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Screening to Prevent to Carential and Metabolic Disease and HPTNS of Equids Grazing Forage Grasses with Unbalanced Levels of Minerals, Through the Mineral Profile and Creatinine Clearance Ratio for Ca and P Assessment

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Abstract: In the present study, 229 horses were evaluated, belonging to 76 properties, 25 Municipalities of the Center-Western region of São Paulo State, covered for a period of clinical observations from 1984 to 2010. Were performed sampling grazing elephant grass and various supplements for dosages minerals, being macro and microminerals and oxalate content in dry matter (DM) of *Brachiaria humidicola* and elephant grass (*Pennisetum purpureum*). The equids of farms with a history of skeletal or osteoarticular problems were evaluated for Calcium (Ca), Phosphorus (P) and Creatinine (Cr) in urine and serum to determine the percentage of Cr depuration for the Ca and P (%DRCr-Ca and P). The clinical observations of osteopathies were determined. The main lesions found in animals after death were evaluated. The analysis leaf mineral of *B. humidicola* presented values ranging from 2.40 g Ca to 3.68 g Ca/kg DM and 0.40 g P to 0.74 g P/kg DM. The high toxic levels of oxalate ranged from 1.15-3.10% oxalic acid in DM of *B. humidicola* and 4% in DM of elephant grass. The higher Ca/Oxalate ratio was 0.37 (*B. humidicola*) and the lower of 0.08 (*P. purpureum*). The largest amounts were excreted 58 mg of Ca and P 47.50 mg/dL in the urine and the lowest was 4 mg Ca and 3.50 mg P/dL of urine. The clearance of ratio, ranged from 0.63-6.28% (μ = 2.48; sd ± 1.28; cv = 51.61%) for %DR- Ca, while the %DR- P ranged from 1.23-35.92% (μ = 17.33; sd ± 8.35; cv = 48.15%). 50% of values of %DRCr-Ca and 95% of values of %DRCr-P were considered abnormal, thus predisposing to osteopathy and renal dysfunction as glomerulonephritis and nephritis. Above of 50% of the animals of properties, presented one or more types of skeletal tissue disease, affecting bones flat (osteochondrosis) or long bone (epiphysitis). Young animals showed accelerated growth in bone problems of members. Animals at play and hard work showed symptoms of nutritional secondary Hyperparathyroidism (HPTNS) when presenting the fibrous osteodystrophy ("swollen face disease") in addition to problems with the locomotor apparatus and in general with the skeletal system. The diagnosis in a timely manner the problem of imbalance in the diet using the technique of %DRCr-Ca and P possible prevention of osteopathies, those reversible. The change of grazing animals to forage less oxalic acid content (<0.50%) and appropriate supplementation prevented the problem, except frames and pathophysiology of chronic glomerulonephritis due to HPTNS. Young animals in accelerated growth showed bone problems of members. Animals in reproduction and hard work showed symptoms of HPTNS, presented signs of fibrous osteodystrophy, besides problems with the locomotor system and in general with the skeletal system. The diagnosis in time habile of the problem of imbalance of diet through of the assessment of %DRCr- Ca and P, enabled prevent the osteopathies, those reversible. The change of the animals to pasture with forages of minor oxalic acid content (<0.50%) and receiving adequate supplementation prevented the problem, except frames chronic pathophysiological of HPTNS and glomerulonephritis.

Key words: Nutritional secondary hyperparathyroidism, calcium, phosphorus, renal clearance, equine, metabolic disease

INTRODUCTION

The equids (*Equus*) are associated with vast patrimony of importance social, cultural, economic, ecological, scientific exploration, education and research. The horse industry has become a very important scene in many areas of livestock country. A major increase in research

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on physiological factors affecting production and performance has resulted from the popularity of the horse and the economic impact of the industry; however, many of the problems have not been solved. The diseases resulting from nutrition, cause enormous prejudice. In equids, the abnormalities of the skeleton are limiting factor in farm animals development. The absence of mechanisms that make one shielding the skeleton in order to avoid reabsorption of bone minerals and its destructuring, makes the animals susceptible, when the deficiencies or nutritional imbalance, the number of osteopathy (Schryver *et al.*, 1974a,b; Raisz and Kream, 1981; Hintz, 1996; Carvalho *et al.*, 2010).

In the mineral deficiency of Calcium (Ca) and Phosphorus (P) in diet and the increased demand by exercise (work), pregnancy, lactation or growth, the body in this conditioning to maintain homeostasis in bloodstream of Ca and P, at the expense of the reserves of the skeleton. The parathyroid is sensitive the decline in Ca in bloodstream, so the organism makes use of the reserves of calcium in bone tissue. If there is deficiency or imbalance of calcium and phosphorus in the diet, bone reserves are not restored via mechanisms to increase intestinal Ca absorption and reabsorption of filtrate (Argenzio *et al.*, 1974; Hintz and Schryver, 1976). Under these circumstances, in continuous production of parathyroid hormone, nutritional secondary Hyperparathyroidism (HPTNS) arises. In persistent cases, the kidney function can also be compromised leading the nephritis and glomerulonephritis. Imbalance or deficiency in the tissue followed by demineralized bone resorption can be filled with fibrous scar tissue, causing the swelling of the facial bones in the process called fibrous osteodystrophy, characterized by a bilateral swelling of the facial bones, known as swollen face, that when in advanced stage is clearly visible on the face, mandible and maxilary of the equids (Krook and Lowe, 1964; Krook, 1968; Joyce *et al.*, 1971).

Although irreversible, these lesions can be controlled if detected. Over 99% of body calcium found in bones and teeth. According to Wasserman (1984), the bone mineral crystal structure of the apatite series consists of hydroxyapatite of formula $Ca_{10}(PO_4)_6(OH)_2$ and Ca salts type tricalcium phosphate and calcium carbonate. According to Krook (1968), the constant renewal of bone tissue is the basis for classification of morphology and pathogenesis of metabolic bone disease characterized by osteopenia.

According to Tisserand (1986) between the particularities of the horse's digestion, 60-70% of the Calcium is absorbed in the intestine is excreted by the kidneys (indepedent of Ca ingested) and the endogenous Ca fecal loss equal to about 20 mg to 25 mg/kg Body Weight/day (BW/d) with isotope marked (Schryver *et al.*, 1970a,b). Because of the loss endogenous, assume if the requeriment for replacement of 30 mg/kg BW/d.

In turn, the endogenous loss of P-labeled radioactive isotope is about ±10 mg P/BW/d (Scrhyver *et al.*, 1971a). During intake of high roughage rations precaecal P secretion increase and the large intestine is the major site of P absorption, while feeding concentrates P is preferently absorbed in the small intestine. Fermentation in large intestine may increase the endogenous losses of P by 20%.

According to Meyer (1986) the recommendation for the daily mineral supply in horses are in maintenance, 50 mg Ca and 30 mg P/kg BW/day and heavy work 67 mg Ca and 49 mg P/kg BW/d. The utilization of ingested minerals (%) are 60% Ca with lower rates and with high phytic P, oxalic acid intake and vitamin D deficiency and 40% of P with higher rates for anorganic P compounds (Schryver and Hintz, 1972). The Ca: P ratio has been a source of much debate in the past. The major sites of Ca absorption in the small, P in the large intestine, it is not surprising that a Ca excess is less detrimental on P absorption than a P excess on Ca absorption. The absolute levels of these minerals are adequate, the horse can tolerate as wide range of Ca: P ratios. However, the lower Ca than P intake should be prevented. However, Staun (1986) in practical aspects of horse nutrition, considered the rate 0.6% Ca (4.5 g/100 kg live weight) to 0.4% P (3.0 g/100 kg BW).

The daily calcium requirements for lactation range from 1.2 g/kg of fluid milk during the first postpartum week to 0.8 g/kg of fluid milk during weeks 15 to 17 postpartum. For a 500-kg mare producing 15 kg of milk/day in early lactation and with a calcium absorption efficiency of 50%, the daily dietary calcium requirement would be 36 g (15 kg x 1.2 g/0.5) for milk production, in addition to 20 g for maintenance (Schryver *et al.*, 1986).

Calcium homeostatic mechanisms maintain serum calcium within a narrow range. Therefore, serum calcium may not be a reliable indicator of calcium status. In the mature horse, inadequate dietary calcium can result in weakening of the bones and an insidious shifting lameness (Krook and Lowe, 1964).

Inadequate calcium intake by the developing foal can lead to rickets which is characterized by poor mineralization of the osteoid tissue and the probability of enlarged joints and crooked long bones. From a survey of the severity of metabolic bone disease in yearlings and diet analyses in horse farms. In these farms were reported a negative linear relationship between dietary calcium intake and perceived severity of metabolic bone disease in young horses. Farms with yearlings having the lowest incidence of metabolic bone disease were fed diets containing 1.2% calcium, whereas yearlings with the most severe metabolic bone disease were on farms that fed diets with 0.2% calcium (Knight *et al.*, 1985).

Diets with calcium:phosphorus ratios of 1.16:1 (0.43% Ca) and 4.12:1 (1.96% Ca) and observed a greater proportion of lamellar bone than osteonic bone in the high-calcium horses; however, no clinically deleterious effects or gross morphologic differences were detected. Calcium has been fed at more than 5 times the required level without detrimental effects, provided the level of phosphorus is adequate (Jordan *et al.*, 1975).

Phosphorus makes up 14-17% of the skeleton. In addition, it is required for many energy transfer reactions associated with Adenosine Diphosphate (ADP) and Adenosine Triphosphate (ATP) and for the synthesis of phospholipids, nucleic acids and phosphoproteins (El Shorafa *et al.*, 1979).

The efficiency of estimated true phosphorus absorption ranges from 30-55% and varies with the age of the animal and with the source and concentration of dietary phosphorus. Phosphorus absorption is likely to be higher in foals fed milk than in older horses.

Phosphorus in the form of phytate is poorly absorbed; however, phytate phosphorus may be partially available because there is some phytase in the equine lower gut (Hintz *et al.*, 1973). Phosphorus retention is depressed in natural diets containing high concentrations of total oxalates (Walthall and Mckenzie, 1976; Swartzman *et al.*, 1978; Blaney *et al.*, 1981; Mckenzie *et al.*, 1981a,b). In this document, a true phosphorus absorption efficiency of 35% is used for all idle horses, gestating mares and working horses because they consume primarily plant sources of phosphorus; a value of 45% is used for lactating mares and growing horses because their diets are typically supplemented with inorganic phosphorus.

At a phosphorus absorption efficiency of 35%, the phosphorus requirement for maintenance is 10 mg/0.35 or 28.6 mg/kg of body weight/day or 0.87 g of phosphorus/Mcal of DE/day. A mature, 500-kg horse would, therefore, require 14.3 g of phosphorus/day for maintenance. The phosphorus requirements for optimal bone development in growing foals are based on estimates that growing horses deposit approximately 8 g of phosphorus/kg of body weight gain. Thus, a 215-kg foal with a phosphorus absorption efficiency of 45 percent and gaining 0.85 kg/day would require about 15.1 g of phosphorus (8 g x 0.85 kg/0.45) in addition to its maintenance requirement of 4.8 g (215 kg x 10 mg/0.45). During late gestation and lactation, phosphorus requirements increase. Phosphorus requirements for the products of conception for mares in months 9, 10 and 11 of pregnancy have been estimated to be 7, 12 and 6.7 mg/kg of BW/d, respectively. At 35% absorption efficiency, the daily phosphorus requirement for a 500-kg mare for products of conception during gestation months 9, 10 and 11 would be 10, 17.1 and 9.6 g, respectively (mean 12.2 g/day). The mean daily phosphorus deposition for the last 3 months of gestation was added to maintenance needs and divided by the mean DE requirement for the same period. This value (14.1 g of P/Mcal of DE/d) was then multiplied by the daily DE requirement for each month (Schryver *et al.*, 1974a,b; NRC, 2007).

The P concentration of mares' milk ranges from 0.75 g/kg of fluid milk in early lactation to 0.50 g/kg of fluid milk in late lactation If the absorption efficiency is 45%, the daily P requirement above maintenance for lactation would be 25.0 g for a mare averaging 15 kg of milk/day in early lactation and 11.1 g for the mare producing 10 kg of milk/day during late lactation. At these rates of milk production, a 500-kg mare would require 36 and 22.2 g of phosphorus/day in early and late lactation, respectively.

The Ca:P ratio is an important criterion for equine diets. Ratios less than 1:1, i.e., when phosphorus intake exceeds calcium intake, may be detrimental to calcium absorption. Even if the calcium requirements are met, excessive phosphorus intake will cause skeletal malformations (Schryver *et al.*, 1971b; Nazario *et al.*, 1986). Ratios of Ca to P as high as 6:1 in diets for growing horses may not be detrimental if P intake is adequate (Jordan *et al.*, 1975).

Ca and P are considered together because they are so closely interrelated in the mineralization of osteoid. A deficiency of either will result in reduced mineralization. A clinical manifestation if improper mineral nutrition is so-called “big-head” disease (Nazario *et al.*, 1986; Hintz, 1996) or HPTNS caused by diets containing insufficient calcium or excessive P. This condition usually occurs when horses are fed high grain diets combined with poor quality roughage but it may also occur when there is malabsorption of calcium or horses that have grazed in *Setaria* sp. grass containing high amounts of oxalate and oxalate has been shown markedly inhibit calcium absorption by ponies. Lameness in horses with big-head disease has been attributed to preferential resorption of the outer circumferential lamellae of cortical bone, with consequent loss of osseous support for the periosteum, tendons and ligaments (Swartzman *et al.*, 1978; Mckenzie *et al.*, 1981a,b).

Young horses and ponies fed calcium-deficient diets grow normally in height and weight, at least under experimental conditions when the calcium deficiency is neither prolonged nor severe. This is due in part to the large reserve of Ca already in the skeleton which may be drawn upon and used for new bone growth and mineralization. The evolution of cases of HPTNS are characterized by facial enlargement, thickening of the mandibular rami and transitory lameness.

The kidney plays a distinct role in the homeostasis of Calcium (Ca) and P in the equine species. The practice of calculating the DRCr%-P is considered applicable in specific situations where there is subclinical and clinical signs consistent with dysfunction of mineral determinants of acute or chronic bone disease, among others, fibrous osteodystrophy, osteitis, angular deformities, contracture flexor tendons, osteochondrosis, lameness, fractures of lumbar and sacral vertebrae (Hintz *et al*., 1979; Mason *et al*., 1988). In these, as well as chemical analysis, complete food, biochemical analysis of P in serum and urine can be accomplished, given the complexity of the interrelationships minerals, animal metabolism and wide variations in the true digestibility of P of different ingredients diet (Van Soest *et al*., 1991). Thus, this research aimed to evaluate the efficiency of %DRCr- P, to detect early, through biochemical measurements in serum and urine sampled simultaneously at different sampling times, the nutritional status of P in horses fed diets with levels P variables in controlled experimental conditions.

So overall, will be needed a collection of urine for 24 h to determine the renal loss of electrolytes.

Importance of electrolyte metabolism in the pathogenesis and diagnostic and management of a broad array of clinical syndromes. The concentration of electrolytes is a function of intestinal absorption, renal reabsorption, bone deposition, fecal loss, sweat loss and renal excretion.

This is due to the variation in water content of urine and the reciprocal variation in electrolyte concentration. Unfortunately, collection of a 24-h urine sample is impractical under clinical conditions. One alternative to measuring the total volume of excreted electrolyte is to quantify electrolyte clearance. Fourtuitously, the clearance if creatinine by the horse is almost constant, being primarily a function of glomerular filtration. Since creatinine is being produced at all times by metabolism of muscle energy, there is no need to inject creatinine into the horse measure creatinine clearance. Thus, the clinician can compare the clearance of any electrolyte to the clearance of endogenous Creatinine (Cr) and determine with reasonable confidence the rate of renal electrolyte output and multiplying the equation by 100: [(X (U)/ x (S)) x (Cr (S)/Cr(U))] x 100 (Knudsen, 1959; Traver *et al*., 1976, 1977; Rawlings and Bisgard, 1975).

Previous discussion of parathyroid pathophysiology can be evaluated by technical of %Cr-Ca and P. Under normal circumstances, horses excrete large amounts of Ca and because of the alkalinity of herbivorous urine, calcium carbonate ($CaCO_3$) crystals are often copious, settling to the bottom of the urinary bladder. The crystals can be dissolved by adding acid to a given aliquot of urine but the Ca is spuriously distributed in the bladder. The PO^-_4 increased in four important clinical situations: 1- primary or pseudo hyperparathyroidism, 2- renal failure, 3- PO^-_4 wasting nephropathy associated with multifocal recurrent lamness and 4- HPTNS. The animal shows clinical signs and the percentage of creatinine clearance is high. Grain-producing areas where the diet is high grain. Because one the actions of parathormone is to increase excretion of PO_4 via urine. P wasting nephropathy may be linked to multifocal recurrent lameness. I saw this condition in Arabian stallion that had a %Cr PO^-_4 value of 45% (Coffman, 1980).

Minerals are involved in a number of functions in the body, including formation of structural components, enzymatic cofactors and energy transfer. Some minerals are integral parts of vitamins, hormones and amino acids. The horse obtains most of the necessary minerals from pasture, roughage and grain. The mineral content of feeds and the availability of minerals vary with soil mineral concentrations, plant species, stage of maturity and conditions of harvesting. The resulting variations should be considered in assessing an animal's mineral status and formulating appropriate diets. The higher number of possible of minerals must be analyzed. Between they, seven macrominerals (calcium, phosphorus, potassium, sodium, chloride, magnesium, sulfur) and eight microminerals (cobalt, copper, fluorine, iodine, iron, manganese, selenium, zinc) are of the utmost importance to the development of the skeleton and need to be assessed (Argenzio *et al*., 1974; Hintz and Schryver, 1976; Carvalho *et al*., 1988; 1990; 2008a,b). Between the macrominerals, the Ca makes up about 35% of bone structure and is involved in other body functions including muscle contractions and blood clotting mechanisms. The true absorption efficiency is believed to decline with age and to range from as high as 70% in young horses to 50% in mature horses. For purposes of estimating calcium requirements herein, a calcium absorption efficiency of 50% is used for all ages of horses (El Shorafa *et al*., 1979).

The values obtained for cortical area gave a clear indication of the formation and resorption of osseous tissue during growth and aging.

Other factors affecting calcium absorption include the dietary concentrations of Ca, P, oxalate and phytate. Swartzman *et al*. (1978) reported that 1% oxalic acid in equine diets reduced Ca absorption approximately 66%. Total dietary oxalate concentrations of 2.6-4.3% produced negative calcium balance, during which fecal calcium doubled and urinary calcium decreased in comparison to control horses (Mckenzie *et al*., 1981a,b). Blaney *et al*. (1981) observed similar negative balances for calcium and phosphorus in horses fed various tropical grass hays of more than 0.5% total oxalate. They concluded that HPTNS may occur when the calcium-oxalate ratio on a weight-to-weight basis is less than 0.5. Hintz and Schryver (1976) reported no difference in absorption of calcium from alfalfa containing 0.5 and 0.87% oxalic acid, in which the calcium:oxalate ratios were 3 and 1.7, respectively.

Estimates of endogenous losses of 20 mg of calcium/kg of body weight/day have been made (Schryver *et al*., 1970a; 1971b). If the absorption efficiency of dietary calcium is 50%, a 500-kg horse would require 20 g (500 kg x 20 mg/0.5) of dietary calcium or 0.04 g of calcium/kg of body weight/day or 1.22 g of calcium/Mcal of DE/day for maintenance. Schryver *et al.* (1974a,b) estimated that growing foals deposit approximately 16 g of calcium/kg of gain. Thus, a 215-kg foal gaining 0.85 kg/day and having a calcium absorption efficiency of 50% would require 27.2 g/day (16 g x 0.85 kg/0.5) of dietary calcium for skeletal growth plus 8.6 g/day (215 kg x 20 mg/0.5) to meet endogenous losses.

Any increase in the calcium requirement associated with exercise (work) appears to be readily met by an obligatory increase in calcium intake as dry matter consumption increases to meet energy requirements. Special calcium requirements occur in late gestation and during lactation. Approximately 11.1, 25.3 and 11.4 mg of calcium/kg BW of mare are deposited daily in the fetus and membranes of mares in months 9, 10 and 11 of gestation, respectively (Teeter *et al.*, 1967; Hintz *et al.*, 1979; NRC, 1978; Meyer, 1986). If the absorption efficiency is 50%, the calcium requirement of a 500-kg mare during months 9, 10 and 11 of gestation would be 11, 25 and 11 g/d (µ = 15.9 g/d) respectively, for fetal development. Data on the deposition rate of minerals in the fetus are very limited; therefore, a mean deposition rate was used for the last 3 months. The mean dietary calcium required for tissue deposit was added to maintenance needs and divided by the mean DE requirement for the same period. This value was then multiplied by the daily DE requirement for each month.

Parathyroid Hormone (PTH) increases the concentration of Ca in the extracellular fluid through its effects on bone, kidney and gut. In turn, the increase of plasmatic Ca through the negative-feedback mechanism determines on the parathyroid gland the suppression of secretion of the hormone. The high Ca is the most potent regulator of PTH. The PTH increases the concentration of calcium in the extracellular fluid through its effects on bone, kidney and gut. In turn, the increase of plasmatic Ca through the negative-feedback mechanism to determine on the parathyroid gland suppression of secretion of the hormone. The high Ca is the most potent regulator of PTH (Habener, 1981).

Three differentiated cell types, the chondrocyte, the osteoblast and the osteoclast, are involved in skeletal development. The chondrocytes and the osteoblast are derived from the same mesenchymal precursors but develop quite differently (Raisz and Kream, 1981).

The monogastric herbivore equid, has a wide tolerance for variations in Ca and P in the diet. The pastures provide some of the essential nutrients and elsewhere must be provided in the form of supplements. However, if the pasture is rich in complexation with calcium, or fungi present hepatogenous (hepatotoxic), the example of *Phytomyces chartarum*. This should be considered in the formulation additional (Swartzman *et al.*, 1978; Malavolta *et al.*, 1988). In this research, pastures of *B. humidicola* in the region center-west showed variable content of 2.4-4.0 g Ca/kg DM and 0.5-1.1 g P/kg DM of forage, denoting insufficiency, mean as much Ca P. showed also high levels ranging from 1.0-2.0% of oxalic acid (average of 1.5%) in the DM of forage. Several of these grasslands also showed the presence of the fungus *Pitomyces chartarum*, photosensitization agent (Carvalho *et al.*, 1990; Schenk *et al.*, 1991).

In the present study were evaluated equids belonging of properties in municipalities of the center-west of São Paulo State, covered by clinical observation period from 1984 to 2010. Samples were grazing elephant grass and various supplements, dosages for minerals, macro and micro minerals and oxalate content of *B. humidicola* and elephant grass. The equids of property with a history of skeletal or osteoarticular problems were evaluated for Ca, P and creatinine in urine and serum to determine the percentage of creatinine clearance for the Calcium and Phosphorus (%DRCr-X).

MATERIALS AND METHODS

The study was developed in the equine-rearing farms in the center-west region west of Sao Paulo State. To study the seasonal behavior of the forage in *Brachiaria humidicola* or Elephant grass accessions purple (*Pennisetum purpureum* Schum), utilized for grazing equids in this region of the state were randomly selected twenty randomly selected properties in the municipalities studied represented by Avaí, Agudos, Arealva, Lucianopólis, Uru, Pongai, Bauru, Cabrália Paulista, Iacanga, Lins, Reginopólis, Promissão, Jau, Dois Córregos, Brotas, Itirapina, Torrinha, São Carlos, Bocaína, Mineiros do Tiête, Duartina, Piratininga, Barra Bonita, Pederneiras and Itapólis. Samples of the grass were analyzed for minerals, macro: Ca, P and Mg and micro elements: Cu, Zn, Co, Fe, Mn (AOAC, 1990). The activities of field survey, collecting material for laboratory analysis, extended the period 1984 to 2002 and also the clinical observations made beyond this period is extended until the year 2010. The sampled animals showed clinical signs of arthropathy and were aged between 8 months to 12 years of age. In the present study were evaluated clinically 229 equids belonging of 76 properties in 25 municipalities of the center-west.

Among the population of equids that showed signs and symptoms of skeletal disease, animals with symptoms classical of osteopathies were selected for clinical monitoring and sampling and laboratory analysis. In this properties, were performed studies complete with measurements of field and laboratory, being sampled all of the dietary ingredients, including pasture and concentrate for analysis of macrominerals and trace minerals.

The properties had pasture land divided into paddocks and other management area constructed in masonry, concrete floor and provided with cement feeders where the animals were given feed concentrate. The final diet (concentrate more pasture forage) were classified according to the proportion of Ca and P in the final diet: Ca normal: P higher, $\geq$2.1 Ca:>1.0 P ratio; Ca lower:P normal, <2.0 Ca: 1 P ratio; Ca lower: P higher, <2.0 Ca: >1 P ratio.

The chemical components of the ingredients of the experimental diets were analyzed in the laboratory analysis mineral of Section of Carential and Metabolic Diseases of Institute Biológico-SP, according to the methodology described by AOAC (1990). The Dry Matter (DM) of the standardization sample was obtained after drying in the oven at 65°C for 24 h for standardization of DM in 90%. After grinding in Wiley mill type knife with stainless steel and free from contamination by minerals, the samples were transferred to Erlenmeyer flask and subjected to acid digestion in a hot plate with the aid of digestor wrapped inside chapel of exhaust gases. After digestion hot of the dry matter in the presence of the mixture sulfuric, nitric and perchloric acid, the mineral composition of Ca, Mg, Mn, Zn, Fe, Co and Cu of the experimental diets and of bone ash. The extract obtained by wet digestion was transferred to volumetric flask and measured the volume with distilled and demineralized water obtained in distiller and deionized apparatus, being both with internal circuit entirely of glass. The analysis were determined by atomic absorption spectrophotometry brand Varian®. Phosphorus was determined colorimetrically by the method of Fiske and Subbarow (1925). The reading obtained for the mineral in question was applied the formula to calculate the concentration according to the volume used in the dilution of concentrated extract obtained.

Biochemical analysis: The full blood to obtain the serum was collected by puncturing the jugular in a glass tube type vacationer without anticoagulant. Urine was collected directly from the bladder with a probe of stainless steel and sterile preceded by thorough asepsis of site. The serum and urine samples were obtained daily with simultaneous sampling of symptomatic animals. To evaluate renal function, urinalysis was performed after centrifugation an aliquot of urine sample for the study of sediment microscopy of crystals, cells desquamation and flora present in the urinary tract.

In the properties which had animals with signs and symptoms of facial swelling, epiphysitis, laminitis and injuries, osteo-articular (Fig. 1) were selected, in order to make viable attempt sampling urine voiding spontaneous or by inserting metal probes directly from the bladder and after obtaining the urine proceeded immediately to collect blood to obtain serum. Of animals sampled for urine and blood serum complete blood counts were performed. All analysis of Calcium (Ca), Phosphorus (P) and Creatinine (Cr) in serum and urine to determine the clearance percent ratio of Ca and P (%DRCr-Ca) were measured by the method Kit Labtest® kit-Diagnostic Systems Ltda., by colorimetric reaction and with reading in absorption spectrophotometer absorption molecular model Micronal 382-B, in Laboratory Animal Pathology of UPD Bauru/APTA-SAA-SP.

Principles of method for spectrophotometric (molecular absorption) determination of calcium in serum and urine: Calcium (Ca) reacts with purple ftaleina in alkaline solution forming a complex of purple color with maximum absorption at 570 nm.

Reagents: Buffer, color reagent and standard of 10 mg/dL. To measure the urine, homogenize, 5 mL and add 1 drop of hydrochloric acid concentrated. For reading, utilize the orange green filter (550-590 nm). Calculate: mg Ca/dL = [(Absorbance of test)/(Absorbance of standard)] x 10. Due to the high reproducibility that can be obtained with the method, the method of factor can be used: Calibration factor = 10/Absorbance of standard; mg Ca/dL = absorbance of the test x Factor. Interpretation and clinical significance: serum Ca is maintained within physiological limits by the combined action of parathormone and vitamin D through their effects on bones, intestines and kidneys. Principle of the method for spectrophotometric (molecular absorption) determination of Phosphorus (P) in serum and urine: Ions Phosphorus (P) reacts with the molybdenum in acid medium to form a yellow complex which in the action of alkaline buffer is reduced to molybdenum blue which is measured colorimetrically at 650 nm. Reagents: Catalyst, Molybdate reagent, buffer and standard of 5 mg/dL. After addition of reagents to a sample of serum, or urine to be measured, the absorbance of the test is read at 650 nm or red filter (640-700 nm) by adjusting the zero, with the white. For dosage in the urine sample, mix, 10 mL and adjust pH between 1 and 3 with concentrated hydrochloric acid to dissolve the phosphate crystals. Dilute urine 1:10 with deionized water and multiply the result by 10.

Read on red filter (640-700 nm). Calculate: mg P/dL = [(absorbance of test)/(absorbance of standard)] x 10. Due to the high reproducibility that can be obtained with the method, the method of factor can be used: Calibration factor = 5/absorbance of standard; mg P/dL = absorbance of test x Factor. Interpretation and clinical significance: Although chronic renal failure and hyperparathyroidism, are the two most common causes of hyperphosphataemia, several conditions can cause hyper phosphatemia transient and asymptomatic.

Creatinine (Cr): Principle of the method for spectrophotometric (molecular absorption) determination of Cr in serum and urine:- Creatinine reacts with picrate solution in alkaline, forming a red color complex which is measured by molecular absorption spectrophotometry. Reagents: Picric acid, Buffer, acidifier and Standard of 4 mg/dL. Take 3 test tubes and prepare the blank and standard test. Homogenize and place in water bath at 37°C for 10 min. Determine the absorbance of the test and standard at 510 nm or green filter (500-540), adjust the zero of the spectrophotometer with the blank. The absorbance of the test will be A1. Add acidifier and after five minutes read the absorbance at 510 nm.

The absorbance of the test will be A2. For dosage urine, homogenize and dilute urine at 1:25 (0.2 mL urine + 4.8 mL of water deionized). Read green filter (500-540 nm). Multiply the result by 25. Calculate: mg Cr/dL = [(A1-A2)/(absorbance of standard)] x 4. Due to the high reproducibility that can be obtained with the method, the method of factor can be used: Calibration factor = 4/absorbance of standard; mg Cr/dL = A1-A2 x factor. Interpretation or clinical significance: The constancy in formation and creatinine excretion index makes it a lot, useful, renal function, particularly glomerular filtration. In diseases chronic renal, creatinine rises more slowly than urea, however, decreases more slowly with hemodialysis.

Oxalic acid in Dry Matter (DM) of *B. humidicola* and Elephant grass accessions purple (*Pennisetum purpureum*, Schum) were determined by the technique of thin layer chromatography, using as a control in the chromatographic run, the standard solution of oxalic acid, by known concentration test identification and quantification of the sample solution.

Statistical analysis of data, the results of the determinations of %DRCr-Ca, Ca and Cr in serum and urine, minerals and oxalate were analyzed for mean, standard deviation and coefficient of variation by using the software "Statistical Analysis System" (SAS, 1994).

RESULTS

After centrifugation of the aliquot, urine samples were subjected to the study of sediment microscopy to analyze the presence of crystals and cell desquamation urinary tract abnormalities and with observation of the flora present in the urine samples were noted changes atypical for the species. The determination of the daily clearance of Cr showed altered metabolism or renal dysfunction of animals under the influence of the treatments during the experimental research. Percentage of the animals showed clinical signs compatible with bony pathologies.

After obtaining the results the values of Ca, P and Cr (X) in serum and urine were interrelated in the formula to calculate the described by Nordin (1958); Knudsen (1959); Rawlings and Bisgard (1975); Traver *et al.* (1976) and Coffman (1980): %DRCr-: [(X U÷X S)/(Cr S÷Cr U)] x 100.

The means for the macro minerals: Ca, P and Mg and micro elements Cu, Zn, Mn, Fe and Co and total oxalate and Ca/Oxalate ratio in grasses analyzed are presented in Table 1.

Table 1: Mineral analysis of *Brachiaria humidicola* and elephant grass accessions purple (*Pennisetum purpureum* Schum) used grazing farms equids

Propriety	Ca	P	Mg	Cu	Zn	Mn	Fe	Co	Oxalate (OX)	Ca/OX ratio
1	0.24	0.10	0.26	6	70	156	116	0.100	1.79	0.13
2	0.28	0.08	0.20	4	28	163	68	0.090	1.82	0.15
3	0.28	0.09	0.23	6	20	148	80	0.100	1.92	0.15
4	0.42	0.06	0.18	4	58	118	234	0.110	1.84	0.23
5	0.40	0.08	0.16	4	34	134	198	0.100	1.73	0.23
6	0.38	0.08	0.15	6.5	24	93	315	0.115	1.78	0.21
7	0.41	0.05	0.17	5	30	78	250	0.090	1.91	0.21
8	0.45	0.08	0.19	4	31	70	301	0.085	2.20	0.20
9	0.25	0.07	0.16	6	32	88	267	0.095	3.10	0.08
10	0.37	0.06	0.15	7	56	102	256	0.085	2.20	0.17
11	0.39	0.09	0.16	5	46	56	312	0.070	4.00	0.10
12	0.43	0.04	0.17	6	57	76	187	0.095	1.15	0.37
13	0.47	0.07	0.15	4	36	92	89	0.065	1.56	0.30
14	0.38	0.11	0.16	7	28	83	230	0.075	1.70	0.22
15	0.36	0.05	0.14	4	42	58	98	0.090	1.60	0.23
16	0.41	0.09	0.19	5	47	79	178	0.100	1.82	0.23
17	0.39	0.07	0.21	7	51	72	205	0.065	1.45	0.27
18	0.29	0.05	0.21	8	45	68	227	0.080	1.69	0.17
19	0.33	0.08	0.17	6	42	70	194	0.075	1.89	0.17
20	0.42	0.07	0.18	6	39	69	175	0.090	2.21	0.19
µ	0.37	0.07	0.18	5.53	40.80	93.65	199.00	0.089	1.97	0.20
sd	0.07	0.018	0.03	1.25	13.05	32.77	76.71	0.014	0.61	0.067
cv	18.26	24.69	16.91	22.6444	31.99	34.99	38.55	15.71	31.19	33.38
Total (%)	20/20 (100%)	2/20 (10%)	-	-	-	-	-	-	20/20 (100%)	20/20 (100%)

µ: Mean; SD: Standard Deviation; CV: Coefficient of Variation.

[a]Total (%): Ca (100%) and P (10%) normal values; Oxalate (100%) and Ca/oxalate (100%) values abnormal in the forages grass

Table 2: Mean % DCr-Ca and P in equids with osteopathies grazing *B. humidicola*

										Urine	
Obs	Ca S[1]	Ca U	Cr S	Cr U	DRCr-Ca (%)	PS	PU	DRCr-P (%)	Ht	pH	D
1	10.00	58.00	1.30	120.00	6.28	3.00	25.00	9.03	42	8	1.046
2	10.50	5.00	1.70	95.00	0.85	4.00	30.00	13.42	37	9	1.028
3	11.00	4.00	1.90	110.00	0.63	4.10	45.00	18.96	36	8	1.025
4	12.00	7.00	2.00	115.00	1.01	5.30	38.00	12.47	37	8.5	1.026
5	11.50	12.00	1.88	72.00	2.72	3.50	28.00	20.89	39	7.5	1.030
6	10.60	15.00	1.70	87.00	2.77	5.25	39.00	14.52	41	8	1.031
7	10.85	8.00	1.89	65.00	2.14	4.32	34.00	22.88	40	7	1.029
8[a]	9.88	6.00	2.20	67.00	1.99	4.30	42.00	32.07	39	8.5	1.032
9[b]	10.45	4.00	1.48	41.00	1.38	4.12	41.00	35.92	33	9	1.033
10	10.95	9.00	1.69	102.00	1.36	4.51	36.00	13.23	42	7.5	1.034
11	11.20	10.00	1.92	126.00	1.36	3.99	37.00	14.13	41	8.5	1.026
12	11.43	34.00	2.12	214.00	2.95	2.12	3.50	1.23	37	8	1.030
13	10.85	23.00	1.76	111.00	3.36	3.15	21.00	10.57	36	7	1.031
14	11.05	13.00	1.87	89.00	2.47	3.56	19.00	11.21	38	8.5	1.030
15	12.40	17.00	1.76	92.00	2.62	3.98	33.00	15.86	39	8	1.029
16	9.99	19.00	1.82	101.24	3.42	4.93	45.00	16.41	41	7.5	1.031
17	11.23	23.00	1.88	156.45	2.46	4.02	47.50	14.20	42	7	1.032
18	11.35	14.00	1.73	65.00	3.28	3.87	41.00	28.20	39	8.5	1.029
19	10.98	15.00	1.45	53.00	3.74	3.46	34.00	26.88	41	8	1.034
20	10.96	21.00	1.33	90.00	2.83	4.25	42.00	14.60	39	9	1.031
μ	10.96	15.85	1.77	98.58	2.48	3.99	34.05	17.33	38.95	8.05	1.031
dp	0.63	12.55	0.24	38.50	1.28	0.75	10.67	8.35	2.42	0.65	0.0043
cv	5.77	79.15	13.41	39.05	51.61	18.79	31.33	48.15	6.21	8.04	0.42
Total[c] (%)					10/20 (50%)			1/20 (5%)			

Obs: Observation; Ca S: Serum Calcium; Ca U: Urine Calcium; Cr S: Serum Creatinine; Cr U: Urine Creatinine; DRCr-Ca (%): Percentage of renal clearance for Ca; PS: Serum Phosphorus; PU: Urine Phosphorus; DRCr-P (%): Percentage of renal clearance for P; Ht: Hematocrit; D: Urine Density (g/cm^3) M: Mean; DP: Standart Deviation; CV: Coefficient of Variation.

[a]Mule (*Equus assinus* x *Equus caballus*) - the animal was being subjected to heavy work.

[b]Mare (*Eqqus caballus*) with proteinuria; [c]Total (%) normal values for DRCr- (%)

For the Calcium, the mean was 3.68 g Ca/kg (sd: ±0.07; cv: 18:26%) and 2.40 g variation of Ca/kg and 4.7 g Ca/kg DM in the *B. humidicola*. For Phosphorus, the mean was 0.74 g P/g (sd: ±0.018; cv: 24.69%) and range from 0.40 g P/kg to 1.1 g P/kg DM in the *B. humidicola*. The levels of oxalic acid ranged from 1.15-3.10% of DM (sd: ±0.61; cv: 31.19%) *B. humidicola* and 4% of MS Elephant grass purple accessions (*Pennisetum purpureum*). The high Ca/oxalate ratio was 0.37 (*B. humidicola*) and the lower of 0.08 Ca/oxalate (Elephant grass accessions purple) and the mean was 0.20 (sd: ±0067; cv = 33.38%). For concentrations greater than 0.5% of oxalic acid in DM of the forage, the lower the ratio of Ca: oxalate increased the risk of bone problems (Table 1). All forage samples, presented satisfactory concentrations of Ca in grasses and in contrast only 10% showed levels of P satisfactory. The higher Ca/oxalate was 0.37 (*B. humidicola*) and the lowest of 0.08 (Elephant grass cession purple) Ca/oxalate and the average was 0.20 (sd: ±0067; cv: 33.38%). In the properties of the cities studied, who presented horses with a problem in the skeleton or multiple osteopathies, urine collections were performed for determination of Ca, P and Cr and total blood for determination of Ca, P and Cr in serum. These values were interrelated in the formula for DR-% Ca and P calculation (Table 2).

The calcium levels ranged from 9.88 mg to 12:40 Ca mg/dL in serum and the average was 10.96 mg Ca/dL (sd: ±0.63). Phosphorus content ranged from 2.12-5.30 mg Ca/dL in serum and the average was 3.99 mg P/dL (sd: ±0.75). The largest amounts were excreted 58 mg of Ca and P 47.50 mg/dL in the urine and the lowest was 4 mg Ca and 3.50 mg P/dL of urine. The clearance of ratio, ranged from 0.63-6.28% (µ: 2:48; sd: ±1.28; cv: 51.61%) for DR-% Ca, while the% DR-P ranged from 1.23-35.92% (µ: 17:33; sd: ±8.35; cv: 48.15%) (Table 2). For the total, 50% of values of % DRCr-Ca and only 5% of values of %DRCr-P were considered next normality (Table 2).

DISCUSSION

The dynamics of nutrients like protein, carbohydrates, fat, minerals and vitamins, represented by the ingestion, digestion, absorption, metabolic route and bone reabsorption or from the renal filtrate, renal excretion or secretion endogenous denote the complexity of physiology of nutrition, hormonal interactions and the importance of dietary balance, in all nutrients and the importance of dietary balance, in all nutrients, viewed the possible interrelations, the example of Ca and P. The organic matrix of bone in the horse's skeleton is adapted to the process of mineral absorption of Ca and P

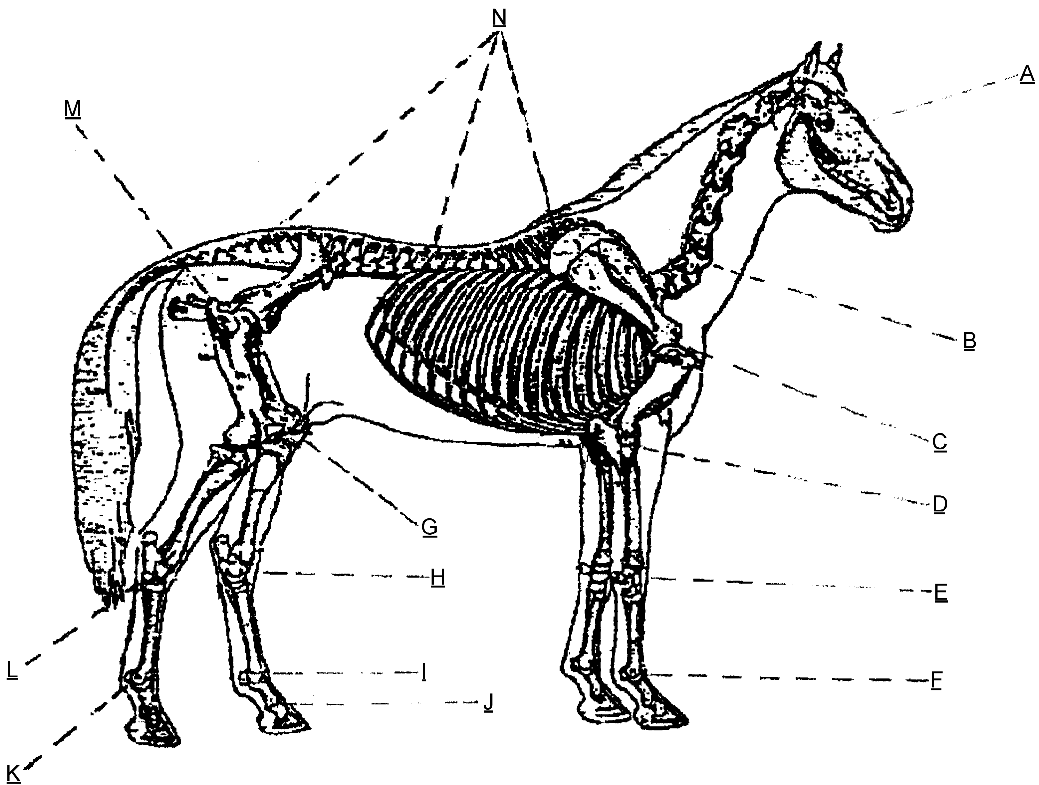

Fig. 1: Frequencies of bone disorders and evaluate of the impact on skeleton and of osteo-articular lesions with clinical manifestations in the skeleton of horses.

I- Osteochondrosis: Group of disorders involving one or more centers of ossification of the bones: A-Facial bones: mandibles, maxillae and nasal bones. Swollen face or big-head or also called fibrous osteodystrophy; B-Cervical vertebrae; C-Scapular-humeral joint or shoulder; D-Proximal radius; G-Tibio-femoral articulation or stifle; H-Distal tarsal joints or hock; M-Joint femur ileum or hip joint; N-Vertebrae spine thoracic-lumbar-sacral.

II- Physitis (epiphysis): Group of disorders involving one or more centers of ossification of the epiphyseal plates (growth) of the articular extremities: E-Distal radius; F-Distal metacarpus; I-Distal metatarsus; J-Rear pastern joint; K-Distal tarsal joint; L-Distal tibia

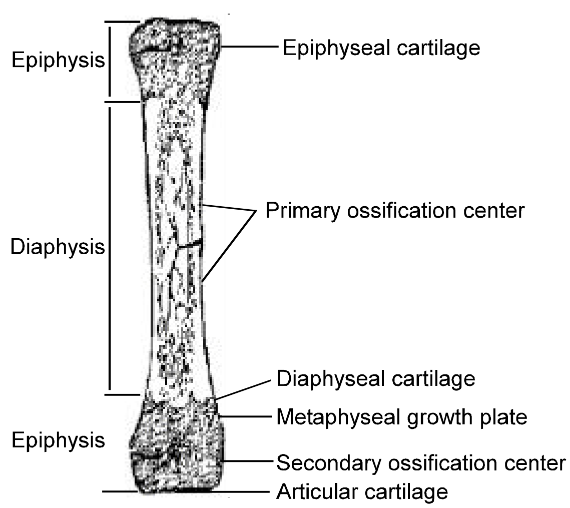

Fig. 2: Main sites predisposed at lesions in developing of growing animals

reabsorption of bone as necessary to maintain homeostasis. Since for monogastric herbivores, forages are the main source, if not the sole source of all nutrients essential to its maintenance, growth, reproduction, lactation and work. Forage available for grazing horses, presented as a general rule, the need for supplementation or correction of the appropriate levels of one or more nutrients (Tisserand, 1986; Coffman, 1980; Caple *et al.*, 1982a,b).

The Ca: P ratio should be approximately 2:1 (around of 1.8:1 for growing foals, greater than 2.1 for lactating mares and horses for sport or work heavy). Occurs, that the bones also require calcium, Occurs, that the bones also require calcium, being this that gives consistency to the bones. The first bones to suffer with the removal of Ca are the bones of the face. When calcium is removed from the facial bone, bone tissue needs to be replaced and there is a proliferation of chondrocytes cells without calcium were filled with fibrous tissue at the site where

Fig. 3: A: procedure for obtaining urine through the collection with stainless steel probe directly into the urinary bladder of the mare; B: Mare with foal (with angle defect of the anterior forelegs) at her second pregnancy and breast-feeding after pregnancy presenting successive fibrous osteodystrophy and photosensitization (burning) in parts visible by the white *Pithomyces chartarum*; C: Horse of three years of age presenting epiphysitis (osteo articular lesion) in the distal radius; D, F and H: Reproduction mares with foals on breast-feeding raised on pasture feeding of *B. humidicola* presenting unbalanced diet in macroelements Ca and P microminerals and growing animals with symptoms of bone disorders (laminitis, epiphysitis, osteochondrosis), angulation deformities and limb flexion; G and J: Mare and mule used in traction (heavy work) with advanced process of fibrous osteodystrophy, osteochondrosis and epiphysitis; I: Mare after the third pregnancy presenting laminitis, epiphysitis, osteochondrosis, bone fragility and glomerulonephritis due to prolonged use of calcium-deficient diet with excess P-grain cereals (corn, oats) and forages low quality

Fig. 4: Observations of macroscopic lesions of the facial bones of horses with chronic process of osteopathies: A-B-C-D-E-F-I-J: Represent the view of the dorsal and lateral sides of the mandible of horses which died after the chronicity of nutritional secondary hyperparathyroidism and glomerulonephritis; G: Right and left ventral edge of mandible with bone fibrous osteodystrophy and dorsal surface of the same mandible; A-B-H-K-L-M-N-P-Q-R-S-T-U-V: Showed fibrous osteodystrophy of the joints and removal of teeth (Gomphosis) and loss of molar teeth and several fractures. Enlargement of the bone, cancellous bone, fractures, detachment, loosening and fall of the molars, complicated with implantation periodontal disease

Table 3: Composition of diets and formulations based on calcium and phosphorus diets of farms studied

		Ca and P ratio		
Period	Equids sampled	Ca normal and P higher $\geq$2.1 Ca: >1 P (P: 1.5% to >3.0%)	Ca lower and P normal <2.1 Ca: 1 P (Ca: 0.5%-0.8%)	Ca lower and P higher <2.1Ca: >1P (0.5-0.8% Ca: >3.5% P)
1984-1989	128	70	1	057
1990-2005	101	43	0	058
Total (%)	229	113 (49.34%)	1 (0.44%)	115 (50.22%)

it was bone. This tissue has a larger volume, giving the appearance that the horse is with a swollen face (Joyce *et al*., 1971; Hintz and Schryver, 1976; Van Doorn *et al*., 2004).

In this research, considering averages above 2.1 g% Ca and averages next to 1.0 g% P as satisfactory for forage of equids, all samples showed unsatisfactory levels of Ca and only 10% of samples of forage of property were satisfactory for the levels of P (Table 1). However, considering that on average only 60% of the diet consists of forage and another 40% on average is obtained by supplementation of diet-based cereal grains, can be inferred that reside in the supplementation, the opportunity to correct or balance the diet in all nutrients, especially in minerals Ca and P. On the other hand, percentages ranged from 35-39% for Ca, 14-17% for P and 0.32-0.85 for Mg in the ash of bone from horses aged 1 day to 33 years. Values did not indicate a relationship between age and percentage of these elements in the ash and the value of the sum of the three cations was fairly constant. Ratio of calcium to phosphorus ranged from 2.1-2.6 with no relationship to age observed. A positive correlation was found between bone ash expressed as percentage of the dry, fat-free bone and age of the horses, up to 7 years. The bone ash values found in this study, ranging from 50 to 65% for horses of all ages (El Shorafa *et al*., 1979). These data above confirmed the that if proposed to present research, ie, correct the Ca: P ratio in all diets through supplementation of the ration end with concentrated balanced with the minerals mainly Ca and P.

To the other macro mineral, Phosphorus, the endogenous phosphorus loss in mature horses has been estimated at 10 mg/kg of BW/d. Assuming an average absorption of 45% of the phosphorus in feedstuffs, a pony requires about 21 mg of phosphorus/kilogram body weight per day to balance this loss. A similar estimate is obtained from the regression analysis of retention on intake which predicts that retention is zero when intake is 18 mg of phosphorus/kg BW/d. These estimates of the maintenance requirement are less than the 32 mg/kg BW/d recommended for ponies or 27 mg for mature light horses. The endogenous fecal phosphorus excretion did not vary with intake. This finding contrasts with that in ruminants and in rats in which fecal endogenous phosphorus has been shown to depend on dietary phosphorus. The endogenous fecal excretion of P may be an important path way in the maintenance of P homeostasis in ruminants. Unlike the horse, the urinary excretion of P is always very small in ruminants and is little affected by diet. The endogenous fecal excretion of P of rats was affected by both Ca and P intake (Schryver *et al.*, 1971a,b).

The kidney plays a distinct role in the homeostasis of Ca and P in the equine species. Schryver *et al.* (1970a) and Whitlock *et al.* (1970) asserted that the true digestibility of P had a mean 45% when administered in diet at maintenance level. However, the absorption of dietary P can vary 30-50% and endogenous fecal loss has had an average of 10 mg/kg BW/d (Schryver *et al.*, 1971b; Cymbaluk and Christensen, 1986) and is relatively constant over wide variation in intake of these minerals (Schryver *et al.*, 1974a,b). Unlike ruminants, the urinary excretion of Ca (Schryver *et al.*, 1970a; 1974b) and P is always significant in horses and is affected by diet (Schryver *et al.*, 1971b). In the present research, similar observations to of authors above were confirmed by measurements of %DRCr-Ca and P. All diets were imbalanced in Ca and P, except one in Ca. The values of %DRCr-P showed high levels of dietary P requiring correction of balance Ca:P. Other authors have pointed out the importance of absorption, metabolism and endogenous excretion and renal Ca and P (Gans and Mercer, 1984).

Thus, according to Schryver *et al.* (1971b; 1974a,b), renal endogenous excretion increased from 1.5 mg (0.20% P- requiring maintenance) to 42 mg/kg BW/d with high phosphorus (1.19% P) diet, suggesting linearity increasing P excretion in the urine due to the increase in consumption (Cymbaluk and Christensen, 1986; Schryver *et al.*, 1972). Therefore, if horses with high dietary P absorb and excrete higher levels of phosphate (PO^-_4) in urine. In these cases, determining the rate of clearance of creatinine (Cr) to PO^-_4 (%DRCr-P) according to Traver *et al.* (1976; 1977); Coffman (1980); Caple *et al.* (1982a,b); Mason *et al.* (1988), is considered a strategic tool for early detection of ionic imbalance of P, by permitting, diagnose and correct the problem before irreversible damage occurs to the skeletal tissue (Knight *et al.*, 1985).

In HPTNS, Joyce *et al.* (1971) characterized the disease, fibrous osteodystrophy, as being bilaterally symmetrical, firm, pyramidal enlargement of facial bones immediately above and anterior to the facial crests by thickening of the horizontal rami of the mandible and transitory lameness of 1 or more limbs detected in seven horses (Ca:P ratio of 1:10.5). Low Ca (ranging of 4.2 mg to 10.8 mg Ca/dL) and high P concentrations occurred in the urine (ranging of 116 mg a 224 mg P/dL) of these horses and PTH is unable to maintain Ca homeostasis because of the continued high intake of P. Serum P ranged of 1.4 mg to 7.8 mg P/dL. Occurs as a result of focal periosteal avulsion, torn or detached ligaments or tendons or subephyseal microfractures. Avulsion of ligaments may occur during work. Later in more severe cases the teeth become loosened, both the upper and lower jaws become enlarged and affected equids gradually become cachectic because mastication in painful and difficult. Spontaneous fractures occurs commonly. Installs if periodontal disease (Camargo, 1981; Nazario *et al.*, 1986; Sousa *et al.*, 1986). Normal values as reported by one investigator for Ca and PO^-_4 % DRCr-, were proposed: Ca: > 2.5% and PO_4 < 4.0% (Caple *et al.*, 1982a); Ca: > 3.5% and PO_4: < 2.52% (Carvalho *et al.*, 2008a,b) and PO_4: 0-0.5% (Traver *et al.*, 1976; Coffman, 1980). In the present study, all animals studied showed abnormal values for %DRCr-Ca and P, when observed the readings of both in the same animal. Similar results to the present research were also presented by Nordin (1958) that evaluated the primary and secondary HPTNS found regression in 95% of samples with normal serum 2.5 mg P/dL, P excretion was low and the% CR-P was 0.2% (Rawlings and Bisgard, 1975). HPTNS documented that may occur in vitamin D levels, evidencing the symmetric enlargement of the parathyroid glands in renal glomerular failure in chronic process; Mason *et al.* (1988) proposed the diagnostic, treatment and prevention of HPTNS thoroughbred in race horses in Hong Kong. Animals showed compression, fractures of the vertebrae T13, L4, L6 and of the sacrum. They observed that the fibrous osteodystrophy was resultant, when soft and fibro-osseous tissues were replaces the previously mineralized areas of bone tissue, leaving a weakened skeleton. Mentioned, still, that the imbalance (diets with excess P) results in increases levels of calcium reserve being excreted in urine with the P. The osteodystrophies produced at the beginning of HPTNS, it is difficult to detect and manifests clinically as a lameness or as sore areas at the insertion lines of ligaments and tendons. In these cases, the DRCr%-P was higher than 20% when the C: P ratio was 1:4.3 and %-P DRCr of more than 10% at the C: P ratio of 1:2.5. The authors have used the rate C: P ratio of 6:1 to treat animals with osteopathies. However, they recommended Ca: P ratio of 2.2-2.5:1 and Caple *et al.* (1982b); Nazario *et al.* (1986) mentioned that some centers in Australia, where there was high incidence of fibrous osteodystrophy, the diet was composed of approximately 80-90% by weight grain, to

10-20% by weight of chaff. They inferred that, considering the normal range of %DRCr-Ca >2.5 and DRCr%-P <4.0, among the 229 horses analyzed, 60% of the horses sampled had adequade Ca intake, 44% had excessive intakes of P and 25% of the horses were excreting more P in urine than Ca, indicating that these animals were subject to HPTNS (Table 3; Fig. 3 and 4). Was conducted, in present research the comparing of the clearance ratio, with the Ca:P ration end and clinical signs obtained from the farms, according to that described by Knight *et al.* (1985), in which the conditions referred to the metabolic bone disease include from a disturbance in the change of the skeleton into functional bone. Clinical manifestations include the phyistis, osteochondrosis, juvenile osteoarthritis and limbs deformities, contracted tendons, epiphysitis. The increases in protein, Ca and P levels that have been recognized as necessary in the diets of yong, growing horses may be inducing deficiencies in several trace minerals. Diets containing higher levels of calcium and phosphorus have been shown to interfere with the absorption of copper and zinc. Soybeans meal, commonly used as the protein source in corn-oat-based diets, contains phytic acid, a phosphorus-containing compound which forms insoluble complexes with zinc and several other minerals (Robinson and Slade, 1974). Still, the results of present research, agrees with the demonstrated by the authors Krook (1968); Joyce *et al.* (1971); Mason *et al.* (1988); Traver *et al.* (1976) to inferred that skeletal abnormalities are a prominent feature of Ca, P, Cu, Mn and Zn deficiencies in chicks, pigs and cattle. These skeletal changes include a decrease in osteoblastic activity and chondrogenesis broadened epiphyseal cartilage, limb deformities, lameness, enlarged joints, contracted tendons and ataxia. Similar bone and joint problems are recognized in the horses. There has been very little documentation in horses of the role of trace minerals in the development of the various manifestions. It appears likely that calcium, phosphorus, copper and zinc deficiencies are involved in the development of epiphysitis, contracted tendons and Osteochondrosis dissecans. Bone fragility by mineral imbalance of Ca and P, may predispose to accentuation of the severity of the process of kyphosis or lordosis.

The periodontal diseases implant if given the inflammatory process results in destruction of the attachment apparatus, loss of supporting alveolar bone and if untreated, tooth loss. Periodontal disease is one of the most common diseases of the oral cavity and is the major cause of tooth loss in adults. Recently, there has been increasing interest in the relationship of periodontal disease to important systemic diseases, such as cardiovascular disease and complications in pregnancy (Krook and Lowe, 1964; Krook, 1968; Joyce *et al.*, 1971).

In the present research, similar observations were made and showed the severity of the clinical pictures presented by animals in advanced stages of carential and metabolic disease (Fig. 3 and 4) irreversible. All the analysis showed if agreement with the values considered abnormal presented by Caple *et al.* (1982a,b), have proposed that adequate calcium intake exists in horses excreting more than 15 μmol calcium/ mosm (0.6 mg/L, 6 mg Ca/100 mL) of urine solute when calcium:creatinine clearance ratio (%DRCr-Ca) is greater than 2.5% and by Carvalho *et al.* (2008a) that found similar values normal ranging from 5.27% (37.57 mg Ca/dL) to 10.14% (47.20mg Ca/dL) for %DR- Ca.

Many of the consequences are manifested in the signs of deficiency or excess of dietary inadequate dietary phosphorus will, like inadequate calcium and vitamin D, produce rachitic changes in growing horses and osteomalacic changes in mature horses. As indicated, excess phosphorus reduces the rate of calcium absorption and leads to chronic calcium deficiency and HPTNS, also known as big head disease, bran disease, osteofibrosis and fibrous osteodystrophy (Hintz, 1996). HPTNS is characterized by a shifting lameness and in advanced cases by enlargement of the upper and lower jaws and facial crest (Krook and Lowe, 1964). Identical findings were presented in the present research in Fig. 3 and 4.

Serum inorganic phosphorus may be more indicative of dietary phosphorus status, than serum calcium is of calcium status because the homeostatic mechanisms for phosphorus are less sensitive than for Ca (Schryver *et al.*, 1970a,b; 1971a,b). Caple *et al.* (1982a) determined that horses excreting more than 15 μmol of P/mosmole (4.65 mg P/dL) of urine solute and having a phosphorus: %DRCr-P ratio greater than 4 had excessive phosphorus intake and were subject to HPTNS. The NRC (1980) suggested that a maximum tolerable level of dietary phosphorus in horses fed adequate dietary calcium is 1%. Similarly, Carvalho *et al.* (2008b) found similar values ranging from 1.01% (1.38 mg P/dL) to 1.57% (4.18 mg Ca/dL) for %DR-P. So like at present research, all animals presented a mean of P in urine greater than that described in the normality of the diet, except animal number 12 (Table 2) had lower average of 3.50 mg P/dL of urine, %DRCr-P equal 1.23%, meaning that the diet was unbalanced in P and indicating the need for intervention with correction of the diet through chemical analysis of its constituents.

The monogastric herbivore - equids - present restrictions for variations in Ca and P in the diet. The pastures provide part of the essential nutrients and other party must be provided in the form of supplements. However, if the pasture is rich in compounds which complex calcium, or presents fungi hepatogenous (hepatotoxic), the example of Phytomyces chartarum, this must be considered in the formulation supplementary. In this research, Pastures *B. humidicola* in the region center-west, presented variable content of 2.4-4.0 g Ca/kg DM and 0.5 g to 1.1 g P/kg DM of forage, denoting insufficient middle so much Ca as P. These pastures also had high levels ranging from 1.0-2.0% of oxalic acid (average of 1.5%) in DM of the forage. Several of these pastures also showed the presence of the fungus *Pitomyces chartarum*.

Unlike ruminants, in which the main route of excretion of P is endogenous and shows a wide variation depending of the amount ingested, absorbed and metabolized in horses endogenous excretion of P is constant and is about 10 mg/kg BW determined experimentally in ponies 200 kg live weight by Schryver *et al.* (1971a) and Hintz and Schryver (1972) with radioactively labeled P^{32}. It has suggested that earlier recommendations were inadequate or excessive. In homeostasis, the P are involved tissues (organs) of the intestines that are responsible for the absorption and endogenous secretion the skeletal tissues which respond by stock of P combined when in the form of Ca salts apatite and hydroxyapatite and are the main reservoir of the body easily adjustable by hormonal mechanisms for deposition and removal of salts in the form of phosphate. Finally, the kidneys are responsible mainly reabsorption of glomerular filtrate P or excretion of excess absorbed and not metabolized. Still, in the metabolism of P exert important regulatory activity the parathyroid hormones, calcitonin and vitamin D_3. Calcium in horse showed according to experiments with ponies 200 kg of live weight (BW) performed by Schryver *et al.* (1970a) and Hintz and Schryver (1972) endogenous fecal excretion constant 20 mg/kg BW. In the present research, behavior similar for the %DRCr-P described by the authors above, for the metabolism of P was observed. Diet with excessive P, directly reflected in increased absorption and renal excretion.

Quality forages will usually provide adequate Ca and P for maintenance. Ca and P ratios are important for horses. Ca and P ratios of 1.5:1 or 2:1 in the total diet are recommended for most classes of horses and should never be below 1:1. The NRC (1989) states that if P is adequate Ca:P ratios can be as high as 5:1. Recent research suggests Ca:P ratios should not be above 2.5 for growing horses. Supplementation of grass hay diet should be done with equal Ca:P or with more Ca than P. Diets can be balanced by including proper supplementation in the grain portion of the diet. Grains are high in P and low in Ca so they must be appropriately supplemented as needed. Offering a free-choice mineral containing Ca and P may help meet requirements but horses cannot be counted on to consume needed quantities of supplements. Using oats or other grains or add to commercial diets is not recommended because it may alter Ca:P ratios and dilute mineral and vitamins. There are feeds designed to be mixed with oats and these are more appropriate choices. Trace mineral requirements are usually met by feeding good quality hay and free-choice trace mineral salt. However, there are some deficiencies in forages that require close attention. Selenium and copper are generally deficient and adequate supplementation is necessary. In the present research, all the results of %DRCr- indicated that the animals' diet, was imbalanced in Ca and P, requiring correction of mineral supplementation. Attended the proportion recommended by the NRCs (1989, 2007), the values of %DRCr- returned to normal after 48 h for Ca and 72-120 h for P which is in agreement with Caple *et al.* (1982a,b) and Carvalho *et al.* (2008a,b).

Bioavailability: In the present research, one of the ways in against-proof, for the quality of nutrition in Ca and P provided by or pasture or hay is the clinical observation of animals. Any, clinical manifestation or signs of imbalance of mineral dietary Ca and P can be observed by the pathognomonic signs of nutritional disorder. In the face swollen disease or fibrous osteodystrophy, the animal shows swelling facial visible to the observer. Animals subjected to pregnancy, or gestation and lactation are very susceptible, as well as animals in stages of rapid growth are also likely pathophysiology of nutrition (Fig. 1, 2 and 3). The efficiency of absorption was decreased, if the Ca intake was excessive (Schryver *et al.*, 1970a). Source of Ca, Oxalate and Ca:P ratio could also influence efficiency (Whitlock *et al.*, 1970).

Some mineral complex may be present in some species of forage used by grazing horses. In pastures of *B. humidicola*, there may be increasing levels of oxalate which complex calcium in the plant and bind calcium in the form of calcium oxalate is not available to the animal metabolism. Moreover, the free oxalate in forage may complexing Ca feed in the digestive tract, or if absorbed may complexing Ca in the bloodstream forming insoluble salts that can deposit in renal glomeruli. Levels greater than 0.5% of Dry Matter (DM) of forage can reflect problems to metabolism of Ca. Agreeing with that observed in the present research, Mckenzie *et al.* (1981a,b) investigated the effect dietary containing 2.6% or 4.3% total oxalate on Ca and P balances in horses. The urinary loss of Ca was reduced and that of P increased. The main Ca and P loss was in faeces. The HPTNS or fibrous osteodystrophy is a disease of stabled horses fed diets high in P and low in Ca (high grain or bran component). However, various species of tropical grasses have their full ion Ca bound to anion oxalate $\{[Ca^{++} (COO^-)_2 \leftrightarrow Ca^{+2} + 2COO^-]\}$, ion Ca^{+2} should present themselves dissociated, however, Ca was unavailable for absorption by the horses and thereby produced the disease. Still, observations identic to the present research were reported by Mckenzie *et al.* (1981a,b) analyzed Horses grazing manily *Cenchrus ciliaris* and or *Panicum maximum* pastures on over 30 properties in southern central Queensland developed lesions of osteodystrophia fibrosa. Fibrous osteodystrophy: characterized by osteoclastic bone resorption and intertrabecular fibrosis with unmineralized trabecular bone containing a large amount of unmineralized osteoid. There was also hypertrophy and hyperplasia of the parathyroid glands which is consistent with HPTNS. Horses on individual properties in coastal Queensland grazing Setaria anceps, *Brachiaria mutica* or *Pennisetum clandestinum* also developed the disease. Lameness and fibrous swellings of nasal bones, maxillae and mandibles were observed. Ca and P levels of pasture were normal but all the above pasture species contained oxalates which were suspected of causing the disease. The authors mentioned that the control of HPTNS, in grazing horses with calcium plus P supplementation. That was concluded pasture hazardous during this time. Total oxalate presented content of above 0.50% and Ca: Oxalate of ratio below 0.5%. The supplementation of horses with limestone plus dicalcium phosphate resulted in some recurrences but the addition of rock phosphate in molasses for previous mixture corrected the problem.

Large ruminants have the ability to detoxify oxalates by converting them to carbonates and bicarbonates in the rumen. Walthall and Mckenzie (1976); Swartzman *et al.* (1978) and Mckenzie *et al.* (1981a,b) studying oxalate nephropathy in a horse report that although the horses are relatively resistant at least the oxalates nephropathy caused by Na and K. However, describe a case of oxalate poisoning with subsequent nephropathy in a horse.

The equids has selective grazing habits for the most tender parts of plants more palatable and more protein and therefore of higher oxalate content. In rainy months plants synthesize more protein and therefore also more oxalate. Thus, results identical to of this research, were also reported in studies of Carvalho *et al.* (1988; 1990), Swartzman *et al.* (1978) for the months January to May the percentage of oxalate ranged from 0.93-1.97% in horses grazing in the central region west of the state of Sao Paulo. The elephant grass used as supplement in the diet had an average of 4% of oxalate in dry matter. In pastures, the relationship Ca:Oxalate ranged from 0.30-0.39. Ca varied from 0.36-0.60% in those grassland studied. The appearance of bone disorders associated with the imbalance of nutrients minerals, among them the relationship of mineral Ca: P can be accelerated and as an additive factor, the presence of oxalate which complex with Ca sometimes in the absorption, sometimes in the bloodstream and leads to the appearance of pathologies of skeleton such as the osteodystrophy, epiphysitis, arthritis and others that are often of degree irreversible of process (Pupo, 1984).

However, given the possible variations in the utilization of total digestible nutrients, diet, unfavorable results of clinical evaluations should be interpreted taking into account the possible existence of complexation with Ca, phytates and oxalates in the feed formulation based on true digestibility of the ingredients (Swartzman *et al.*, 1978; Mckenzie *et al.*, 1981a,b; Hintz and Schryver, 1972; Carvalho *et al.*, 1988; 1990). In the present study, were performed the anamneses of animals in properties where the animals showed pictures of osteopathies, the analyze of the ingredients minerals of forage and complete feeds of rations oxalate, urinalysis, %DR-Ca and P (Fig. 1-3).

Still agreeing with the above mentioned, in the present study, determined the total oxalic acid in forages were greater than three to four times the amount of 0.50% DM in the grass. Oxalic acid levels above 0.50% in the diet may bind Ca in the plant, interfering with absorption of calcium by forming insoluble salts by complexing with Ca or being absorbed bind to the Ca blood (Swartzman *et al.*, 1978). Given the presence of antinutritional factors and possible adverse interactions or antagonisms unfavorable macro and micro minerals has led authors (Mckenzie *et al.*, 1981a,b; Caple *et al.*, 1982a,b; Mason *et al.*, 1988), the utilize in addition chemical analysis of foods, the assessment of renal function by determination of creatinine clearance for the Ca ion (%DRCr-Ca) as a complementary diagnostic biochemical test capable of assessing the status of the metabolism of Ca (Table 2).

The technique used by several authors was based on work carried out by Knudsen (1959) that by determining the %Cr filtered in parallel to DRCr-ion electrolyte in question (%DRCr-X), was considered of diagnostic value, with the assertion 95%. In the specific case of Ca ion, the standardization of the diet, ie, the quantification of Ca in the final diet, taken together the results of %DRCr-Ca, in present research, allowed interpretation of results and comparative analysis the values of literature.

In the regulation of homeostasis of Ca and P under the influence of parathormone and vitamin D_3, the digestive system, kidneys and skeleton play physiological mechanisms primordial (Schryver *et al.*, 1971a,b). The efficiency of intestinal absorption, increases even during periods of greater intensity in bone mineralization, such as growth and pregnancy and also during lactation. With relation to the absorption of phosphate in horses, the percentage absorbed in the diet is relatively constant and depends mainly of the source. Basically, the soluble phosphates are more easily absorbed and can estimate the absorption of P (around of 33%) in mean the variation of 29-35%, concentrates of 44-46% (around of 45%) in feed and as general average 58% (and around 55% in mineral supplements of good quality) in the best minerals supplements (Hintz *et al*., 1973; Schryver *et al*., 1972; Hintz and Schryver, 1976).

The kidney filters uninterruptedly, a large amount of calcium non-bonded proteins, reabsorbing around of 98%. Also reabsorb 80-97% of filtered phosphorus. When the plasma level of P increases, the filtration and reabsorption also increased, however, the mechanisms of reabsorption are rapidly saturated and excretion at that time increases in proportion to the filtered phosphate (Gans and Mercer, 1984).

The skeleton functionate with a reservoir potential for Ca and P and under hormonal regulation makes it available for the maintenance of normal blood levels of these elements. Bone formation and mineralization, as well as the reabsorption depends on the plasma concentration of calcium and phosphorus. In turn, the skeleton is the major reservoir of calcium and phosphorus to maintain normal levels of these minerals in the blood. The long bones, if they develop from cartilage by a process of endochondral ossification. In the fetus, the bone matrix is composed only of cartilage. Ossification centers develop in the center of the future long bone (diaphyseal region) and ends (epiphysis, distal and proximal epiphysis). With the onset of ossification, a bony epiphysis evolves each end and a bone diaphysis is developed there. Between these two centers of ossification, is the growth plate metaphyseal (also called the physis) which allows the elongation of the bone which accompanies growth after birth. At the appropriate moment, there is ossification of the physis. The closure of the physis of each bone, occurs at different times, as a rule generally may if say that the distal bones close before of the proximal bones. The hormonal regulation is done mainly through PTH, Calcitonin and Vitamin D_3 (cholecalciferol). Also participate indirectly estrogens, adrenocortical, thyroxine, glucagon and somatotropin interact in the regulation of plasma Ca and P. The PTH increases blood levels so that receives stimulus by declining levels of calcium in the blood. Vitamin D_3 increases Ca absorption from the gut by stimulating the decrease to normal levels in the bloodstream. Conversely, calcitonin produced in C cells, increases your thyroid levels when above-normal levels in the bloodstream are detected.

The maintenance of homeostasis of Ca^{+2} and PO^{-}_4 ions depend mainly on the intestinal tract, skeleton and kidneys. The liver as the central organ of metabolism is modulated by hormones and responds to physiological needs, synthesizing or resynthesized essential nutrients (Raisz and Kream, 1981; Wasserman, 1984).

The level of Ca and P in the blood of animals is very variable and wherefore it serves as a parameter for mineral status of these elements [(Ca±10.0 mg to 14.0 mg/dL and P±2.0 mg to 5.5 mg/dL); (Schryver *et al*., 1971b; Coffman, 1980)].

Phytates and oxalates if bind to cations, reducing their absorption. The grasses of tropical and subtropical grasslands may have a high level of oxalate reducing the absorption of Ca and may cause HPTNS (Walthall and Mckenzie, 1976). The P complexed in the form of phytate in cereal grains are responsible for the decrease in digestibility of P.

In disorders provoked by carential and metabolic disease of Ca and P, due to some peculiarities of the species, are more predisposed the lesion members in the equids which was observed in present research (Table 1; Fig. 1-3). Thus, the fore foot is composed of five toes (called phalanges) and the corresponding five proximal long bones forming the metatarsus. Similar to the fingers of the hand, the bones of the toes are called phalanges and the big toe has two phalanges while the other four toes have three phalanges. The joints between the phalanges are called interphalangeal and those between the metatarsus and phalanges are called metatarsophalangeal. Osteochondrosis is a developmental defect in the cartilage or of cartilage and bone seen in specific locations on the surface of the tarsocrural joint; Laminitis is the vascular disease, causing ischemia or homeostasis.

In present research, might if ray to the clinical frame of osteopathies may be defined by: epiphysitis - When growth cartilage is not replaced by bone and the vessels can't penetrate the area of growth, bone not if organizes, there is necrosis in this area and we can observe clinically enlargement at the site of the growth plate of distal radius bone and metacarpal and metatarsal distal to foals of up to two years; fibrous osteodystrophy - In adult animals with advanced disease, we can observe severe lesions, especially enlargement of the bones of the mandible and maxilla, a disorder known as swollen face disease (Fig. 1 to 4) and lameness evident. These signals appear due to the increase of osteoclastic activity of bone external lamellae and rupture of the tendon insertions and the trabecular bone that support the articular cartilage. The bones of the face, there is an excess deposition of connective tissue and osteoid by osteoblast stimulation, hence, the progressive expansion of the bone and osteochondrosis - It's a defect of osteochondral ossification which results in various manifestations, depending the place where she is it installs. It may come in the form of cystic lesions in the bone or as a defect in the articular cartilage (Fig. 1, 2 and 3: C-D-I). Agreeing with the described above, Hintz and Schryver (1976) mentioned that frequently, these lesions are painful and often only diagnosed by radiological examination or when there is manifestation of pain, particularly in the intensification of training. The nutritional imbalance especially related to the macro minerals is crucial for the appearance of these lesions and the reported by Schryver *et al*. (1971a,b) and Jordan *et al*. (1975) on the appearance of osteopathies in physical activity has a role relevant in the performance of young animals. The most common lesions are inflammatory reactions in the joints, small fractures and exostosis in response mainly to inadequate mineral nutrition. To prevent bone lesions or HPTNS, some nutritional use high levels of dietary Ca, however, levels above those recommended over-stimulate calcitonin and may interfere with the absorption of trace minerals such as zinc.

Besides of the imbalance of Ca and P diets observed in present research, other aggravating the absorption, metabolism and excretion of Ca and P were diagnosed for toxic levels of oxalate and the presence of *P. chartarum* in *B. humidicola*. About the toxic levels of oxalate, important studies were performed by Swartzman *et al*. (1978), Mckenzie *et al*. (1981a,b) and Carvalho *et al*. (1990) inferring that horses grazing mainly *Cenchrus ciliaris* and/or *Panicum maximum* var. trichoglume pastures on over 30 properties in southern central Queensland developed lesions of osteodystrophia fibrosa. Horses on individual properties in coastal Queensland grazing Setaria anceps, Brachiaria mutica or Pennisetum clandestinum also developed the disease. Ill-thrift, lameness and fibrous swellings of nasal bones, maxillae and mandibles were observed. Calcium and phosphorus levels of pasture were normal but all the above pasture species contained oxalates which were suspected of causing the disease and by Nunes *et al*. (1990a) and Nunes and da Silva (1998) reporting that the *B. humidicola*, originally from tropical forage continent african and adapted to our climate and soil conditions, often acidic and low fertility, very used as pastures equines farms. In spite of the good acceptance by equines has an impact on mineral metabolism of the animals demonstrated clinical and metabolic abnormalities of bone growth, weight loss and fractures. This forage grass presents unbalanced levels, the main nutrients such as proteins, lipids, vitamins and minerals, in addition, high fiber content (Nunes *et al*., 1990a,b).

Among the osteodystrophy, the most notorious is the swollen face or “big head”, characterized by a bilateral swelling of the facial bones. In deficiency in plasma Ca, parathyroid gland is stimulated to produce parathormone, which mobilizes Ca from skeletal reserves into the bloodstream. Spaces bone decalcified are replaced by fibrous scar tissue which leads to the increase in volume of the bones flat of skeleton, being more visible on the face. In the continuous stimulation of PTH by calcium deficiency, or excess P in the diet, appear to HPTNS (Camargo, 1981; Nazario *et al*., 1986). Once these changes are irreversible skeletal, alternatively, correction of the imbalance of the pasture can be accomplished through mineral supplementation orientated to the problem of animals on the property in study. If the pasture provides only 60% of requirements in MS (forage), the addition of 40% concentrate will return balanced mineral nutrients in deficiency based on body weight. Among the sources of Ca available, the most common are calcium carbonate, calcium chloride, tricalcium phosphate and oyster meal.

For concentrations higher than 0.5% of oxalic acid in DM of the forage, as lower the ratio of Ca:oxalate, higher the risk of bone problems. Nunes *et al*. (1990a) mentioned that *B. humidicola* showed 1.80% of oxalate and calcium/oxalate ratio equal 0.23 and others forages had values ranging from 1.30-2.80% and Ca: oxalate ratio ranging from 0:10 to 0:28. In this research, the values of levels of oxalic acid ranged from 1.15% to 3.10% of DM in the *B. humidicola* and 4% of MS Elephant grass purple accessions. The high Ca/oxalate ratio was 0.37 (*B. humidicola*) and the lower of 0.08 Ca/oxalate (elephant grass accessions purple) and the mean was 0.20 (sd: ±0067; cv = 33.38%). (Table 3). The ecotype of *B. humidicola*, is originally from Africa and was introduced in Colombia in 1978, has perfectly adapted climatically edaphic and shows acceptance by cattle and horses (Nunes and Silva, 1998). However, horses underwent grazing, showed disturbances of mineral metabolism, since this plant, besides presents low protein content, nutrient imbalance high levels of oxalates (Helene *et al*., 1989; Carvalho *et al*., 1990), besides being associated to *Pithomyces chartarum* (Malavolta *et al*., 1988), the causative agent of hepatic photosensitization (Schenk *et al*., 1991). Usually, the horses that make it permanent use, presented, delayed development, low work performance and affect the immune competence and cause metabolic disorders (Krook and Lowe, 1964; Joyce *et al*., 1971). We observed several cases of osteopathy, osteodystrophy as the "swollen face" in horses subjected to prolonged grazing (Gronendyk and Seawright, 1974; Mackenzie *et al*., 1981a,b).

Summarizing for results presented in this research, concluded if that the horses are very vulnerable when if say respect to predisposition for the removal of minerals from the skeletal reserves to maintain the homeostasis of Ca and P in tissue systems organic and particularly in the bloodstream. Any signaling, below normal levels in the bloodstream, parathormone is stimulated via cell receptors in the parathyroid gland, aiming to increase the absorption of Ca and P, the intestine, potentialized the action of vitamin D_3 and action on the kidney to the glomerular filtrate reabsorption of Ca^{++} and PO^{-}_{4}. In Ca-deficient diet or with an excess of P, the Parathyroid is continuously stimulated which may lead the HPTNS, the renal dysfunction (chronic interstitial nephritis and glomerulonephritis) and consequently the dysfunctions metabolic bone and articulars dependent of Ca and P. More than 50% of the animals examined had renal dysfunction as a result of toxic levels of oxalate in the diet.

At present research, 50% of horses grazing in *B. humidicola* presented the clinical picture, with multiple bone lesions and articular. In stages more advanced metabolic disease or deficiency the most visible lesions were fibrous osteodystrophy "swollen face" epiphysitis, osteochondrosis and lameness. These animals lost reproductive and of work potential, besides hampering the management of property.

In the reform of pasture grasses for horses of the genus *Panicum* sp. (*Panicum maximum*) were preferred. However, the coast cross (*Cynodon dactylon*, Linneau) has shown no toxicity problems have low levels of oxalate and absence, minimal incidence or nonexistence of *Pitomyces chartarum*.

In assessment of clearance for Ca and P ratio, 50% of equids presented values below the acceptable %DRCr-Ca and 95% of the equids presented values higher than the acceptable %DRCr-P.

The monitoring of diet, through the analysis of the feed ingredients and mineral analysis, mainly Ca and P end of diet, constituted if in practice of screening to be followed by biochemical %DRCr-Ca and P. Allied to these practices, the assessing the content of oxalate of the forages. Clinical examination of animals with application of semiotic veterinary techniques, allied the obtaining of anamneses of the animals, tools that were added the diagnostic accuracy in assessing the initial profile of the carential diseases or disorders of the bone in early of establishment.

In practice of determination of %DRCr-Ca and P together to chemical analysis when the properties presented the sum of forages (pasture) and supplementation rich in cereal grains with contents end, especially imbalanced in minerals in the diet and particularly of Ca and P, allowed to prevent in time the appearance of pictures subclinical or clinical trials initial of osteopathies, before that the bone pathologies if aggravate and if become irreversible.

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