Growth and Skeletal Development in Great Dane Pups Fed Different Levels of Protein Intake[1]

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ABSTRACT Feeding a dog of a large breed with a diet exceeding the National Research Council (1974) recommendations for energy, protein, calcium, phosphorus and vitamin D may result in disturbances of skeletal development. The effects of excess energy and various calcium:phosphorous ratios *per se* have been reported by others. The role of dietary protein, especially with regard to calcium metabolism and skeletal development, in large breed-dogs is reported in this article. Seventeen Great Dane pups, 7 wk of age, were divided into three groups. During 18 wk each group received isoenergetic dry food (~15 kJ metabolizable energy/g) containing 31.6, 23.1 or 14.6% protein on dry matter basis. No differences were found among the high (H-Pr), normal (N-Pr) and low protein (L-Pr) groups for the height at the shoulder. Significant differences were found between the H-Pr and L-Pr groups for body weight and plasma albumin and among all three groups for plasma urea. The differences in protein intake *per se* had no demonstrable consequences for calcium metabolism and skeletal development. A causative role for dietary protein in the development of osteochondrosis in dogs is unlikely. J. Nutr. 121: S107–S113, 1991.

INDEXING KEY WORDS:

• *symposium* • *dog* • *protein* • *calcium metabolism* • *growth*

In young dogs of large breeds, disturbances in enchondral ossification may lead to severe alterations in both articular and physeal cartilage, clinically known as osteochondrosis (OC) and resulting in severe lameness and skeletal deformities (1, 2). Diet composition plays an important role in enchondral ossification. Of the many possible variables in the diet, attention has thus far been focused on the influence of the total intake, energy and calcium-to-phosphorous ratio.

Hedhammer et al. (3) induced OC in Great Dane pups by feeding excess energy, protein, calcium, phosphorous and vitamin D. Excess energy per se in a balanced diet did not cause an increased incidence of skeletal abnormalities (4). By increasing only the calcium content of the diet, Hazewinkel et al. (5) found increased occurrence and severity of OC in Great Dane pups. Results of follow-up studies with various calcium and phosphorous intakes demonstrated that high calcium intake (independent of the ratio to phosphorous) is an important determinant of disturbances in enchondral ossification (6). Another important diet component, i.e., protein, has not yet been investigated as a single variable with regard to the skeletal development in large breeds of dogs.

There are at least two reasons to pursue this matter. First, from the multi-variable study of Hedhammer et al. (3) it was suggested that a high protein content in the diet contributed to the development of OC. Second, there is evidence from studies in humans and rats that protein excess influences calcium absorption, skeletal mineralization and calcium excretion (7–14). There have been no studies on the protein requirements in growing dogs of large breeds (15).

The present study was primarily designed to test the hypothesis that high protein intake plays a causative role in the pathogenesis of disturbed enchondral ossification. The second objective was to increase understanding of protein requirements for growth in large breeds of dogs. In this report, the clinical, routine laboratory, biochemical, radiographic and histological

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results, as well as results of calcium kinetic studies, in Great Dane pups fed different levels of protein will be presented.

MATERIALS AND METHODS

Animals. Seventeen Great Dane dogs (11 males, 6 females), 7 wk of age and originating from three litters, were randomly divided into three groups: a high protein group (H-Pr; $n = 6$), a low protein group (L-Pr; $n = 6$) and a control group (N-Pr; $n = 5$). In all groups the sexes were represented as equally as possible. At the end of the study, i.e., 27 wk of age, all dogs were killed for pathological investigation, by use of an intravenous overdose of sodium thiopental.

Housing. The dogs were housed in individual metabolism cages for 2 wk during each calcium kinetic study (W 1+2, W 7+8, W 13+14, and W 19+20). Between these 2-wk periods the animals were housed in individual cages, had access to an outside run and were allowed free exercise once a week for 4 h.

Diet and water. The dry diet was formulated to meet the recommendations of the U.S. National Research Council's Nutrient Requirements of Dogs (1974) (16). During the first 2 wk all dogs received the N-Pr food with 23.1% protein expressed on dry matter basis (% DM). From W 3 onward, dogs of the H-Pr group received diet with 31.6% protein, and those of the L-Pr group 14.6% protein (**Table 1**). The three diets were isoenergetic, with ~15 kJ metabolizable energy (ME) per g DM. This was achieved by exchanging carbohydrate for protein in the H-Pr diet and the reverse in the L-Pr diet, as compared with the N-Pr group (**Table 2**). The protein sources of the diets are given in **Table 3.** The protein of the diets used had a lysine content of 6 g/100 g of protein, calculated from standard reference values. The relatively low content of sulphur-containing amino acids in the protein-rich ingredients was compensated by the addition of methionine to the diet. Lysine was added to the ingredients of the L-Pr diet to maintain a lysine content of 6 g/100 g protein.

For restricted feeding, the daily amount offered, expressed as kJ (ME) per unit of metabolic body weight ($kJ/BW^{0.75}$), was decreased stepwise from 1500 at W 1–4 to 1200 at W 16–20. The nonconsumed food was weighed, and food intake was calculated. The proximate composition (Weende analysis) of the diet was determined in triplicate at the beginning and at the end of the experiment (Table 2). Dogs had free access to drinking water.

TABLE 1

Protein content of high (H-Pr), normal (N-Pr) and low (L-Pr) protein diets

Protein content[1]	H-Pr	N-Pr	L-Pr
% DM	31.6	23.1	14.6
g/1000 kJ ME	19.5	14.3	9.0
Pr:E ratio	0.29	0.21	0.13

[1] % DM; percentage of dry matter; ME, metabolizable energy; Pr:E ratio, protein energy to total energy ratio.

TABLE 2

Proximate (Weende) analysis of high (H-Pr), normal (N-Pr) and low (L-Pr) protein food

	H-Pr	N-Pr	L-Pr
Crude protein[1]	30	21	12.9
Crude fat[1]	9.7	9.9	9.7
Crude fiber[1]	2.8	3.0	3.1
N-free extract[1]	41.4	49.5	58.6
Ash[1]	6.1	6.6	5.7
Moisture[1]	10	10	10
Ca[2]	10	10	10
P[2]	9	9	9
Vitamins[3]			

[1] g/100 g.
[2] g/kg.
[3] Each kg of diet contained 5000 IU of retinyl acetate, 1000 IU of cholecalciferol and 50 IU of *dl*-α-tocopheryl acetate as guaranteed by the producer.

Physical examination. The dogs were observed twice daily at feeding time. The height at the shoulder was measured in all dogs once weekly. Body weight was recorded three times weekly at regular intervals, and a physical examination was performed once weekly.

Chemistry. Blood samples were collected once a week by jugular venapuncture with the dogs in sitting position after an overnight 9-h fast. This was done without prolonged occlusion of the vein. The following measurements (by the methods in parentheses) were carried out in blood, serum or plasma, as appropriate: packed cell volume (PCV); white blood cell count (WBC) (Sysmex system F800, Sysmex-ToA Medical Electronics Co. Ltd., Kobe, Japan) and differentiation; total protein (biuret); albumin (bromcresol-green); protein electrophoresis (cellulose acetate, staining by Ponceau S); total calcium (o-cresolphtalein); inorganic phosphate (molybdate without deproteinization); urea (urease glutamic dehydrogenase); creatinine (Jaffé method, initial rate at 30°C); alkaline phosphatase [(AP) EC 3.1.3.1] and alanine aminotransferase [(ALT) EC 2.6.1.2] (both kinetic according to International Federation of Clinical Chemistry recommendations at 30°C); γ-glutamyltransferase, γ-GT (EC 2.3.2.2) (kinetic, L-γ-glutamate-3-5-dibromo-4-hydroxyanilide, 30°C).

Serum calcium concentrations were adjusted using the formula: $calcium_{adjusted}$ = total plasma calcium (mmol/L) + 0.875 − 0.025 × (albumin concentration, g/L) (17).

Statistics. Differences between two groups were investigated with the Student's *t* test. One-way anal-

TABLE 3

Ingredients of the high (H-Pr), normal (N-Pr) and low (L-Pr) protein food

Ingredient	I.F.N.[1]	H-Pr	N-Pr	L-Pr
		g/100 g DM		
Bloodmeal	5-00-381	8.1	5.2	2.3
Casein (dehydrated)	5-01-162	7.8	4.8	2.2
Soyabean flour	5-04-593	13.6	8.7	3.8
Corn glutenmeal	5-02-900	8.0	5.0	2.2
Hominy feed	4-02-887	8.5	12.5	15.3
Potato starch	4-07-850	21.5	25.0	29.5
Wheat middlings	4-05-205	14.7	19	21.9
Sugar		2.5	5.0	7.5
Tallow	4-08-127	5.9	6.0	6.0
Soyabean oil	4-07-983	2.0	2.0	2.0
Cellulose		1.0	1.0	1.0
dl-Methionine		0.4	0.2	0.02
l-Lysine		—	—	0.13
Vitamin/mineral supplement[2]		6.0	6.0	6.0
Butylated hydroxy toluene		0.1	0.1	0.1

[1] I.F.N., International Feed Numbers.

[2] Vitamin, mineral and trace-element mix (6 g; Trouw, Putten, The Netherlands) contained the following: Vit A, 6250 IU (retinyl acetate); vit D_3, 100 IU (cholecalciferol); vit E, 6.9 IU (*dl*-α-tocopheryl acetate); vit C, 3.7 mg (ascorbic acid); thiamin, 0.7 mg; biotin, 21 μg; Vit B-12, 5 μg (cyano cobalamin); δ-panthotenic acid, 1.9 mg; nicotinic acid, 5 mg; folic acid, 0.12 mg; pyridoxine-HCl, 0.7 mg; menadione-sodium bisulfide, 0.25 mg; choline chloride, 83 mg; iron, 7.3 mg; copper, 0.5 mg; zinc, 10 mg; manganese, 4.5 mg; cobalt, 0.025 mg; iodine, 0.11 mg; selenium, 0.01 mg; sodium chloride, 0.5 g; limestone, 0.6 g; dicalciumphosphate, 2.75 g; potassium chloride, 0.6 g.

ysis of variance (ANOVA) was applied when three groups were involved. When significant, this analysis was followed by Student-Newman-Keuls multiple comparisons method. $P = 0.05$ was chosen as a level of significance.

Radiology. Mediolateral radiographs of the right radius and ulna were made with the dogs in right lateral recumbency, using a conventional diagnostic X-ray system (Maximus M-150, Philips NV, Eindhoven, The Netherlands), on regular black-and-white films (Cronex 4 DDS, DuPont de Nemours GmbH, Frankfurt, Federal Republic of Germany), in combination with high-detail intensifying screens (Cronex Detail Screens). Exposure settings ranged from 48 to 54 kVp and 10 to 16 mA, depending on size of the object, with a focus-film distance of 100 cm and table-top technique. Radiographs were made four times, at 6-wk intervals, i.e., W 3, 9, 15 and 21. These radiographs were used to assess the development of the ulnar styloid process and the anconeal process, the architecture of the distal ulnar metaphysis and the growth in length of the radius and ulna. The radial diaphyseal length was measured between the proximal and distal metaphyseal border through the axis of the bone (18). The length of the ulna was measured between the proximal border of the olecranon, adjacent to the apophyseal growth plate, and the epiphyseal border of the distal ulnar growth plate, through the axis of the bone. Measurements were made with a curved ruler, corrected for geometric magnification and expressed in centimeters. The average length (±SD) of the radius and ulna was calculated for each group of dogs at different ages.

Calcium kinetics. Calcium kinetic studies with ^{45}Ca were performed four times (W 1+2, W 7+8, W 13+14 and W 19+20) in all dogs, as described elsewhere (6).

Pathology. Costochondral junctions of the ninth ribs were obtained by surgical resection in W 3 (left rib, $n = 7$), W 9 (left rib, $n = 8$) and W 15 (right rib, $n = 17$). At necropsy the right proximal humerus and tibia, the right distal radius and ulna and the costochondral junction of the 10th rib were collected. After removal of soft tissues, 2-mm-thick midsagittal slices were cut from these bones. Tissue preparation for light microscopic studies was as described previously (19). Histomorphometric studies in bone tissue were done in undecalcified rib sections. The following determinations were made: total volume of bone (V%b), relative osteoblast-covered (Obs) and osteoclast-covered (Ocls) trabecular bone surface and the number of osteoclasts per microscopic field (Ocl; the mean of 15 fields, 25× objective) (20). Average width of the physeal growth plate of the rib was determined from the mean of five measurements at fixed spaced sites, using an ocular micrometer. Sections of heart, lung, liver, kidney, spleen, gut, thyroid, parathyroid, adrenal glands and cervical spinal cord were processed (19).

RESULTS

The mean daily food intake was not different among groups and decreased gradually from ~1400 kJ ME/$kg^{0.75}$ in W 1 to 1100 kJ ME/$kg^{0.75}$ in W 20 in all groups. Physical examination revealed no abnormalities. No significant differences were found for the height at the shoulder throughout the study. The increase in body weight was lower in the L-Pr dogs than in the H-Pr dogs throughout the study, although only statistically significant in the period of W 6–8 (**Fig. 1**).

After W 3, significant differences occurred in the H-Pr and L-Pr groups for plasma albumin (**Fig. 2**) and in all three groups for urea (**Fig. 3**). Linear regression demonstrated a significant increase in plasma total protein and creatinine concentration during the experiment for all groups. Differences between groups for plasma creatinine (**Fig. 4**) and total protein (**Fig. 5**) were not significant at any time. However, as already suggested by Figures 4 and 5, the mean differences

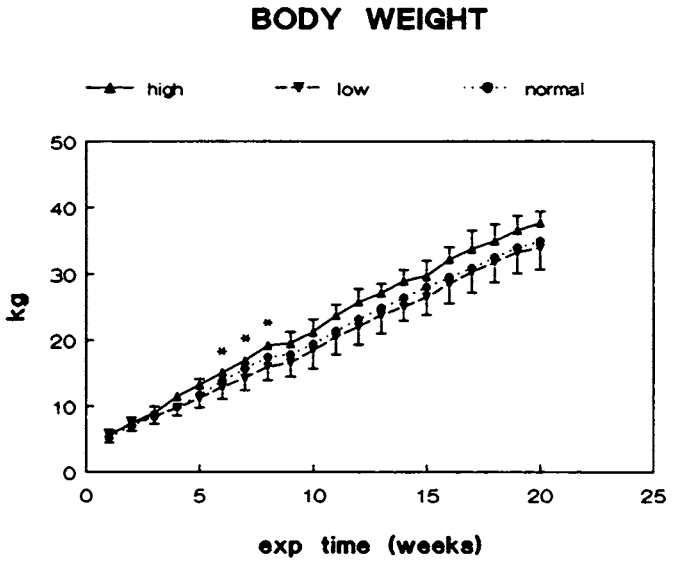

FIGURE 1 Mean (±SD) body weights (BW) of Great Dane pups fed high (H-Pr, n = 6; 31.6%), normal (N-Pr, n = 5; 23.1%) or low (L-Pr, n = 6; 14.6% protein on dry matter basis, % DM) during the experiment. (*) significant difference ($P \leq 0.05$) between H-Pr and L-Pr group. For clarity, SD of the N-Pr group is not printed.

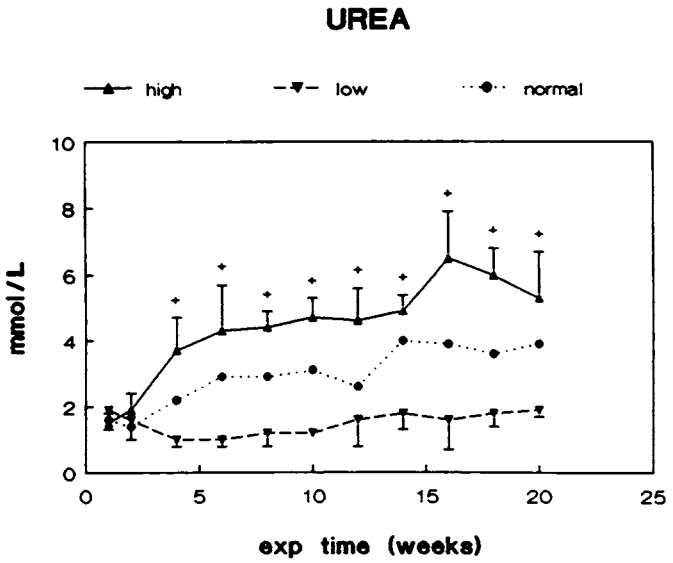

FIGURE 3 Mean (±SD) plasma urea concentration of Great Dane pups fed three different protein levels. (+) significant difference among all three groups. See legend to Figure 1 for further explanation.

were significantly different from zero. Differences between groups were not significant for PCV, WBC, γ-globulin, total plasma calcium, $calcium_{adj}$, phosphorous, AP, ALT and γ-GT. In all dogs, the shape of the ulnar styloid process developed from rectangular or square in W 3 to cone-shaped in W 9. Partial or complete ossification of the apex of the styloid process was present in W 9, while at that time complete fusion of the ossified apex with the styloid process had occurred in one H-Pr dog. The latter stage was reached in all dogs in W 15. The anconeal process was partially or completely ossified in all dogs in W 9 and fusion of the anconeal process with the olecranon had occurred in one N-Pr and one H-Pr dog. In W 15, the anconeal process was completely ossified in all dogs and fused with the olecranon in all five N-Pr, five H-Pr dogs and three L-Pr dogs. In all dogs except for one L-Pr dog, radiologically detectable fusion of the anconeal process with the olecranon had occurred in W 21. Flattening or indentation of the physeal border of

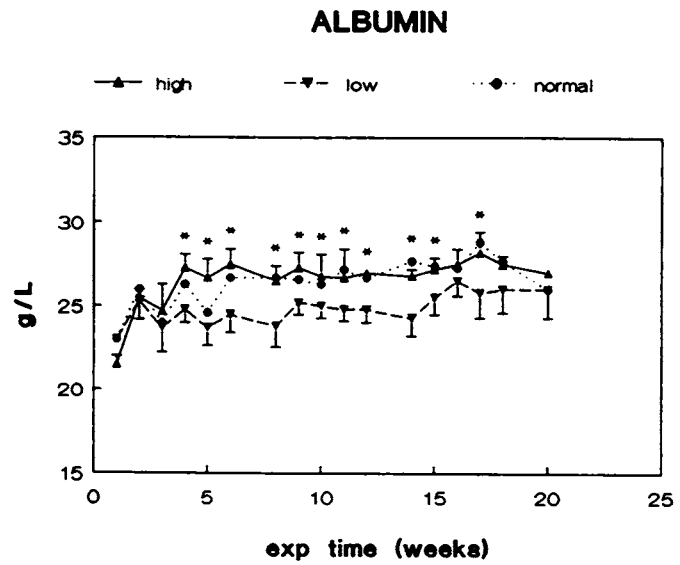

FIGURE 2 Mean (±SD) plasma albumin concentrations of Great Dane pups fed three different protein levels. See legend to Figure 1 for further explanation.

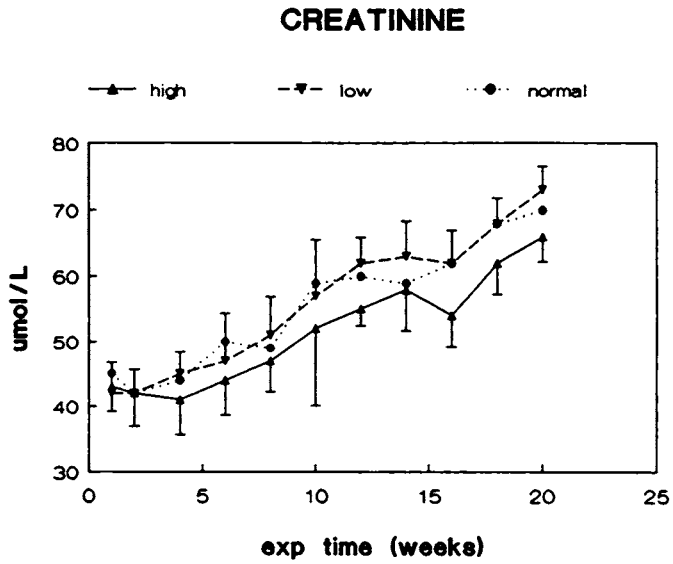

FIGURE 4 Mean (±SD) plasma creatinine concentration of Great Dane pups fed three different protein levels. No significant differences were found among groups at any time. See legend to Figure 1 for further explanation.

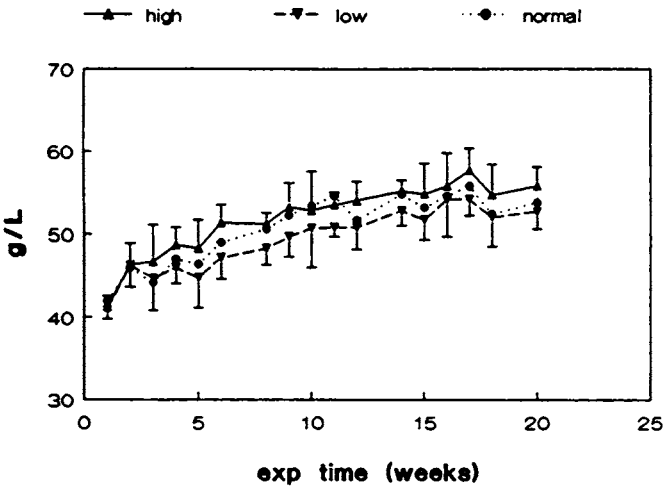

FIGURE 5 Mean (±SD) plasma total protein concentrations of Great Dane pups fed three different protein levels. No significant differences were found among groups at any time. See legend to Figure 1 for further explanation.

the distal ulnar metaphysis, or even a retained cartilage cone in the distal ulnar metaphysis, was present in all dogs at some point in W 3, 9 or 15. Improvement of architecture of the distal ulnar metaphysis was noted in W 21, resulting in a normal shape of the distal ulnar metaphysis in four N-Pr dogs, three H-Pr dogs and four L-Pr dogs. The abnormalities in the remaining dogs were confined to a small remnant of a cartilage cone in one H-Pr dog and minor flattening of the distal ulnar metaphysis in the other dogs. A distinct bony spur at the palmar aspect of the distal ulnar metaphysis, which was present in all dogs at some point in W 3, 9 or 15, had disappeared in W 21. There was no difference in the mean length of the radius and the ulna between the groups in W 3, 9, 15 and 20.

In all groups food intake was such that there were no significant differences in calcium intake (V_I). In addition, no significant differences were found in calcium kinetics among all groups, including the percentage calcium absorption from the intestinal tract (α), calcium-accretion (V_0^+), and calcium resorption from the skeleton (V_0^-) (**Table 4**).

At pathological examinations no macroscopic lesions were present in the various organs of any dog. In one dog in the L-Pr group an ununited anconeal process (UAP) was found. Along the midsagittal cut surface of the long bones, irregularities of retained cartilage of varying degrees of both articular and physeal growth plate cartilage were present. Such osteochondral lesions were especially seen in the caudocentral part of the proximal humeral articular cartilage, in the distal ulnar physeal growth plate cartilage and in the growth plate of the ribs. The severity of these lesions differed between dogs and was equally present in each group. Histomorphologically the mean width of the growth plate cartilage of the costochondral junction of the ribs decreased with increasing age (**Table 5**; physis). Allowing for age, no significant differences were calculated between groups of dogs. There were also no differences in the amount or thickness of the metaphyseal bone trabeculae of both primary and secondary spongiosa between the groups. The same applied for the amount and thickness of osteoid seams on the trabeculae and the amount of remaining cartilage matrix in the trabeculae. Histomorphometric data on specific areas of trabecular bone in the rib are given in Table 5. Neither the V%b nor the Obs, Ocls and Ocl were different among the groups. The amount of parafollicular cells in the thyroid and the activity of the parathyroid glands did not differ histologically among the three groups. In cervical spinal cord there were minimal degenerative changes, especially in the segments between the third and fifth cervical vertebrae in some dogs, but these lesions occurred equally in all three groups. There were no histological lesions in other soft tissues.

DISCUSSION

Protein requirements in dogs have been discussed for over 50 y and numerous experiments have been carried out to determine the optimal protein content

TABLE 4

Calcium kinetic measures in three groups of Great Dane pups fed different levels of protein intake[1]

Measure	W	H-Pr (n = 6)	N-Pr (n = 5)	L-Pr (n = 6)
V_I, *mmol/kg$^{0.75}$ per day*	1	20.2 ± 0.5	19.3 ± 2.0	18.5 ± 1.7
	7	12.4 ± 1.7	12.5 ± 1.5	13.7 ± 1.5
	13	9.4 ± 1.2	10.1 ± 0.7	10.7 ± 0.8
	19	7.8 ± 1.5	8.3 ± 1.1	8.5 ± 0.5
α, %	1	77 ± 8	79 ± 8	77 ± 7
	7	66 ± 10	70 ± 7	50 ± 17
	13	74 ± 7	60 ± 5	55 ± 4
	19	72 ± 12	67 ± 19	67 ± 14
V_0^+, *mmol/kg$^{0.75}$ per day*	1	36.2 ± 4.0	33.8 ± 1.7	34.3 ± 2.0
	7	32.2 ± 2.3	33.6 ± 1.4	29.3 ± 1.6
	13	23.1 ± 3.5	24.7 ± 2.5	22.2 ± 1.4
	19	16.2 ± 1.9	16.4 ± 1.7	16.0 ± 1.9
V_0^-, *mmol/kg$^{0.75}$ per day*	1	21.9 ± 5.3	19.9 ± 3.0	20.6 ± 3.0
	7	24.9 ± 2.5	25.8 ± 0.6	24.0 ± 2.9
	13	16.7 ± 3.2	18.8 ± 3.1	17.4 ± 1.2
	19	10.9 ± 2.7	11.3 ± 3.3	10.9 ± 2.1

[1] Values are means ± SD Ca intake (V_I), percentage intestinal Ca absorption (α), Ca accretion (V_0^+) and Ca resorption from the skeleton (V_0^-) at W 1, 7, 13 and 19 in Great Dane pups fed high (H-Pr), normal (N-Pr) or low protein (L-Pr) food. No statistically significant differences were found. V_I, V_0^+ and V_0^- are expressed as mmol/kg metabolic body weight (kg$^{0.75}$).

TABLE 5

***Histomorphometric measures of three groups of Great Dane pups fed different levels of protein*[1]**

Measure	W	H-Pr	N-Pr	L-Pr
Physis, μm	3	109 ± 8 (3)	116 ± 11 (1)	117 ± 29 (3)
	9	96 ± 14 (3)	100 ± 9 (3)	96 ± 11 (3)
	15	92 ± 14 (6)	94 ± 15 (5)	87 ± 18 (6)
	21	69 ± 26 (11)	78 ± 25 (7)	72 ± 24 (10)
Obs	3	49.0 ± 7.3 (3)	45.4 (1)	44.0 ± 2.1 (3)
	9	47.8 ± 2.0 (3)	49.7 ± 7.4 (3)	46.9 ± 3.1 (3)
	15	59.0 ± 8.1 (6)	56.5 ± 4.4 (5)	56.6 ± 7.0 (6)
	21	53.0 ± 7.1 (11)	60.6 ± 5.7 (7)	56.0 ± 7.5 (10)
Ocls	3	16.5 ± 3.6 (3)	10.5 (1)	14.4 ± 4.8 (3)
	9	16.4 ± 4.8 (3)	15.2 ± 3.6 (3)	13.0 ± 2.7 (3)
	15	14.8 ± 4.6 (6)	13.8 ± 4.8 (5)	12.5 ± 2.3 (6)
	21	11.8 ± 4.8 (11)	10.2 ± 3.7 (7)	11.8 ± 4.3 (10)
Ocl	3	14.3 ± 2.1 (3)	10.3 (1)	1.4 ± 1.9 (3)
	9	12.8 ± 1.8 (3)	11.9 ± 1.3 (3)	10.1 ± 2.8 (3)
	15	10.3 ± 2.9 (6)	9.4 ± 1.5 (5)	9.2 ± 1.5 (6)
	21	7.0 ± 2.1 (11)	6.2 ± 1.5 (7)	6.9 ± 1.6 (10)
V%b	3	20.9 ± 3.0 (3)	17.2 (1)	18.8 ± 3.6 (3)
	9	20.8 ± 0.9 (3)	19.5 ± 2.6 (3)	17.6 ± 0.7 (3)
	15	20.9 ± 3.0 (6)	18.8 ± 2.9 (5)	17.9 ± 4.9 (6)
	21	23.5 ± 2.5 (11)	22.7 ± 5.7 (7)	19.5 ± 5.3 (10)

[1] Values are means ± SD; no. of biopsies evaluated in parentheses. Biopsies were taken of the 9th or 10th rib at the following times: W 3 (9th left), W 9 (9th left), W 15 (9th right) and W 21 (10th). Physeal height (physis), percentage of trabecular bone covered by osteoblasts (Obs) and by osteoclasts (Ocls), number of osteoclasts (objective 25×) per microscopic field (Ocl) and the volume percentage of trabecular bone (V%b) did not differ significantly among groups.

of the food and the optimal protein sources (21–25). The use of different bases in the literature to express the protein content of the diet, i.e., as percentage in the product or on a dry matter basis, as grams protein/1000 kJ, or as protein to energy ratio is confusing.

The minimal protein requirements reported in the literature differ between studies from 11.5% (26) to 22% DM. (27). The protein requirement depends on factors such as digestibility, amino acid composition, proper ratios among the essential amino acids and their availability from the protein source, energy density of the food and physiological state of the dog (26).

The growth in length of the dogs receiving food only differing in protein content did not differ, as revealed by measurements of height at the shoulder and length of the radius and ulna measured on radiographs. The significant differences in body weight in W 6–8 between the H-Pr and the L-Pr groups may have been the result of the high protein requirements at that very young age (26, 28, 29), the protein supply in the L-Pr group being suboptimal. The body weights finally reached were about the same as those observed by Hedhammer et al. (3) in Great Danes fed ad libitum.

In this study the differences in protein intake definitely had consequences on some biochemical measures. The serum albumin concentration of the L-Pr group was lower than that of the H-Pr group. Although the values in the L-Pr group were still within the reference range, this finding indicates that the protein content in the food of 14.6% (% DM), i.e., 13% of energy as protein, with the protein quality as used in our experiment, was just below optimal requirements for growing dogs of giant breeds under 27 wk of age. This is in agreement with recommendations of a minimum requirement of 16% of energy as protein for growing dogs (29–31).

There were also differences in metabolites of protein metabolism, i.e., urea and creatinine. In the H-Pr dogs, plasma urea concentration was higher and the creatinine concentration was lower than in the L-Pr dogs. This is in accordance with the finding that excessive dietary protein is metabolized and increases the glomerular filtration rate (32, 33). There was no histological evidence for kidney damage in any of the groups.

The alterations in protein metabolism had no demonstrable consequences on calcium metabolism, osteoblastic activity and calcium accretion. Thus, the influence of changes in dietary protein on calcium kinetics observed in several other species (7, 9–13) was not seen in this study. It seems unlikely that the protein content of the food is an important determinant of disturbances in enchondral ossification in large breeds of dogs.

The radiographic and histologic examinations nevertheless revealed changes compatible with disturbed enchondral ossification. The changes were equally distributed among the groups, indicating that they were not related to protein intake but rather to genetic factors or another food constituent, most likely calcium, as was demonstrated in previous experiments with Great Danes (5, 6, 19).

It is concluded that in this study the differences in protein intake per se did not affect the occurrence of disturbed skeletal development in young Great Danes, and that an etiologic role for dietary protein in the development of osteochondrosis in dogs is unlikely. From the differences in body weight and the relatively low plasma albumin concentrations in the L-Pr group, it is concluded that 14.6% protein on a dry matter basis (13% of energy as protein) in the food is marginal for giant breeds of dogs during growth.

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LITERATURE CITED

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