

# Maintenance energy requirements and the effect of diet on performance of racing Greyhounds

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**Objectives**—To determine maintenance energy requirements and effect of diet on performance of racing Greyhounds.

**Animals**—7 adult racing Greyhounds.

**Procedure**—Dogs were fed a higher fat and protein (HFP) or a lower fat and protein (LFP) diet for 8 weeks in a crossover design. Dogs were exercised for 15 minutes twice daily in a paddock and raced 500 m twice weekly. Blood gas, hematologic, and serum biochemical analyses were performed before and after racing, and race times were compared at the end of each diet period.

**Results**—Mean race time was significantly shorter ( $32.81 \pm 0.65$  seconds vs  $33.05 \pm 0.71$  seconds), and mean racing speed over 500 m was significantly faster ( $15.25 \pm 0.30$  vs  $15.13 \pm 0.30$  m·s<sup>-1</sup>) when dogs were fed the HFP diet than when they were fed the LFP diet. Diet had little or no effect on results of blood gas, hematologic, and serum biochemical analyses, except that Hct was 4% greater before and after racing when the HFP diet was fed than when the LFP diet was fed. Mean SD metabolizable energy intake from weeks 1 through 16 was  $155 \pm 9$  kcal·kg<sup>-0.75</sup>·d<sup>-1</sup>.

**Conclusions and Clinical Relevance**—Racing Greyhounds ran faster when fed a diet containing higher fat and protein and lower carbohydrate contents. Their maintenance metabolizable energy requirement was slightly higher than that of moderately active dogs. (*Am J Vet Res* 2000;61:1566–1573)

Greyhound racing is a major industry in Florida and around the world, but there is little scientific information concerning the relationship between nutrient composition and performance in racing Greyhounds. Toll et al<sup>1</sup> describe a study in which Greyhounds ran faster when fed a diet containing a moderate fat content (31% of metabolizable energy [ME] as fat), compared with a diet with an extremely high fat content (75% of ME as fat). Other authors have suggested that Greyhounds should be fed a diet containing a high carbohydrate content and less fat to maximize muscle glycogen content and minimize the severity of lactic acidosis that develops during a race.<sup>2,3</sup> Evidence from

endurance racing dogs, however, suggests that dogs perform better when fed a high-fat diet.<sup>4-11</sup>

Maintenance energy requirements of racing Greyhounds are also unknown. The National Research Council (NRC) has suggested<sup>12</sup> that the mean maintenance energy requirement of adult dogs in kennels undertaking moderate exercise is 132 kcal·kg<sup>-0.75</sup>·d<sup>-1</sup>. Thus, the maintenance energy requirement for a 30-kg Greyhound would be 1,700 kcal·d<sup>-1</sup>. An Australian survey<sup>13</sup> suggested that Greyhound trainers fed their dogs 2,400 to 2,600 kcal·d<sup>-1</sup>, but food consumption surveys are notoriously inaccurate. Grandjean and Paragon<sup>14</sup> have estimated that Greyhounds require 150 to 190 kcal·kg<sup>-0.75</sup>·d<sup>-1</sup>, or somewhat more than is required by moderately active dogs, but have provided no empirical evidence to support this conclusion.

The purposes of the study reported here, therefore, were to determine whether a diet containing higher fat and protein and lower carbohydrate contents altered performance (ie, racing speed) or severity of lactic acidosis in racing Greyhounds and to measure the energy intake of racing Greyhounds maintaining a constant body weight while in training.

## Materials and Methods

**Experimental animals**—Eight Greyhounds (3 females, 5 males) weighing  $31.2 \pm 2.3$  kg (mean  $\pm$  SD) that were 22  $\pm$  2 months old were used in the study. Greyhounds had been trained to chase a lure on a racetrack and were donated by Greyhound breeding kennels. During the study, 1 of the female dogs developed a foreign body (stick) abscess in a front foot, and although the dog returned to racing after 2 weeks of treatment, data from this dog were excluded from analyses.

All dogs were vaccinated against rabies, distemper, leptospirosis, hepatitis, and parvovirus infection. Dogs were considered to be in good health on the basis of results of physical examination, CBC, serum biochemical analyses, urinalysis, and fecal examination for parasites. Results of serologic tests for heartworm antigen and antibody to *Ehrlichia canis* and *Rickettsia rickettsii* were negative. Some dogs had low titers of antibodies to *Babesia canis*, but evaluation of blood smears did not reveal any evidence of overt parasitism during the study.

The anthelmintic milbemycin oxime<sup>a</sup> (23 mg) was administered PO every 4 weeks. Praziquantel<sup>b</sup> (142 mg) was administered every 8 weeks to eliminate tapeworms, and dogs were bathed<sup>c</sup> every 4 weeks to remove any fleas or ticks. Testosterone<sup>d</sup> (1 mg/kg) was administered to female dogs IM every 2 weeks to prevent estrus (testosterone is widely used to prevent estrus in female racing Greyhounds in training). Repeated fecal examinations indicated that 1 dog had a patent *Capillaria* spp infection that was resistant to anthelmintic treatment. This dog was included in the study, because results of hematologic testing, thoracic radiography,

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bronchoscopy, and rhinoscopy failed to reveal any clinically important abnormalities. Necropsy of another affected Greyhound revealed worms in the frontal sinus but did not reveal any other abnormalities. Bone scintigraphy and radiography of the forelimbs and hind limbs were performed on all dogs to evaluate the importance of slight joint abnormalities observed during physical examination. There was mild radiographic evidence of degenerative joint disease and some slight scintigraphic evidence of bone remodeling in some dogs, but these changes appeared to be minor and not influence racing performance.

Dogs were cared for according to the principles outlined in the NIH *Guide for the care and use of laboratory animals*<sup>15</sup>; the study protocol was approved by the Institutional Animal Care and Use Committee. All dogs were housed in 1.4 × 1.9 m cages in a room with a 12-hour light:dark cycle, a constant temperature of 24 C, and 13 to 18 air changes·h<sup>-1</sup>. All dogs were exercised for 15 minutes twice daily in a 30 × 30 m grass paddock and raced twice a week. Each race was 500 m (five-sixteenths of a mile) in length and performed on a 400 m (1/4 mile) oval soft sand-clay track with 10° banking on the corners. Dogs chased a mechanical lure maintained 10 to 20 m in front of the lead dog. Race times were measured with the aid of a photofinish camera.<sup>c</sup> Ambient temperature and humidity were measured at the track before each race.

Dogs were raced in groups of 4, and starting position on the track was assigned according to racing preference of the dogs (eg, dogs that preferred to race along the rail were given inside starting positions to minimize interference and bumping). Several combinations of dogs were tested during an acclimation period to determine which combination resulted in the least interference during racing, and this combination was used for the duration of the study.

All dogs were fed once daily after their morning exercise (at 9 AM); water was available at all times. Each dog was offered food in excess of its estimated requirement and allowed to eat for 30 to 40 minutes. Excess food was removed once each dog had voluntarily stopped eating. The amount of food offered and any residual food left after eating were weighed to determine food intake. Each dog was weighed after defecation and urination but immediately before exercise each week. Dogs were photographed during the final week of each diet period.

Dogs were fed 1 of 2 extruded diets: a **higher fat and protein (HFP)** diet (32% of ME as fat, 25% of ME as protein, and 43% of ME as carbohydrate) or a **lower fat and protein (LFP)** diet (25% of ME as fat, 21% of ME as protein, 54% of ME as carbohydrate). Both diets were composed of wheat, poultry meal, and meat and bone meal with vitamins and minerals added to conform to **American Association of Feed Control Officials (AAFCO)** recommendations for adult dogs.<sup>16</sup> The amount of wheat was increased by 15% and the amount of poultry meal was decreased by 15% in the LFP, compared with the HFP, diet. Each diet was manufactured as a single batch, and a representative 1 kg sample of each diet was stored at -20 C for subsequent analysis. Initially, all dogs were fed a mixture of the 2 test diets (50% HFP diet:50% LFP diet) during a 7-week acclimation period. Dogs were then randomly assigned to 2 groups of 4 dogs. One group of dogs was fed the HFP diet for weeks 1 through 8 of the study and the LFP diet for weeks 9 through 16. The other group was fed the LFP diet for weeks 1 through 8 of the study and the HFP diet for weeks 9 through 16. Randomization of dogs to diets was stratified to ensure that there were 2 dogs from each diet group in each race.

Blood samples were obtained, and rectal temperature was measured in the kennel before and at the track 5 minutes after each twice-weekly race during the fourth and eighth weeks of each diet period. A heparinized blood sample was

immediately transported on ice for blood gas analysis. Blood was collected in 2-ml evacuated tubes containing potassium EDTA (7.5%) for hematologic analyses, in 5-ml evacuated tubes containing sodium fluoride (10 mg) and potassium oxalate (12.5 mg) for analysis of serum lactate concentration, and in 6-ml evacuated tubes coated with silica for clot activation and containing a gel for clot separation for serum biochemical analyses. Samples for serum biochemical analyses and plasma lactate concentration were centrifuged at 500 × g for 15 minutes at 4 C. Plasma was stored at -20 C, and plasma lactate concentration was measured colorimetrically<sup>d</sup> in duplicate within 1 month. Automatic analyzers<sup>g,j</sup> were used to determine RBC count, **mean cell volume (MCV)**, hemoglobin concentration, Hct, and WBC count; to measure serum sodium, potassium, chloride, urea nitrogen, creatinine, total calcium, glucose, total bilirubin, cholesterol, albumin, globulin, triglyceride, and inorganic phosphate concentrations and **alkaline phosphatase (ALP)**, **alanine aminotransferase (ALT)**, **aspartate aminotransferase (AST)**, **amylase**, **creatine kinase (CK)**, and lipase activities; and to measure venous blood pH, bicarbonate concentration, P<sub>CO</sub><sub>2</sub>, P<sub>O</sub><sub>2</sub>, and base excess. Analyses were performed on the day of collection. Reproducibility was established by repetitive analysis of standard samples and routine standardization of electrodes. The time that dogs panted after each race was also recorded.

All feces passed from noon Monday until noon Friday on the eighth week of each diet period were collected by means of direct catch into plastic bags.<sup>k</sup> Feces were weighed and frozen at -20 C for subsequent analysis. Fecal samples were lyophilized and reweighed to determine **dry matter (DM)** weight. Samples from each dog for each week were pooled and homogenized by grinding in a blender<sup>l</sup> to the consistency of powder.

Standard methods were used by the Analytic Laboratory of the Waltham Center for Pet Nutrition to measure major nutrient<sup>17</sup> and dietary fiber<sup>18</sup> contents of each food sample and each pooled fecal sample. Protein content was analyzed by use of a method based on the Dumas principle,<sup>19</sup> using a nitrogen analyzer.<sup>m</sup> Protein, fat, ash, insoluble fiber, and moisture contents were subtracted from the total to obtain the **nitrogen-free extract (NFE)** content. Digestibilities of nutrients were calculated for each food as 100 minus the percentage of each nutrient consumed by each dog that was collected in the feces during the 4 days of fecal collection. The ME density and the percentage of the ME provided by each major nutrient were calculated by multiplying these mean digestibilities by the gross energy values of 4.4 (5.65 - 1.25 for nitrogen energy loss in the urine), 9.4, and 4.15 kcal/g for protein, fat, and NFE, respectively.<sup>12</sup>

The essential nutrient composition of the food samples was also measured by the Waltham Center for Pet Nutrition Laboratory. To measure essential fatty acids content, fat was extracted by use of a cold mixture of chloroform, methanol, and water.<sup>20</sup> The extracted fat was saponified and methylated with an internal standard, using methanolic sodium hydroxide and boron trifluoride complex under reflux. The resulting fatty acid methyl esters were extracted into heptane and analyzed by means of capillary gas chromatography with a flame ionization detector. To measure mineral contents, samples were ashed in a porcelain crucible at 550 C. They were then cooled, and 5% w/v ammonium nitrate was added until the ash had fully dissolved to complete oxidation of the organic matter. Samples were evaporated, and 10 ml of 6N HCl was added. They were left to dry, and 10 ml of 10% HCl was added to solubilize the chloride salts. Samples were filtered, rinsed, and mixed with deionized water in calibrated 200 ml flasks. Calcium, copper, iron, magnesium, manganese, potassium, sodium, and zinc concentrations in the solution were analyzed against matrix-matched standards

using an atomic absorption spectrometer. This solution was also used to measure phosphorus concentration spectrophotometrically at 420 nm, after a colorimetric reaction with ammonium vanadate-molybdate.

Vitamin B1 (thiamine) and B2 (riboflavin) contents were measured by use of **high-performance liquid chromatography (HPLC)** with fluorescence detection; riboflavin content was measured directly, and thiamine content was measured after formation of the thiochrome derivative. Analytes were extracted from the sample simultaneously, using hot dilute HCl and subsequent enzymatic incubation. Niacin, pantothenic acid, vitamin B6, vitamin B12, folate, and biotin contents were analyzed by means of microbiologic turbidimetric methods that closely followed procedures advocated by the Association of Official Analytic Chemists.<sup>21</sup> Nicotinic acid and pyridoxine were extracted with dilute HCl, and contents were assayed by use of *Lactobacillus plantarum* and *Saccharomyces cerevisiae*, respectively. Vitamin B12, pantothenic acid, and folic acid were extracted with sodium acetate buffer and enzymes, and contents were assayed by use of *Lactobacillus plantarum* (vitamin B12 and pantothenic acid) and *Enterococcus hirae* NCIMB 6459 (folic acid). To measure vitamin A and E contents, each sample was hydrolyzed with ethanolic potassium hydroxide solution, and the vitamins were extracted into petroleum ether. The petroleum ether was removed by evaporation, and the residue was dissolved in isopropanol. Vitamin A and E concentrations in the isopropanol extract were determined by use of reversed-phase HPLC.

**Statistical analyses**—Results are reported as mean  $\pm$  SD. Normal probability plots of the data were inspected visually, and the Shapiro-Wilke test was performed to assess whether data were normally distributed. Values for any parameter that appeared to not be normally distributed at all sample times or that appeared to have unequal variances were log transformed prior to analysis. Mean daily food intake (weight of food as fed) and mean ME intake relative to metabolic body weight ( $Hg^{0.75}$ ) were calculated for each week of the study. Data were analyzed as a 2-period crossover design with a within-period repeated-measures factor, using generalized least-squares estimation. Measurements over time within each period (eg, weekly [mean food and ME intake] and semiweekly measurements [race times]) and measurements obtained before and after each race were treated as within-subject repeated measures. An autoregressive heterogeneous covariance structure was used to explicitly model the correlation between measurements taken at these times. Moreover, a random effect for each subject was included. Diet, period, sequence, race, and a diet  $\times$  race interaction were included as factors in the model. If sequence effects were found, then data from only the first diet period (study weeks 1 through 8) would have been used to test for differences attributable to diet. The method of analysis was essentially the method of analysis first suggested by Hill and Armitage.<sup>22</sup>

Postrace serum biochemical concentrations were adjusted for hemoconcentration by use of the following equation:  $C_{adj} = C_{obs} \times P_1/P_2$ , where  $C_{adj}$  is the adjusted postrace concentration,  $C_{obs}$  is the observed postrace concentration,  $P_1$  is the total protein concentration before racing, and  $P_2$  is the total protein concentration after racing. Adjusted and unadjusted postrace concentrations were compared with prerace concentrations separately. Ambient temperature and humidity were regressed against race time, using data from all dogs for the duration of the study. Given a type-I error of 0.05 and the expected SD for differences between values obtained when dogs were fed 1 diet versus the other if all 8 dogs had completed the study, we calculated that we would be able to detect differences of 2 kg in body weight, 30 g·d<sup>-1</sup> in food intake, 10 kcal·kg<sup>-0.75</sup>·d<sup>-1</sup> in ME intake, 0.5 seconds in race

time, 0.03 in pH, 0.3 mg·dl<sup>-1</sup> in protein or creatinine concentration, 1 mEq·L<sup>-1</sup> in potassium, calcium, or phosphorus concentration, 2 mEq·L<sup>-1</sup> in sodium or chloride concentration, 2 mg·dl<sup>-1</sup> in urea nitrogen concentration, 10 mg·dl<sup>-1</sup> in glucose and triglycerides concentrations, 10 U·L<sup>-1</sup> in ALT, AST, and ALP activities, 300 U·L<sup>-1</sup> in CK and lipase activities, 3% in Hct, and 2 fl in MCV (ie, differences equal to the estimated SD of the difference between the 2 groups) with a type-II error of 0.2. Detectable differences were slightly (10%) greater when only 7 dogs completed the study.<sup>23</sup> All analyses were performed by use of computer software.<sup>24</sup> Values of  $P \leq 0.05$  were considered significant.

## Results

The HFP diet contained 24.6% of ME as protein, 32.5% of ME as fat, and 42.6% of ME as NFE; the LFP diet contained 20.9% of ME as protein, 24.9% of ME as fat, and 54.3% of ME as NFE (Table 1). Vitamin and mineral contents of the 2 diets were similar (Table 2), and any differences were probably inconsequential.

Table 1—Proximate analysis, fatty acid content, fiber content, and estimated metabolizable energy (ME) content of 2 experimental diets used to evaluate the effect of diet on performance (ie, racing speed) of racing Greyhounds

Nutrient	Higher fat and protein diet	Lower fat and protein diet
Protein		
As fed (%)	27.0	21.9
% of ME	24.6	20.9
Fat		
As fed (%)	14.6	10.7
% of ME	32.5	24.9
Linoleic acid (mg · kcal <sup>-1</sup> )	3.8	4.6
Linolenic acid (mg · kcal <sup>-1</sup> )	0.7	0.4
Arachidonic acid (mg · kcal <sup>-1</sup> )	0.06	0.05
Nitrogen free extract		
As fed (%)	42.2	50.0
% of ME	42.6	54.3
Ash (mg · kcal <sup>-1</sup> )	17.0	14.5
As fed (%)	6.8	5.8
Insoluble fiber (mg · kcal <sup>-1</sup> )	12.0	17.2
As fed (%)	4.8	6.4
Soluble fiber (mg · kcal <sup>-1</sup> )	2.5	3.9
As fed (%)	1.0	1.4
Moisture (% as fed)	4.6	5.2
Energy density (kcal · g <sup>-1</sup> )	4.01	3.71

Table 2—Mineral and vitamin contents of 2 experimental diets used to evaluate the effect of diet on performance of racing Greyhounds

Nutrient	Higher fat and protein diet	Lower fat and protein diet
Calcium (mg · kcal <sup>-1</sup> )	2.7	2.4
Phosphorus (mg · kcal <sup>-1</sup> )	2.1	2.0
Sodium (mg · kcal <sup>-1</sup> )	1.6	1.5
Potassium (mg · kcal <sup>-1</sup> )	1.2	1.2
Magnesium (mg · kcal <sup>-1</sup> )	0.2	0.3
Iron (μg · kcal <sup>-1</sup> )	91	74
Copper (μg · kcal <sup>-1</sup> )	1.9	2.3
Zinc (μg · kcal <sup>-1</sup> )	16	17
Manganese (μg · kcal <sup>-1</sup> )	9	11
Vitamin A (U · kcal <sup>-1</sup> )	3	4
Vitamin E (μg · kcal <sup>-1</sup> )	8	15
Vitamin B <sub>1</sub> (μg · kcal <sup>-1</sup> )	0.6	1.1
Vitamin B <sub>2</sub> (μg · kcal <sup>-1</sup> )	2.5	2.5
Niacin (μg · kcal <sup>-1</sup> )	13	15
Vitamin B <sub>6</sub> (μg · kcal <sup>-1</sup> )	0.4	0.5
Vitamin B <sub>12</sub> (μg · kcal <sup>-1</sup> )	32	21
Pantothenic acid (μg · kcal <sup>-1</sup> )	7	6
Folic acid (μg · kcal <sup>-1</sup> )	0.1	0.6

Table 3—Digestibilities of major nutrients in 2 experimental diets used to evaluate the effect of diet on performance (ie, racing speed) of racing Greyhounds

Nutrient	Digestibility (%)	
	Higher fat and protein diet	Lower fat and protein diet
Organic matter*	88.9 ± 1.2	86.9 ± 2.0
Dry matter	86.0 ± 1.3	84.3 ± 2.4
Protein	83.1 ± 1.6	80.4 ± 3.4
Fat*	95.1 ± 0.4	91.9 ± 1.1
Nitrogen free extract*	97.7 ± 0.6	96.6 ± 0.5

Data are given as mean ± SD and represent values for 7 Greyhounds.  
\*Values are significantly ( $P \leq 0.05$ ) different between diets.

The HFP diet had a higher energy density, and digestibilities of organic matter, fat, and NFE in the HFP diet were slightly, but significantly, higher than in the LFP diet (Table 3). Both diets conformed to AAFCO recommendations for adult dogs.<sup>16</sup>

Mean ± SD ME intake from weeks 1 through 16 was  $155 \pm 9 \text{ kcal} \cdot \text{kg}^{-0.75} \cdot \text{d}^{-1}$ . Dogs consumed significantly ( $P < 0.001$ ) more food ( $583 \pm 41$  vs  $536 \pm 39 \text{ g} \cdot \text{d}^{-1}$ ) when the LFP diet was fed than when the HFP diet was fed, but ME intake was not significantly different ( $155 \pm 10$  vs  $154 \pm 11 \text{ kcal} \cdot \text{kg}^{-0.75} \cdot \text{d}^{-1}$ ) because of the greater energy density of the HFP diet, and mean body weight when dogs were fed the HFP diet ( $33.9 \pm 2.4 \text{ kg}$ ) was not significantly different from mean body weight when dogs were fed the LFP diet ( $33.9 \pm 1.9 \text{ kg}$ ) (Fig 1). Subjectively, the photographic appearance of the dogs was also not affected by diet. During the 16 weeks of the study, mean body weight increased from 32 to 34 kg, but dogs maintained good body condition and did not become obese. Most of the increase in body weight occurred during weeks 1 through 3; dogs then maintained the same mean body weight (34 kg) for the rest of the study and for a further 22 weeks after the study was complete.

Dogs appeared to require 4 weeks at the start of each diet period to adjust to the change in energy density of the diet. Slight sequence effects were also found for a few blood measurements at week 4 but not at week 8. Blood measurements and race times were, therefore, compared using data obtained during the final 3 weeks of each diet period. Mean race time was significantly ( $P = 0.02$ ) shorter ( $32.81 \pm 0.65$  seconds vs  $33.05 \pm 0.71$  seconds) and mean racing speed over 500 m was significantly ( $P = 0.02$ ) faster ( $15.25 \pm 0.30$  vs  $15.13 \pm 0.30 \text{ m} \cdot \text{s}^{-1}$ ) when dogs were fed the HFP diet than when they were fed the LFP diet. Race times varied significantly ( $P < 0.001$ ) from week to week, but dogs in both groups were affected similarly (Fig 1), suggesting that weekly variations in race time were likely attributable to some unmeasured factor, such as track firmness. Ambient temperature varied from 11 to 25 C, and humidity varied from 24 to 57%, but neither was significantly correlated with race time ( $r^2 < 0.01$ ).

Hematocrit, RBC count, hemoglobin concentration, albumin concentration, cholesterol concentration, and CK activity were significantly increased when the HFP diet was fed, compared with values obtained when the LFP diet was fed (Tables 4 and 5). Statistically significant, but physiologically unimpor-

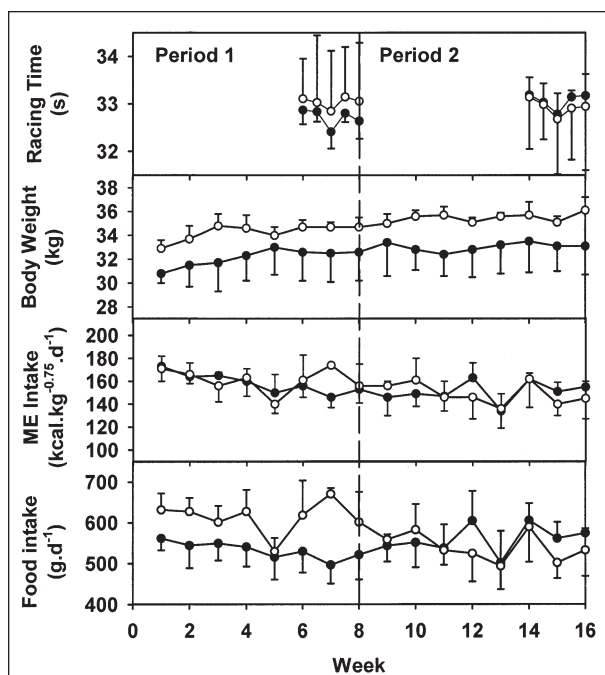


Figure 1—Mean racing times, body weight, metabolizable energy (ME) intake, and food intake (mean ± SD) of 2 groups of racing Greyhounds during 2 consecutive periods of 8 weeks. Group-1 dogs (solid circles,  $n = 4$ ) were fed a higher fat and protein diet (HFP) during period 1 (weeks 1–8) and a lower fat and protein diet (LFP) during period 2 (weeks 9–16); group-2 dogs (open circles, 3) were fed the LFP diet during period 1 and the HFP diet during period 2.

Table 4—The effect of diet on hematologic variables before and after racing in 7 racing Greyhounds

Variable	Higher fat and protein diet		Lower fat and protein diet	
	Before racing	After racing	Before racing	After racing
Hct (%)*†	60 ± 1	70 ± 3	56 ± 3	66 ± 4
RBC count ( $\times 10^6 \cdot \mu\text{l}^{-1}$ )*†	8.8 ± 0.3	10.2 ± 0.7	8.2 ± 0.4	9.6 ± 0.8
Hemoglobin ( $\text{g} \cdot \text{dl}^{-1}$ )*†	21.1 ± 1.3	23.3 ± 0.5	19.6 ± 0.8	22.6 ± 0.8
Mean corpuscular volume (fl)†	68 ± 1	70 ± 3	69 ± 2	69 ± 2
WBC count ( $\times 10^3 \cdot \mu\text{l}^{-1}$ )†	6.9 ± 2.0	8.6 ± 2.3	7.7 ± 2.5	9.0 ± 3.2

Data are given as mean ± SD. Dogs were fed each diet for 8 weeks; values were obtained during the eighth week of each diet period.  
\*Values are significantly ( $P \leq 0.05$ ) different between diets. †Values obtained after racing are significantly ( $P \leq 0.05$ ) different from values obtained before racing.

tant, differences in lipase activity, urea nitrogen concentration, ALP activity, and AST activity were also attributable to diet. There was a statistically significant, but physiologically unimportant, interaction between racing (pre- vs postrace concentration) and diet in regard to phosphorus concentration. These effects of diet were similar regardless of whether adjusted or unadjusted postrace concentrations were considered. Hematocrit; RBC count; hemoglobin concentration; MCV; WBC count;  $\text{PvO}_2$ ; lactate, sodium, calcium, total protein, albumin, globulin, creatinine, urea nitrogen, glucose, triglyceride, and cholesterol concentrations;

Table 5—The effect of diet on results of serum biochemical analyses before and after racing in 7 racing Greyhounds

Variable	Higher fat and protein diet			Lower fat and protein diet		
	Before racing	After racing		Before racing	After racing	
		Observed	Adjusted*		Observed	Adjusted*
Sodium (mEq • L <sup>-1</sup> )†‡	148 ± 1	159 ± 1	123 ± 4	148 ± 1	159 ± 3	126 ± 6
Potassium (mEq • L <sup>-1</sup> )†‡	4.5 ± 0.2	4.0 ± 0.2	3.1 ± 0.2	4.5 ± 0.2	4.1 ± 0.4	3.2 ± 0.4
Chloride (mEq • L <sup>-1</sup> )‡	113 ± 1	111 ± 2	86 ± 4	113 ± 1	112 ± 3	89 ± 4
Calcium (mg • dl <sup>-1</sup> )†‡	10.4 ± 0.7	11.4 ± 0.4	8.9 ± 0.4	10.4 ± 0.5	11.3 ± 0.4	9.0 ± 0.6
Phosphorus (mg • dl <sup>-1</sup> )†‡§	3.0 ± 1.3	2.8 ± 0.7	2.2 ± 0.5	3.7 ± 0.7	2.1 ± 1.2	1.6 ± 0.9
Total protein (g • dl <sup>-1</sup> )†	5.9 ± 0.2	7.6 ± 0.4	5.9 ± 0.2	5.9 ± 0.2	7.4 ± 0.3	5.9 ± 0.2
Albumin (g • dl <sup>-1</sup> )†§	3.1 ± 0.1	4.0 ± 0.3	3.1 ± 0.2	3.0 ± 0.2	3.8 ± 0.2	3.0 ± 0.2
Globulin (g • dl <sup>-1</sup> )†	2.8 ± 0.2	3.6 ± 0.3	2.8 ± 0.2	2.9 ± 0.2	3.6 ± 0.3	2.9 ± 0.2
Creatinine (mg • dl <sup>-1</sup> )†	1.3 ± 0.1	1.6 ± 0.3	1.3 ± 0.3	1.4 ± 0.1	1.6 ± 0.2	1.2 ± 0.2
BUN (mg • dl <sup>-1</sup> )†‡	16 ± 2	18 ± 2	14 ± 1	14 ± 1	16 ± 1	12 ± 1
Glucose (mg • dl <sup>-1</sup> )†‡	96 ± 20	143 ± 24	110 ± 17	92 ± 12	134 ± 24	106 ± 19
Triglyceride (mg • dl <sup>-1</sup> )†‡	37 ± 13	141 ± 33	109 ± 25	39 ± 7	145 ± 25	115 ± 18
Cholesterol (mg • dl <sup>-1</sup> )†	169 ± 21	211 ± 29	163 ± 22	151 ± 24	193 ± 28	153 ± 23
ALT (U • L <sup>-1</sup> )†‡	42 ± 12	58 ± 12	45 ± 10	42 ± 9	60 ± 10	47 ± 8
ALP (U • L <sup>-1</sup> )†‡	63 ± 18	77 ± 22	59 ± 16	74 ± 20	84 ± 26	67 ± 19
AST (U • L <sup>-1</sup> )†‡	50 ± 16	76 ± 23	59 ± 18	33 ± 6	66 ± 7	52 ± 7
CK (U • L <sup>-1</sup> )†	359 ± 260	340 ± 174	262 ± 133	120 ± 58	274 ± 170	218 ± 137
Lipase (U • L <sup>-1</sup> )	377 ± 211	NA	NA	558 ± 212	NA	NA
Amylase (U • L <sup>-1</sup> )	627 ± 147	NA	NA	696 ± 139	NA	NA

Data are given as mean ± SD. Dogs were fed each diet for 8 weeks; values were obtained during the eighth week of each diet period.

\*Values obtained after racing were adjusted for hemoconcentration, using change in total protein concentration as an estimate of degree of hemoconcentration. †Observed values after racing are significantly ( $P \leq 0.05$ ) different from values obtained before racing. ‡Adjusted values after racing are significantly ( $P \leq 0.05$ ) different from values obtained before racing. §There was a significant interaction between racing and diet. ||Values are significantly ( $P \leq 0.05$ ) different between diets. NA = Not applicable.

Table 6—The effect of diet on rectal temperature, results of venous blood gas analyses, and results of serum biochemical analyses before and after racing and on time panting stopped after racing in 7 Greyhounds

Variable	Higher fat and protein diet		Lower fat and protein diet	
	Before racing	After racing	Before racing	After racing
Rectal temperature (C)*	38.8 ± 0.4	40.6 ± 0.5	38.7 ± 0.7	40.6 ± 0.8
Venous pH*	7.38 ± 0.02	7.05 ± 0.02	7.38 ± 0.01	7.03 ± 0.04
Bicarbonate (mEq • L <sup>-1</sup> )*	24.5 ± 0.7	6.3 ± 1.0	26.1 ± 3.1	6.1 ± 1.4
PvCO <sub>2</sub> (mm Hg)*	42 ± 2	24 ± 4	44 ± 5	24 ± 6
PvO <sub>2</sub> (mm Hg)*	55 ± 8	87 ± 8	50 ± 6	86 ± 11
Lactate (mmol • L <sup>-1</sup> )*	0.8 ± 0.3	29.9 ± 5.8	0.6 ± 0.1	35.7 ± 7.9
Panting time (min)	NA	47 ± 16	NA	44 ± 18

\*Values obtained after racing are significantly ( $P \leq 0.001$ ) different from values obtained before racing. NA = Not applicable.

and ALT, AST, ALP, and CK activities were all significantly increased, and venous pH; PvCO<sub>2</sub>; and bicarbonate, potassium, and phosphorus concentrations were all significantly decreased after racing, compared with values obtained before racing (Tables 4, 5, and 6). Adjusted postrace sodium, potassium, chloride, calcium, phosphorus, and urea nitrogen concentrations and ALP activity were significantly decreased, and adjusted glucose and triglyceride concentrations and ALT and AST activities were significantly increased, compared with prerace values.

## Discussion

In the present study, racing Greyhounds ran, on average, 0.2 seconds or 0.1 m • s<sup>-1</sup> faster over a distance of 500 m when fed a diet containing higher fat and protein and lower carbohydrate contents than when fed a diet containing lower fat and protein and higher carbohydrate contents. This time difference was small, but equivalent to a 3-m lead for a dog at the end of a 500-

m race and could represent the difference between winning and losing a race.

Racing speeds (15.2 m • s<sup>-1</sup>) of dogs in the present study were comparable to those for dogs in other scientific studies<sup>24</sup> but slower than those observed at commercial race tracks. Most likely this was attributable to a difference in track surface; commercial race tracks typically have a hard surface, whereas we used a soft sand training track to minimize the risk of injury in the present study. Dogs were also donated for this study because they were too slow for commercial racing.

It is possible that an increase in protein content, an increase in fat content, a decrease in carbohydrate content, or a change in a minor nutrient in the diet could have been responsible for the observed beneficial effect of the HFP diet on performance. It is also possible that changes in 2 or more nutrients acted synergistically. It is not possible in a single experiment to separate these effects, and any explanation as to why dogs ran faster

when fed the HFP diet must, therefore, be speculative until further experiments have been performed.

One possible explanation for why dogs ran faster when fed the HFP diet is that the protein content of the diet increased from 21 to 25% of the ME. The amount of poultry protein also increased, relative to the amount of wheat protein, and meat protein has a higher biological value than wheat protein. The higher dietary fat content of the HFP diet may have also improved protein utilization. The protein content of both diets was higher than the minimum requirement recommended for healthy adult dogs by the NRC (> 8% of ME as protein)<sup>12</sup> or the AAFCO (> 18% of ME as protein).<sup>16</sup> However, most racing Greyhounds are fed meat containing > 30% of ME as protein, and the protein requirements of racing Greyhounds may be higher than those of other dogs.

In racing sled dogs, increasing the dietary protein content from 28 to 32% of ME prevented a decline in Hct during training.<sup>25</sup> This training-induced anemia was exacerbated in dogs fed a vegetable protein diet, compared with dogs fed an animal protein diet,<sup>26</sup> and changes in fat composition were implicated as a cause for this decrease in Hct. In the present study, Hct, RBC count, and hemoglobin concentration all increased when the HFP diet was fed, which may have resulted in increased oxygen delivery to the tissues that may, in turn, have improved performance. The increase in Hct could also reflect a decrease in plasma volume, not an absolute increase in RBC count. However, plasma volume increased, rather than decreased, in racing sled dogs when dietary protein content was increased to 40% of ME.<sup>27</sup> The small increase in urea nitrogen concentration when dogs were fed the HFP diet probably also reflected increased protein turnover but was considered physiologically unimportant, because the change was small and values were within reference limits. This was also true of several other changes in blood measurements attributable to diet.

It is also possible that in the present study dogs ran faster when fed the HFP diet because dietary fat content increased from 25 to 32% of ME, and dietary carbohydrate content decreased from 54 to 43% of ME. The only previous study<sup>1</sup> of the effect of diet on performance in racing Greyhounds found that dogs ran faster when fed a moderate fat diet (31% of ME), compared with a high fat diet (75% of ME), suggesting that excess dietary fat or too little carbohydrate may be detrimental. In combination with results of the present study, this suggests that performance of racing Greyhounds may increase and then decrease as dietary fat content is increased from 25 to 32 to 75% of ME. If this holds true, then results of this study contradict conventional recommendations that high carbohydrate diets be fed to Greyhounds to maximize performance and minimize severity of lactic acidosis.<sup>2,3</sup> Increasing dietary carbohydrate content from 40 to 51% of ME in the present study was associated with a decrease in performance. In addition, mean posttrace lactate concentration was slightly (15%) higher in dogs fed the higher carbohydrate diet, and although this difference was not statistically significant, the power of the study was insufficient to detect

small increases in lactate concentration because of the wide individual variation.

High carbohydrate diets increase muscle glycogen stores and stamina in humans, but high fat, low carbohydrate diets increase stamina in dogs undertaking endurance exercise.<sup>9,10,28</sup> In dogs, muscle fat stores are larger, albumin binds more free fatty acids, the concentration of free fatty acids in the blood is higher, delivery of free fatty acids to the tissues is enhanced, and the amount of energy from fat oxidation at rest and during exercise is greater than in less aerobic species such as humans and goats.<sup>4,5,8</sup> Furthermore, most limb muscle fibers in Greyhounds and crossbred dogs have high oxidative activity,<sup>6,7</sup> and high-fat diets increase resting serum triglycerides and free fatty acids concentrations, mitochondrial volume, and maximal energy expenditure in sled dogs undertaking endurance exercise.<sup>11</sup> Thus, glucose oxidation is the principle source of energy at high rates of energy expenditure (80 to 85% of  $\text{VO}_2 \text{max}$ ), but fat oxidation still provides some energy and may affect maximal energy expenditure.<sup>8</sup> In the study reported here, increased dietary fat content did not increase serum triglycerides concentration, but serum cholesterol concentration and racing speed increased, which suggests that maximal energy expenditure may have increased because of increased fat oxidation.

Changes in blood measurements observed after racing in the present study were similar to those reported previously for Greyhounds that raced more than 500 m.<sup>24,29-32</sup> A marked posttrace acidosis (pH, 7.04) resulted from a 50-fold increase in lactate concentration to 33 mmol·L<sup>-1</sup>. This was partly compensated by a decrease in  $\text{PvCO}_2$ . Total protein concentration increased by 27%, probably as a result of fluid shifts and dehydration. Serum concentrations of many analytes also increased a small amount, but when adjusted for dehydration, serum concentrations of most analytes decreased after racing. Adjusted posttrace serum glucose concentration and ALT and AST activities increased slightly, and serum triglycerides concentration increased markedly, which suggests these analytes were released into the circulation in response to increased circulating concentrations of epinephrine and cortisol.

Unadjusted serum phosphorus concentration decreased during racing in the present study but was reported to increase in previous studies.<sup>29,32</sup> In some dogs, serum phosphorus concentration was low (< 2 mg·dl<sup>-1</sup>) before racing and even lower (< 1 mg·dl<sup>-1</sup>) after racing. Serum phosphorus concentration decreased more when dogs were fed the LFP diet. Phosphorus is required for glycolytic intermediates and ATP. Hypophosphatemia may result in RBC lysis and can cause muscle damage and limit performance.<sup>33</sup> The LFP and HFP diets contained 0.8 and 0.9% phosphorus on a dry matter basis (2.0 and 2.1 mg·kcal<sup>-1</sup>), respectively. This is greater than the NRC's minimum recommendation for adult and growing dogs (1.2 mg·kcal<sup>-1</sup>) and similar to the AAFCO recommendation for growing dogs.<sup>12,16</sup> Further experiments are needed, however, to determine the reason for these low phosphorus concentrations or whether these low phospho-

rus concentrations can be corrected by feeding more phosphorus in the diet.

In the present study, serum CK activity was not substantially increased after racing. A clinically important CK activity ( $> 1,000 \text{ U}\cdot\text{L}^{-1}$ ) was recorded on only 3 occasions, and none of the dogs developed overt signs of rhabdomyolysis. Some dogs appeared slightly stiff after racing but did not have any increase in serum CK activity immediately or 24 hours after racing. Nevertheless, more subtle measures of muscle damage may have revealed subclinical damage.

Greyhounds in the present study maintained their body weight while consuming  $155 \pm 9 \text{ kcal}\cdot\text{kg}^{-0.75}\cdot\text{d}^{-1}$  ( $1,990 \text{ kcal}\cdot\text{d}^{-1}$  for a 30-kg dog), racing twice weekly, and exercising for 15 minutes twice daily. This is slightly more than the  $132/\text{kcal}\cdot\text{kg}^{-0.75}\cdot\text{d}^{-1}$  ( $1,690 \text{ kcal}\cdot\text{d}^{-1}$  for a 30-kg dog) recommended by the NRC for moderately active dogs<sup>12</sup> but is lower than previous estimates<sup>13,14</sup> of maintenance energy requirements of Greyhounds ( $150$  to  $190 \text{ kcal}\cdot\text{kg}^{-0.75}\cdot\text{d}^{-1}$  and  $2,500 \text{ kcal}\cdot\text{d}^{-1}$ ). This suggests that Greyhounds have only slightly higher energy requirements than other breeds undertaking moderate exercise. The energy required for exercise is proportional to distance traveled, rather than speed, so each 500-m race requires little energy ( $75 \text{ kcal}/\text{race}$ ).<sup>14,34</sup> Dogs were raced twice weekly and indulged in only short bursts of sprinting during each exercise period, so it is not surprising that their energy intake was similar to that suggested by the NRC.<sup>12</sup> It is possible that maintenance energy requirements may be slightly higher relative to total body weight in Greyhounds, because lean body mass represents a greater proportion of body weight (57 vs 43%) in Greyhounds than in other breeds.<sup>35</sup>

In the present study, ME of the diet was estimated, taking into account digestibility of the major nutrients. Atwater factors commonly used to calculate the ME content of human foods (4, 9, and  $4 \text{ kcal}\cdot\text{g}^{-1}$  for protein, fat and carbohydrate, respectively) assume protein, fat, and NFE digestibilities of 91, 96, and 96%, respectively.<sup>36</sup> Modified Atwater factors recommended by the AAFCO to calculate the ME content of pet foods (3.5, 8.5, and  $3.5 \text{ kcal}\cdot\text{g}^{-1}$ , respectively)<sup>16</sup> assume protein, fat, and NFE digestibilities of 80, 90, and 85%, respectively. In the present study, the ME ( $\text{kcal}\cdot\text{g}^{-1}$ ) provided by protein, fat, and carbohydrate were high, because the digestibilities of these nutrients were similar to those assumed for human foods. The digestibility of NFE was higher than is usually estimated for pet foods, because all insoluble fiber was excluded from NFE. Nutrients in the HFP diet were 2% more digestible than nutrients in the LFP diet, and the HFP diet was more energy dense than the LFP diet, but dogs ate more of the LFP than the HFP diet. Energy consumption, body weight, and body condition were similar irrespective of diet. Dogs were able to self-regulate energy intake; therefore, when fed each diet free choice, they adjusted their food intake during the first weeks of each diet period to accommodate the change in energy density of the diet.

<sup>a</sup>Interceptor, Novartis, Greensboro, NC.

<sup>b</sup>Droncit (56.8 mg/ml), Haver/Diamond Scientific, Mobay Corp, Shawnee, Kan.

<sup>c</sup>Paramite Dip for Dogs, Vet-Kem, Dallas, Tex.

<sup>d</sup>Testosterone propionate (100 mg/ml), Steris, Phoenix, Ariz.

<sup>e</sup>Nikkor 43–86 mm Zoom, Nikon Inc, Melville, NY.

<sup>f</sup>Lactate diagnostic kit, Sigma Chemical Co, St Louis, Mo.

<sup>g</sup>Cell-Dyn 3500 System, Abbott Laboratories, Abbott Park, Ill.

<sup>h</sup>Express chemistry analyzers M550 and M664, Ciba Corning, South Norwood, Mass.

<sup>i</sup>Kodak Ektachem DT60 analyzer, Johnson & Johnson, Atlanta, Ga.

<sup>j</sup>BGElectrolytes blood gas analyzer, Instrument Laboratory, Lexington, Mass.

<sup>k</sup>Whirl-Pak bags, Fisher Scientific, Pittsburgh, Pa.

<sup>l</sup>Osterizer Galaxie, Oster, Laurel, Miss.

<sup>m</sup>LECO, Cheshire, UK.

<sup>n</sup>SAS/STAT version 6.04, SAS Institute Inc, Cary, NC.

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