the ration, then restricted feeding may

either reduce the concentration of a nutrient fed in excess, or it may eliminate the deficiency of a nutrient which is manifested only at a higher overall intake. Care should be taken to identify specific contributing nutritional factors and adjust dietary recommendations accordingly, with optimal nutrition in mind (Kronfeld et al., 1994).

Energy. Energy fed in excess of requirements may contribute to osteochondrosis by increasing growth rate and weight for age or by causing nutritional or hormonal imbalance. In pigs, weight overload may result in mechanical stress on joint cartilage that may be too immature to support body weight. Restriction of energy intake in an attempt to prevent osteochondrosis in pigs has yielded inconsistent results (Nakano et al., 1987).

Bull calves consuming 16% more metabolizable energy, compared to calves fed a control diet, had 25% higher weight gains and a higher incidence of limb joint lesions (Jensen et al., 1981). Broilers fed a high energy diet had a higher incidence of tibial dyschondroplasia than those fed a low energy or high fiber diet (Orth and Cook, 1994).

A “paced growth” restrictive diet low in energy and protein (.55 times NRC) was used to retard growth rate and was claimed to arrest progression of cervical vertebral malformation in 80% of selected weanling horses (Donowick et al., 1989). These data are inconclusive: results were reported without statistical inference, as there were no weanlings fed control rations. Also, the weanlings were kept in the dark in small stalls to inhibit movement.

Foals fed energy at 130% of the NRC recommendation for digestible energy, had higher incidence of histological dyschondroplasia (Savage et al., 1993a). The data

from this study suggest that restriction of energy to one times the NRC recommendation may prevent developmental orthopedic disease, thus eliminating the need for severe dietary restriction, as practiced by Donowick et al. (1989).

Calcium. Calcium excess may affect bone by increasing growth, stimulating the release of calcitonin, and interfering with phosphorus, copper and zinc absorption from the gut, or iodine uptake by the thyroid (Goedegebuure and Hazewinkel, 1986; Kronfeld et al., 1990; Corbellini et al., 1991). Increased dietary calcium levels reduced the incidence of tibial dyschondroplasia in chickens fed diets high in phosphorus (Edwards and Veltmann, 1983). In fetuses of ewes overfed calcium, as compared to fetuses of control fed ewes, width of articular cartilage was increased, and there was histological evidence of delayed conversion of cartilage to bone (Corbellini et al., 1991).

Excessive calcium intake (three times the NRC recommendation) in Great Dane pups caused various irregularities of retained articular and physeal cartilage with disturbances in endochondral ossification (Goedegebuure and Hazewinkel, 1986).

A common nutritional association noted in the field is the expression of developmental orthopedic disease in horses occurs when grain-based concentrates designed to complement grass hay are fed with alfalfa hay (Kronfeld et al., 1996). This practice provides a diet with excessive calcium. There were no differences in histological expression of dyschondroplasia in foals fed calcium at 342% of NRC recommendations, compared to foals fed a diet containing calcium at the NRC recommendation (Savage et al., 1993b), however, this study lacked statistical power

due to the small number of animals. In the same study, foals fed high calcium (342% NRC) in combination with high DE (130% NRC) had a higher incidence of histological dyschondroplasia.

Phosphorus. A higher incidence of dyschondroplasia was found in broilers fed a diet containing a larger amount of available phosphorus (Lilburn et al., 1989), and in broilers fed diets containing high phosphorus and low calcium (Edwards and Veltmann, 1983). Foals fed a diet high in phosphorus (388% of NRC), compared to control fed foals, had a higher incidence of histological dyschondroplasia (Savage et al., 1993b).

Copper. Copper is an essential cofactor for the metalloenzyme lysyl oxidase, which is essential for the cross-linking of collagen. The induction of tibial dyschondroplasia by a copper deficient diet (.7 mg/kg copper) was first noted in Leghorn chicks (Carlton and Henderson, 1964). Cartilage lesions were reproduced in young chicks and associated with copper deficiency (Leach and Gay, 1987; Orth and Cook, 1994).

A negative correlation between copper concentrations in foal diets and scored degree of osteochondrosis was reported by Knight et al. (1985). The foals fed lower concentrations of copper had a higher incidence of gross lesions related to osteochondrosis. This study may be faulted, however, by a lack of statistical power and the inclusion of two outliers which bias the data. The National Research Council (NRC, 1989) considered the data inconclusive, but the papers aroused much interest, and additional data indicated that copper deficiency was associated with increased severity of osteochondrosis in foals (Bridges and Harris, 1988; Williams et al., 1989; Hurtig et al., 1990).

Zinc. Copper deficiency may be manifested by excess dietary zinc. Zinc inhibits copper absorption by inducing metallothionein synthesis in intestinal epithelial cells. Copper binds tightly to metallothionein and is passed from the gut when epithelial cells are sloughed, therefore inhibiting copper absorption (Ostreicher and Cousins, 1985). Zinc is required for alkaline phosphatase, an enzyme considered to be associated with osteoblastic and excessive osteolytic activity. Dietary zinc concentrations were positively correlated with serum alkaline phosphatase activity and negatively correlated with serum copper concentrations in gilts (Hill and Miller, 1983).

Osteochondrosis was found in foals consuming forages contaminated by zinc; a secondary copper deficiency was implicated (Gunson et al., 1982). Foals fed zinc in concentrations of 1000 or 2000 mg/kg, as compared to those fed 40 or 250 mg/kg, were severely lame in less than 10 wk and had fractures of growth plate and articular cartilage through the zone of hypertrophic cells (Bridges and Moffitt, 1990).

Implications. Nutritional factors have been implicated by epidemiological and experimental studies in the etiology of developmental orthopedic disease. Due to its multiple etiological nature, no single form of nutritional intervention has been found effective in preventing or correcting the disease.

Summary

Examination of northern Virginia pasture studies indicate a need for supplementation for optimal nutrition of grazing horses. Considering the seasonal variation in pasture, particularly in carbohydrate fractions, current supplements rich in

hydrolyzable carbohydrate may be lacking. Although it is considered that pasture provides adequate fiber for nutrition of the horse, some supplementation of fiber during periods of rapid pasture growth may be beneficial. Milk composition of mares may be influenced by dietary intake of fat and fiber, and thus foal nutrition may be affected. Endocrine and metabolic fluctuations associated with the feeding-fasting cycle may cause perturbations in normal growth and, in particular, growth and maturation of cartilage and bone.

Our objective in the design of an optimal nutritional supplement suitable for grazing horses was to test a new type of supplement, one that replaced the grain and molasses in the traditional sweet feed with *fat* and *fiber*. The specific objectives fit questions previously addressed in the literature: the effects of supplementation on growth rates and bone development of foals, milk composition of mares and carbohydrate status.