Dietary Supplementation with (n-3) Polyunsaturated Fatty Acids Does Not Affect Insulin Sensitivity in Healthy Labrador Retriever Dogs 1,2

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EXPANDED ABSTRACT

KEY WORDS: • (n-3) PUFA • insulin sensitivity • dogs

Obesity in dogs (>15% recommended body weight) is becoming increasingly common. It has been estimated to affect between 11 and 44% of the canine population, and has been linked to overfeeding and a sedentary lifestyle (1,2). There also appears to be a genetic propensity to obesity in certain purebred dogs, such as Labrador Retrievers, who have an increased risk of developing obesity (3). It is well established from human studies that obesity is a major risk factor for developing insulin resistance and diabetes. As with human subjects, obesity is associated with various conditions in the dog and it is considered the single most important contributory factor in the development of diabetes (4).

Dietary supplementation in humans with (n-3) polyunsaturated fatty acids [(n-3) PUFA4] has been shown to increase insulin sensitivity, by reducing the fasting insulin:glucose ratio and increasing glucose clearance rates (5). The present study was therefore carried out to investigate whether dietary supplementation with (n-3) PUFA was effective in improving insulin sensitivity in the healthy Labrador Retriever dog.

MATERIALS AND METHODS

Six healthy nonobese Labrador Retriever dogs [mean age 5 (SD 2.5) y] were studied. They were placed on a standard maintenance diet: [kJ metabolic energy/d = 460 × body weight (kg)0.75], enriched with (n-3) PUFA in the form of marine fish oil containing 0.08 g eicosapentaenoic acid (EPA) and 0.06 g docosahexaenoic acid (DHA)/100 g, for 6 mo. During an equivalent control period, the dogs were fed the standard diet without marine fish oil supplementation. Dogs received similar exercise levels throughout the study. At the end of each dietary period, insulin sensitivity was assessed with an intravenous glucose tolerance test (IVGTT). After an overnight fast, two venous cannulae were inserted into cephalic veins in the right and left forelimbs. Venous blood was collected for the measurement of plasma glucose and insulin concentrations, before and at frequent intervals for 120 min following an intravenous bolus injection of glucose (300 mg glucose/kg body weight, administered as a 50% w/v solution over 30 s). Indices of insulin sensitivity (SI) and glucose effectiveness (SG) were calculated using the glucose–insulin minimal model adapted from Bergman (6). The insulinogenic index (ΔΙ/ΔG) was calculated from the maximum increment of glucose and insulin above fasting levels. Glucose fractional turnover rate (kIVGTT) was calculated as minus the gradient of the best-fit straight line of natural log glucose against time between 15 and 45 min following glucose administration. Incorporation of (n-3) PUFA into plasma membranes was assessed by measuring fatty acid profiles from red blood cells, collected at the end of each dietary period. Differences between dietary treatments were compared using the paired Student’s t-test; data were examined beforehand to ensure they satisfied the criteria for parametric analyses. The study complied with the Guide for the Care and Use of Laboratory Animals (7) and conformed to the guidelines of the Waltham Ethical Review Committee.

RESULTS

Red blood cell fatty acid profiles were significantly affected by the (n-3) PUFA diet (Fig. 1). There was a significant increase in membrane concentrations of EPA (C20:5) (P < 0.01) and DHA (C22:6) (P < 0.01) with (n-3) PUFA dietary supplementation. (n-3) PUFA supplementation was also associated with a smaller but significant decrease in palmitic acid (C16:0) (P = 0.039) and increase in stearic acid (C18:0) (P = 0.029). However, total saturated fatty acids (palmitic + stearic) were unchanged by the dietary intervention. None of the parameters of insulin sensitivity measured

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4 Abbreviations used: DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; IVGTT, intravenous glucose tolerance test; kIVGTT, glucose fractional turnover rate; PUFA, polyunsaturated fatty acids; SI, insulin sensitivity; SG, glucose effectiveness; SI, insulin sensitivity.
was affected by the (n-3) PUFA-supplemented diet (Table 1). There were no significant differences in fasting plasma glucose or insulin, the insulinogenic index, $k_{IVGTT}$, $S_1$, or $S_G$. However, one dog who demonstrated marked hyperinsulinemia compared to the rest of the group and a degree of insulin resistance had greatly improved insulin sensitivity on the (n-3) PUFA-supplemented diet ($S_1$ on control diet, $0.53 \times 10^{-4}$ min$^{-1}$ pM$^{-1}$; on (n-3) PUFA-supplemented diet, $0.94 \times 10^{-4}$ min$^{-1}$ pM$^{-1}$).

### DISCUSSION

Dietary supplementation with (n-3) PUFA at a level of 1.4 g/kg diet was effective in altering membrane fatty acid composition. One mechanism whereby (n-3) PUFA dietary supplementation has been suggested to affect insulin sensitivity is by incorporating into cell membranes and increasing membrane fluidity (8). However, in spite of the significantly higher incorporation of DHA and EPA into red blood cell membranes, no effect of (n-3) PUFA supplementation was observed for any of the parameters of insulin sensitivity measured. The lack of any significant change in insulin sensitivity may be attributed to the fact that dogs that were already relatively insulin sensitive were used for this study. Current evidence in other species collectively indicates that consumption of (n-3) PUFA, particularly the 20- and 22-carbon fatty acids, could play a preventative role in the development of insulin resistance (9). In rats, 6% marine fish oil supplementation has been shown to be effective in this respect, but ineffective in reversing sucrose-induced insulin resistance (10). Some, but not all, human studies suggest that marine fish oil ingestion by patients who have already developed non-insulin-dependent diabetes may actually exacerbate insulin resistance (10). In this context, it is therefore interesting to note that a single dog who had a degree of insulin resistance had improved insulin sensitivity on the (n-3) PUFA-supplemented diet.

We conclude that dietary supplementation with (n-3) PUFA at the level given in this study is ineffective in further improving insulin sensitivity in healthy, nonobese insulin-sensitive dogs. There is a suggestion that such a diet might be beneficial in more insulin-resistant animals, but further studies are required to confirm this.

### LITERATURE CITED


### TABLE 1

Minimal model and non-model-derived measurements for insulin sensitivity from six dogs fed a diet supplemented with 1.4 g (n-3) PUFA/kg diet$^1$

<table>
<thead>
<tr>
<th>Index</th>
<th>Control diet</th>
<th>(n-3) PUFA diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insulinogenic index ($\times 10^{-9}$)</td>
<td>47.8 ± 15.2</td>
<td>42.4 ± 9.63</td>
</tr>
<tr>
<td>$k_{IVGTT}$ (%/min)</td>
<td>2.30 ± 0.21</td>
<td>2.50 ± 0.32</td>
</tr>
<tr>
<td>$S_1$ ($\times 10^{-4}$ min$^{-1}$ pM$^{-1}$)</td>
<td>0.88 ± 0.21</td>
<td>1.12 ± 0.34</td>
</tr>
<tr>
<td>$S_G$ ($\times 10^{-2}$ min$^{-1}$)</td>
<td>3.81 ± 0.86</td>
<td>2.84 ± 0.69</td>
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</table>

$^1$Values are mean ± SEM. There were no significant differences between the diet groups.