

# Freshpet Canine Vital Complete Meal Maintenance Study

## Introduction

Nutritional adequacy in terms of minimum nutrient requirements for dogs and cats is based on cumulated research and updated periodically in the National Research Council (NRC) Nutrient Requirements for Dogs and Cats<sup>1</sup>. These recommendations are typically based on studies using purified diets rather than commercial diets. To compensate for potential reductions in bioavailability, the Association of American Feed Control Officials (AAFCO) publishes its own nutrient profiles for dogs and cats to account for these reductions and potential processing losses<sup>2</sup>. The AAFCO nutrient profiles are published yearly and are what the pet food industry uses as guidelines in formulating dog and cat commercial diets.

Besides setting minimum nutrient requirements, nutritional adequacy claims for commercial pet foods are based on procedures and protocols established by AAFCO<sup>2</sup>. One method of establishing an adequacy claim in the United States is to conduct a feeding trial following AAFCO established protocols. Adequacy claims can also be made from calculated nutrient content of diet formulas using a nutrient data bank or actual chemical analysis of the diets with or without feeding trials. The pet food industry though uses ingredients than can have a wide variation in actual nutrient content and bioavailability. Therefore, feeding protocols are considered the “gold standard” in evaluating pet food for nutritional adequacy. Because of the increased costs involved, not all pet food companies perform feeding trials on their foods.

Grain-free diets are one of the largest growing segments of the pet food market right now. More and more pet owners are buying into the concept that dogs are strict carnivores like their ancestor the wolf, and that feeding grains can be harmful to dogs. Contrary to popular opinion, recent studies have shown through genomic sequencing that dogs have adapted to a diet rich in starch compared to wolves, through selection of genes for starch digestion<sup>3</sup>. There is no reliable evidence that it is harmful to feed grains to dogs. The majority of dogs and cats are very efficient at utilizing the nutrients provided in grains, including protein, vitamins, minerals, fiber and essential fatty acids<sup>4</sup>.

Many pet food companies are substituting grains with legumes or pulses (beans, lentils, peas) or potatoes in their grain-free diets. In association with more and more dogs eating grain-free diets, the FDA has been investigating a link between dogs being fed these diets and dilated cardiomyopathy (DCM)<sup>5,6</sup>. Diet-related DCM has also been traced to diets formulated for dogs with exotic ingredients (i.e. exotic meats, fruits or vegetables).

The purpose of this AAFCO Adult maintenance trial was to evaluate the nutritional adequacy of Freshpet's Vital Grain Free Complete Meal Chicken, Beef, Salmon & Egg Recipe in a group of large breed dogs. In addition, an evaluation of both plasma and whole blood taurine status was done on all the dogs. Taurine deficiency has been found in some of the dogs that have developed diet-related DCM.

## Methods

**AAFCO Protocol:** Eight healthy adult English Pointer dogs between the ages of 1 – 5 years old were used as the control group, and eight healthy Pointer dogs between the age of 1 – 5 years old were used in the test group. All dogs starting the protocol passed an initial physical examination by a veterinarian. The control group was fed a diet containing rice and oats, Purina One Smart Blend Chicken and Brown Rice canned, exclusively throughout the trial, and the test group was fed Freshpet Vital Grain Free Complete Meal Chicken, Beef, Salmon & Egg Recipe throughout the trial. See Table 1 for ingredient list and analysis. The control group diet<sup>a</sup> had previously passed an AAFCO adult maintenance feeding

protocol. The diets were the sole source of nutrients, except for water which was available at all times. All dogs were fed based on their current weight and energy needs and were kept within 15% of starting weights. To determine food intake, all food was weighed before and after each feeding and the difference determined. The feeding trial was run for 26 weeks, with individual body weight measured and recorded at the beginning, weekly, and at the end of the trial. To pass the feeding trial, no individual dog shall lose more than 15% of its initial body weight, and the average percent body weight change (final compared to initial) shall not be less than 10%.

An AAFCO Adult Maintenance trail requires hemoglobin, packed cell volume (PCV), albumin, and alkaline phosphatase be measured and recorded at the end of the feeding trial. The average final hemoglobin, PCV, albumin and alkaline phosphatase shall not be less the:

1. Hemoglobin – 14.0 g/dL (no individual < 12.0 g/dL)
2. PCV – 42% (no individual < 36%)
3. Albumin – 2.8 g/dL (no individual < 2.4 g/dL)
4. Alkaline Phosphatase – 150 IU/L (no individual >300 IU/L)

Whole blood was drawn at the end of the study for hemoglobin, PCV, Albumin and Alkaline phosphatase analysis. For taurine analysis, an additional 2.0 ml of whole blood was drawn with 1 ml placed in sodium heparin tube, and 1 ml placed in sodium heparin tube and centrifuged with the heparin plasma placed in another clean tube. Both whole blood and plasma tubes were immediately submitted to the Amino Acid Analysis Laboratory at the University of California/Davis for whole blood and plasma taurine analysis.

## Results

Table 2A lists the baseline and weekly weight change from baseline weights for the 8 dogs in the control and test group, and the average weekly weight change for both dog groups.

### **2A. Control Group:** Baseline weight (kg) and weekly weight change from baseline weight (%)

Dog	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7	Wk8	Wk9	Wk10	Wk11	Wk12	Wk13	Wk14
<b>1</b>	23.5	-3.4	-1.7	0	0.8	2.5	4.2	2.5	1.7	1.2	2.1	-0.85	-1.7	-1.7
<b>2</b>	22.5	-6.6	-4.4	-8.0	-3.5	0.8	2.2	1.3	5.7	6.2	2.6	-0.4	-2.2	-1.7
<b>3</b>	20.5	-2.9	-3.4	-1.4	-0.9	-1.9	1.4	1.4	1.9	1.4	0	-1.4	-2.4	-2.9
<b>4</b>	21.5	-6.0	-0.9	-3.2	-2.7	-2.3	-2.7	-3.7	-0.4	-0.4	2.3	-0.9	-2.3	-3.7
<b>5</b>	24.8	-9.2	-10.0	-9.6	-5.6	-2.8	-0.8	-4.0	5.2	5.2	4.8	-0.8	-2.8	-3.6
<b>6</b>	27.2	-5.8	-6.6	-6.9	-4.4	-1.4	-1.4	1.1	2.5	2.5	1.4	1.1	-4.0	-5.1
<b>7</b>	33.8	-4.4	-5.9	-7.3	-6.5	-4.4	-2.9	-3.8	-2.9	-1.4	-0.5	-0.5	-1.4	-2.0
<b>8</b>	23.3	-5.5	-8.5	-8.5	-6.0	-2.1	-0.4	0.4	3.4	4.7	4.7	-0.4	-0.4	-1.2

Dog	Wk15	Wk16	Wk17	Wk18	Wk19	Wk20	Wk21	Wk22	Wk23	Wk24	Wk25	Wk26
<b>1</b>	-2.9	-3.4	-3.8	-2.5	-2.9	-1.7	0.8	0	2.5	3.8	4.2	3.8
<b>2</b>	-1.7	-2.6	-2.2	-0.4	0.4	0	-0.8	-1.7	-2.6	-4.0	-0.8	-0.8
<b>3</b>	-3.4	-3.9	-3.9	-3.4	-1.9	-2.4	1.9	3.9	6.8	8.7	10.2	12.1
<b>4</b>	-3.2	-0.4	0.9	2.3	0.9	2.3	6.0	7.4	9.7	8.3	9.7	8.3
<b>5</b>	-4.4	-3.6	-2.0	-2.0	-2.4	0	0.8	4.0	5.2	6.4	13.3	12.9
<b>6</b>	-5.1	-5.8	-4.4	-1.8	-0.3	-0.3	1.8	1.4	2.5	1.4	3.3	2.9

<b>7</b>	-1.7	-2.3	-2.9	-2.6	-2.3	0.5	2.9	2.9	4.4	4.4	5.0	4.1
<b>8</b>	-2.1	-3.0	-3.8	-1.7	-0.8	0.8	4.7	4.2	6.0	6.0	5.5	4.2

**2B. Test Group:** Baseline weight (kg) and weekly weight change from baseline weight (%)

Dog	Wk1	Wk2	Wk3	Wk4	Wk5	Wk6	Wk7	Wk8	Wk9	Wk10	Wk11	Wk12	Wk13	Wk14
<b>1</b>	24.7	-0.8	0.4	0.8	0.4	-0.8	1.2	1.2	-0.4	-1.2	-0.4	0	-0.4	-0.4
<b>2</b>	20.0	-3.5	-7.0	-10.0	-4.5	1.5	4.0	4.0	6.0	4.5	0.5	1.5	-1.5	-0.5
<b>3</b>	15.4	-3.2	-5.8	-5.1	2.5	4.5	10.3	10.3	12.3	10.3	9.0	2.5	1.9	-4.5
<b>4</b>	20.7	-1.4	-4.3	-2.8	-2.8	0	2.8	1.9	6.2	3.3	-0.9	-0.9	-3.3	-3.8
<b>5</b>	23.7	-0.4	-2.5	-1.2	-0.4	2.9	4.2	3.7	5.4	4.2	2.9	-1.6	-1.6	-4.6
<b>6</b>	30.3	-1.9	-2.3	-2.6	0.3	3.9	4.9	4.6	6.6	5.9	5.6	2.3	1.3	1.3
<b>7</b>	32	-2.1	-3.1	-3.1	-0.6	1.5	1.5	1.5	3.1	1.8	0.6	-1.5	-0.6	-0.3
<b>8</b>	24.8	-1.2	-3.6	-4.4	-2.0	-0.4	2.4	4.0	6.0	5.6	4.8	1.6	0.8	-1.2

Dog	Wk15	Wk16	Wk17	Wk18	Wk19	Wk20	Wk21	Wk22	Wk23	Wk24	Wk25	Wk26
<b>1</b>	0	-0.4	-0.8	-0.4	-0.4	-0.8	-1.2	-1.6	-0.8	-0.4	1.2	2.0
<b>2</b>	-2.5	-2.5	-2	-1	-1	0	-2.5	-4	-2	-2.5	0.5	0.5
<b>3</b>	-5.1	-3.8	-4.5	-3.2	-3.2	-3.2	2.5	-0.6	3.2	4.5	3.8	3.2
<b>4</b>	-4.3	-2.8	-3.8	-2.8	-3.3	-2.8	1.9	3.8	6.7	9.1	10.1	9.6
<b>5</b>	-3.7	-2.9	0	-1.6	-1.6	-0.4	1.6	2.5	2.1	2.5	1.6	1.2
<b>6</b>	-0.9	-2.6	-3.3	-2.6	-3.3	-0.6	0.9	2.3	4.2	4.2	3.9	2.6
<b>7</b>	-1.8	-1.2	-1.2	0.6	0.6	1.8	2.5	1.8	1.8	1.2	1.2	0.6
<b>8</b>	-2.8	-2.4	-1.6	0	0	0.8	0.8	0.8	1.6	0.8	0	0

**Table 2B** Average percentage (%) weight change over the 26-week period for both the control group dogs and test group dogs.

Dog	Control group	Test group
1	0.15	-0.16
2	-1.03	-0.96
3	0.54	1.55
4	1.00	0.59
5	-0.24	0.48
6	-1.26	1.39
7	-1.28	0.27
8	-3.55	0.41

All 16 dogs completed the 26-week feeding trial. No individual dog lost more than 15% of its initial body weight and the average body weight change was not less than 10 % in both the control and test groups.

Table 3A and B lists the ending blood values for control and test group dogs.

**3A. Control group:** Ending hemoglobin, hematocrit (PCV), albumin, and alkaline phosphatase

Dog	Hemoglobin	PCV	Albumin	Alk Phos
1	17.5	52.3	3.2	41
2	18.6	55.3	3.7	33
3	17	51.5	3.5	48
4	20.3	57.5	3.5	27
5	17.2	51.5	3.3	46
6	18.6	54.2	3.4	27
7	18	54.1	3.2	55
8	16.9	51.6	3.5	32
<b>Average</b>	<b>18.01</b>	<b>53.50</b>	<b>3.41</b>	<b>38.62</b>

**3B. Test group:** Ending hemoglobin, hematocrit (PCV), albumin, and alkaline phosphatase

Dog	Hemoglobin	PCV	Albumin	Alk Phos
1	19.3	58	3.3	37
2	17.2	51.5	3.6	38
3	18.9	56.4	3.6	23
4	17.6	51.8	3.2	26
5	18.6	54.8	3.5	49
6	18.7	55	3.4	40
7	17.7	52.3	3.2	27
8	18.5	54.7	3	51
<b>Average</b>	<b>18.31</b>	<b>54.31</b>	<b>3.35</b>	<b>36.37</b>

All blood values within the test group were within normal reference ranges. Since all blood value within the parameters determined by AAFCO for adult maintenance, the test diet passed this portion of the protocol.

Table 4A, and B lists normal taurine values for dogs, and the ending plasma and whole blood taurine levels for the test group dogs and control group dogs.

### Normal Taurine Values (nMol/ml) for Dogs

Plasma		Whole Blood	
Normal range	No Known sign of deficiency	Normal range	No Known sign of deficiency
60-120	>40	200-350	>150

**Test group (Freshpet):** Ending plasma and whole blood taurine levels

Dog	Plasma taurine	Whole blood taurine
1	155	331
2	130	298
3	150	348
4	115	332
5	94	265
6	108	278
7	88	258
8	114	298
<b>Average</b>	<b>119.25</b>	<b>301.0</b>

All plasma and whole blood taurine levels in both groups were within normal reference ranges for dogs.

### Summary

The relatively recent findings that dogs eating “grain-free” diets have an increase risk for dilated cardiomyopathy has caused great concern in the veterinary community, pet food industry and dog-owners. The FDA’s Center for Veterinary Medicine (CVM) and the Veterinary Laboratory Investigation and Response Network (Vet-LIRN), a collaboration of government and veterinary diagnostic laboratories, continue to investigate this potential association. It is believed that the potential association between diet and DCM in dogs is a complex scientific issue that may involve multiple factors.

Taurine is generally not considered an essential amino acid for dogs, because these animals can synthesize taurine from cysteine and methionine. Taurine has important osmoregulation and cardiac contractility functions. It is still unknown how of if taurine deficiency or metabolism may have a role in these reports of canine dilated cardiomyopathy. Current recommendations include whole blood and plasma taurine concentrations be measured in dogs with DCM being fed a grain-free diet, and a dietary taurine supplement should be added if concentrations are found to be low or low-normal. In addition, recommendations include feeding dogs and cats a diet that has undergone extensive feeding trials through the Association of American Feed Control Officials (AAFCO).

FreshPet’s Vital Grain Free Complete Meal Chicken, Beef, Salmon & Egg successfully passed all parameters of the rigorous AAFCO Adult Maintenance feeding trial. In addition, all the dogs fed the FreshPet Vital Grain Free had normal plasma and whole blood taurine levels.

- a. Purina One Smart Blend Chicken and Brown Rice Canine canned. Nestle Purina PetCare. St. Louis USA

Table 1. Calculated dietary composition (g/1000 kcal) and metabolizable energy (ME)

Item	Control Diet (Purina ONE)	Test Diet (FP Vital Complete)
ME (kcal/g)	1.167	1.720
Protein	68.55	98.83
Fat	59.98	58.13
Carbohydrate	22.87	11.62
Moisture (%)	78%	65%

Ingredients:

Control diet: Chicken, chicken broth, liver, pork lungs, brown rice, oat meal, carrots, spinach, egg product, potassium chloride, salt, calcium carbonate, zinc sulfate, ferrous sulfate, copper sulfate, manganese sulfate, potassium iodide, sodium selenite, carrageenan, guar gum, locust bean gum, Vitamin E, Vitamin B-3, Vitamin B-1, Vitamin B-5, Vitamin B-6, Vitamin B-12, Vitamin B-2, Vitamin A, folic acid, Vitamin D-3, Vitamin B-7

Test diet: chicken, chicken liver, beef, salmon, pea protein, eggs, natural flavors, cranberries, spinach, pea fiber, salt, carrageenan, vinegar, beta-carotene, potassium chloride, celery powder, choline chloride, Vitamin E supplement, niacin, calcium pantothenate, biotin, riboflavin, thiamine mononitrate, Vitamin B12 supplement, Vitamin D supplement, pyridoxine hydrochloride, folic acid, zinc proteinate, iron proteinate, manganese proteinate, copper proteinate, sodium selenite, calcium iodate

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