# Fat/Water $^1$H MRI to Investigate Effects of Leptin in ob/ob Mice

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## Introduction:

Leptin, a protein hormone, is an important component in the regulation of body weight. Administration of leptin leads to weight loss due to decreased hunger and food consumption and increased energy expenditure[1]. Our goal in this study was to evaluate the effects of exogenously administered leptin on fat and water in leptin-deficient obese mice by $^1$H MRI.

## Methods:

*Animal model*: Twelve obese leptin-deficient mice (C57BL/∧OlaHsd-*Lep*; ob/ob) were sc-implanted with Alzet osmotic mini-pumps containing either vehicle (n=6) or leptin (0.3 mg/kg/day, n=6). The infusions lasted for seven days. Fat and water MRI of mice were obtained one day prior to implanting the osmotic pumps and then again after seven days of treatment.

*MR parameters*: MR imaging was performed on a 9.4T, 31cm diameter horizontal bore magnet interfaced to a Varian console. Multi-slice spin-echo transaxial images (TR/TE = 2,000/12 ms) with a chemical shift selective (CHESS) technique[2] for fat and water suppression were obtained using a 38 mm diameter birdcage RF coil. Twenty four slices of fat and water images with 2 mm slice thickness and 0.5 mm slice gap were collected with 40 x 40 mm FOV. Two 5-mm diameter reference tubes, one tube containing water and the other containing baby oil, were placed next to the animal. The reference tubes were used for ensuring adequate fat or water suppression and for signal intensity (SI) referencing.

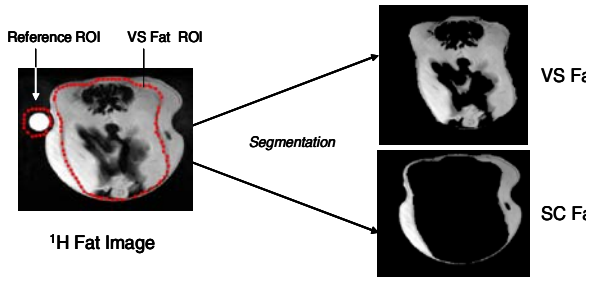


Figure1. Segmentation of 1H Fat Image into VS and SC fat regions

*Data Processing*: Three consecutive slices between the kidneys and the hind legs were selected to determine VS and SC fat volumes and average fat signal intensity in the abdominal region. VS and SC areas were separated in the fat and water images by drawing ROIs and using image processing techniques such as thresholding and image subtraction (Figure 1). Average fat signal intensity (SI) in the VS and SC regions were computed relative to the SI from the oil reference. Liver fat-to-water SI ratio was determined from the average fat and water SI from two rectangular ROIs in the liver normalized with respect to reference SI.

## Results:

Table 1 shows the effect of leptin on animal body weight, VS and SC fat volume in the abdominal region and fat-to-water ratio in the liver. There was a significant decrease in the body weight and daily food intake in leptin treated mice compared to the control. VS fat volume in the abdominal region increased over seven days in control mice while it decreased in leptin treated animals. Average fat SI in the VS region also decreased significantly in leptin treated animals. There was no significant change in SC fat volume in both control or leptin treated mice. Average fat SI in the SC region showed a slight decrease in the control group but did not change significantly in the treated animals. Fat-to-water ratios were decreased in both VS and SC regions in the treated animals. Liver fat-to-water ratio and average SI were significantly reduced in leptin treated animals compared to control animals.

Table 1. Effect of leptin on body weight, VS and SC fat volume and liver fat-to-water SI ratio.

| | CONTROL (n=6) | | TREATED (n=6) | |
|---|---|---|---|---|
| | Before Therapy | After Therapy | Before Therapy | After Therapy |
| Body weight (g) | 46.72 ± 0.89 | 50.02 ± 0.93$^{\dagger}$ | 46.85 ± 0.84 | 43.9 ± 0.81$^{*\dagger}$ |
| VS fat volume (cc) | 2.77 ± 0.04 | 3.06 ± 0.11$^{\dagger}$ | 2.6 ± 0.1 | 2.18 ± 0.10$^{*\dagger}$ |
| SC fat volume (cc) | 1.12 ± 0.16 | 0.98 ± 0.11 | 1.08 ± 0.09 | 1.13 ± 0.20 |
| Liver fat-to-water SI ratio | 2.17 ± 0.41 | 1.98 ± 0.12 | 2.05 ± 0.07 | 1.03 ± 0.06$^{*\dagger}$ |

Values are reported as mean ± SE ; * : $p \leq 0.05$ (control vs. treated); † : $p \leq 0.05$ (before vs. after therapy)

## Conclusions:

Among all the parameters measured the most significant decrease was seen in hepatic fat-to-water ratio with leptin treatment in ob/ob mice. Significant decreases in VS fat volume and average fat SI were also observed in the abdominal region with leptin treatment. SC fat volume and average fat SI in the abdomen do not appear to be good markers for monitoring effects of leptin therapy. $^1$H MRI provides a robust method for monitoring the compartmental changes in fat and water content in small animal models. Simple fat and water MR imaging may prove very useful for evaluating new drugs for the treatment of obesity and other metabolic disorders.

**References:** 1. Pellymounter et al., Science, 269:540-3, 1995; 2. Haase et al., Phys Med Biol, 30(4):341-4, 1985.