Chapter 6

Leptin deficiency per se dictates body composition, insulin action and insulin clearance in ob/ob mice.

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Abstract
Many obese humans appear to be both insulin- and leptin resistant. Since leptin may impact glucose metabolism directly, it is conceivable that lack of leptin signal transduction critically contributes to insulin resistance in these individuals. Furthermore, at this time, it remains unclear if leptin affects glucose metabolism via peripheral and/or central mechanistic routes. To explore the contribution of cerebral leptin deficiency to insulin resistance in leptin deficient ob/ob mice, we infused leptin i.c.v. in ob/ob mice. To also evaluate the impact of adiposity on insulin action in these animals, another group of young ob/ob animals were subjected to severe calorie restriction, so that their body weight became similar to that of wild type mice. Hyperinsulinemic euglycemic clamps and isotope dilution techniques were used to determine insulin sensitivity of hepatic glucose production (HGP) and disposal (GD).

Leptin infusion (i.c.v., 2.5 µg/h for 3 hours) acutely increased both the hepatic insulin sensitivity index and glucose disposal index (9.1±2.4. vs. 5.0±2.7 %·nmol⁻¹ and 25.6±5.6. vs. 13.6±4.8 µmol·min⁻¹·kg⁻¹·nmol⁻¹, respectively; P<0.05 for both comparisons) in ob/ob mice. Furthermore, i.c.v. leptin administration acutely reduced circulating insulin levels during continuous insulin infusion.

Food restriction barely affected body composition although it profoundly curtailed body weight. Insulin suppressed HGP clearly more in the lean ob/ob mice than in their obese counterparts, but its impact remained less than in wild-type mice (% suppression: 11.8±8.9 vs. 1.3±1.1 vs. 56.6±13.0 %·nmol⁻¹, respectively; P<0.05). The glucose disposal index of lean ob/ob mice was also in between that of obese ob/ob and lean wild-types (37.5±21.4 vs. 25.1±14.6 vs. 59.6±17.3 µmol·min⁻¹·kg⁻¹·nmol⁻¹, respectively; P<0.05 wild-type mice vs. ob/ob mice). In conclusion, leptin deficiency per se is not just responsible for hyperphagia and obesity in ob/ob mice, but critically determines body composition, insulin action and insulin clearance from the circulation in these animals. The data suggest that leptin resistance in obese humans may contribute to insulin resistance and hyperinsulinemia in these individuals.

Introduction
Leptin conveys signals pertaining to the size of bodily energy stores to the brain, where it orchestrates behavioral and metabolic adaptations meant to maintain energy
balance in the face of environmental fluctuations in nutrient availability. Leptin may also affect body composition, as suggested by the fact that it specifically reduces fat mass (leaving lean body mass untouched) in rodents.

Leptin deficiency produces severe obesity, insulin resistance and impaired glucose tolerance in ob/ob mice. The ratio of adipose over lean tissue is significantly elevated in these animals. Since in vivo measures of insulin sensitivity correlate strongly with total and regional fat mass in animals and humans, it is tempting to attribute at least part of the metabolic anomalies of ob/ob mice to their altered body composition. However, there is also evidence to suggest that leptin impacts glucose metabolism independently of its effects on food intake and body weight. Accordingly, intraperitoneal administration of leptin acutely reduces glycemia and insulinemia and restores glucose tolerance without affecting body weight in ob/ob mice. Injection of a low dose of leptin into the ventromedial hypothalamus of lean rats promotes basal (insulin independent) glucose uptake in various tissues, suggesting that the central nervous system is a critical target of leptin in the control of glucose metabolism. Leptin deficient ob/ob mice are an ideal model to study the (metabolic) impact of leptin, as exogenously administered leptin inevitably competes with endogenous peptide in wild-type animals. We therefore used ob/ob mice to evaluate the effects of i.c.v. leptin infusion on insulin sensitivity. To determine the simultaneous impact of the obese phenotype of these animals on insulin action, we also subjected young ob/ob animals to severe calorie restriction, so that their body weight became similar to that of wild type mice. In both experimental settings, hyperinsulinemic euglycemic clamps and isotope dilution techniques were used as measures of in vivo insulin action on glucose metabolism.

Research designs and methods

Animals. Wild-type and ob/ob mice were obtained from our breeding colony of C57BL6 ob+/- mice. Mice were individually housed in a temperature-controlled room on a 12:12h light-dark cycle and were fed a standard mouse chow diet with free access to water. All animal experiments were performed in accordance with the regulations of Dutch law on animal welfare and the institutional ethics committee for animal procedures approved the protocol.
I.c.v. leptin experiment. Male ob/ob mice were anesthetized with 0.5 mg/kg Medetomidine (Pfizer, Capelle a/d IJssel, The Netherlands), 5 mg/kg Midazolam (Roche, Mijdrecht, The Netherlands) and 0.05 mg/kg Fentanyl (Janssen-Cilag, Tilburg, The Netherlands). A 25-gauge guide cannula was stereotaxically implanted into the left lateral ventricle using the following coordinates from Bregma: 0.46 mm posterior, 1.0 mm lateral and 2.2 mm ventral. The guide cannula was secured with dental cement (Oral Hygiene Center BV, Amersfoort, The Netherlands) to the skull surface. After a recovery period of 1 week, adequate placement of the cannulae was tested with the feeding response to an i.c.v. injection of NPY (5 µg dissolved in 1 µl of sterile water; Bachem, Bubendorf, Germany). After overnight fasting, a hyperinsulinemic euglycemic clamp experiment was performed under 6.25 mg/kg Acepromazine (Sanofi sante animale, Libourne Cedex, France), 6.25 mg/kg Midazolam (Roche, Mijdrecht, The Netherlands) and 312.5 µg/kg Fentanyl (Janssen-Cilag, Tilburg, The Netherlands) anesthesia. During the entire experiment (basal and hyperinsulinemic period) leptin (1 µg/µl; Bachem, Bubendorf, Germany) or vehicle was infused into the left lateral ventricle at a rate of 2.5 µl/h.

Food restriction experiment. Male and female mice were divided into the following experimental groups: 1) ad libitum-fed ob/ob mice, 2) ob/ob mice food restricted to 55% of the ad libitum wild-type food intake, 3) ad libitum-fed wild-type mice. Three nights preceding the experiments, all groups were food restricted for two nights to 55% of the wild-type food intake (this was done to avoid potential bias of the results attributable to differences in daily caloric intake) followed by overnight fast. The next morning, a hyperinsulinemic euglycemic clamp was performed under 0.5 ml/kg Hypnorm (Janssen pharmaceutica, Beerse, Belgium) and 12.5 mg/kg Midazolam (Roche, Mijdrecht, The Netherlands) anesthesia.

Hyperinsulinemic euglycemic clamp. Hyperinsulinemic clamps were performed at 9.00 a.m. as described earlier. Basal rates of glucose turnover were determined by administering a primed (P) continuous (C) infusion (P: 0.8 µCi, C: 0.02 µCi/min) of 3-3H-glucose (Amersham, Little Chalfont, U.K.). After 60 min, insulin was given as a prime (10 mU) followed by continuous infusion of 0.25 mU/min. A variable infusion of 12.5% D-glucose was used to maintain euglycemia (measured at 10 min intervals via tail bleeding, Freestyle, TheraSense, Disetronic Medical Systems BV, Vianen, The Netherlands). Blood samples (60 µl) were taken at the end of the basal period and during the clamp (40 and 20 min before and by the end of the clamp) for
determination of 3-\(^3\)H-glucose specific activity and plasma insulin, glucose and free fatty acid (FFA) concentrations. At the end of the clamp, mice were sacrificed and tissue samples were taken within 5 min, frozen immediately in liquid nitrogen and stored at \(-20^\circ\text{C}\) for subsequent analysis. Carcasses were stored at \(-20^\circ\text{C}\) for body composition analysis.

**Analytical procedures.** Plasma levels of glucose and free fatty acids (FFAs) were determined using commercially available enzymatic kits (Sigma, St. Louis, MO; Sigma, St. Louis, MO and Wako Chemicals, Neuss, Germany, respectively). Plasma insulin concentration was measured by radioimmunoassay (Linco Research Inc., St. Charles, MO). Total plasma 3-\(^3\)H-glucose was determined in 7.5 \(\mu\text{l}\) plasma and in supernatants after trichloroacetic acid (20\%) precipitation and water evaporation to eliminate tritiated water. Content of triglycerides (TG) in liver and muscle tissue was determined as described before\(^{15}\). Briefly, 10-20 \(\mu\text{g}\) of tissue was homogenized in phosphate buffered saline (PBS) and samples were taken for measurement of protein content\(^ {16}\). Lipids were extracted and TG fraction was separated from the other lipid components by high performance thin layer chromatography (HPTLC) on silica gel plates.

**Body composition analysis.** Carcasses were dehydrated at \(65^\circ\text{C}\) until constant mass was achieved and hydrolyzed in 100 ml ethanolic potassium hydroxide (3M in 65\% ethanol). Aliquots were taken, diluted with PBS and used for determination of body triglyceride content (as detailed in\(^ {17,18}\) by enzymatic measurement of glycerol (Sigma, St. Louis, MO).

**Calculations.** Turnover rates of glucose (\(\mu\text{mol/min/kg}\)) were calculated during the basal period and in steady-state clamp conditions as the rate of tracer infusion (dpm/min) divided by the plasma specific activity of 3-\(^3\)H-glucose (dpm/\(\mu\text{mol}\)). The ratio was corrected for lean body mass. Hepatic glucose production (HGP) was calculated as the difference between the tracer-derived rate of glucose appearance and the glucose infusion rate.

The hepatic insulin sensitivity index was calculated as the ratio of the relative suppression of HGP during the hyperinsulinemic condition to plasma insulin levels during hyperinsulinemic conditions. The glucose disposal index was calculated as the ratio of glucose disposal to plasma insulin levels during hyperinsulinemic conditions.
Statistical analysis. Differences between groups were determined by Kruskall-Wallis and Mann-Whitney nonparametric tests for independent samples. A P-value < 0.05 was considered statistically significant. All values shown represent means ± SD.

Results

1.c.v. leptin experiment.

Animals. Ob/ob mice were 4 months old at the time of the experiments. Body weight was 44.6 ± 6.7 gram in the control group and 44.7 ± 2.2 gram in the leptin group.

Plasma parameters. Plasma glucose, FFA and insulin concentrations in basal and hyperinsulinemic conditions are shown in Table 1. In the basal state, plasma parameters did not differ between leptin- and vehicle-infused animals. In steady state clamp conditions, plasma glucose and FFA concentrations were similar in both groups. However, circulating insulin levels were significantly lower in leptin-infused compared to vehicle-infused animals, despite equal insulin infusion rates.

Glucose turnover. Hyperinsulinemia suppressed the hepatic glucose production to a different extent in both groups. To correct for disparate insulin levels during the clamp, we calculated the hepatic insulin sensitivity index. This index was significantly higher in leptin-infused animals compared to vehicle-infused animals (9.1 ± 2.4. vs. 5.0 ± 2.7 %·nmol⁻¹, respectively; P<0.05, Fig. 1).

Figure 1. Percentual inhibition of the endogenous glucose production (EGP) per nmol insulin determined by hyperinsulinemic clamp in ob/ob mice that received an icv infusion of vehicle or leptin. Values represent the mean ± SD of at least 5 mice per group. * P < 0.05 vs. vehicle.
Hyperinsulinemia increased glucose disposal in both groups. However, the glucose disposal index was significantly higher in leptin-infused- compared to vehicle-infused animals (25.6 ± 5.6. vs. 13.6 ± 4.8 μmol·min⁻¹·kg⁻¹·nmol⁻¹, respectively; P<0.05, Fig. 2).

**Glucose disposal**

*Figure 2.* Insulin mediated glucose disposal (Rd) per nmol insulin as determined by hyperinsulinemic euglycemic clamp in obese ob/ob mice that received an icv infusion of vehicle or leptin. Values represent the mean ± SD of at least 5 mice per group. *P* < 0.05 vs. vehicle.

**Food restriction experiment.**

**Body weight.** Food restriction of ob/ob mice was initiated at the age of 2 months to keep them lean. However, even at 2 months, ob/ob mice were heavier than wild-type mice (Fig. 3). After 4 months of severe food restriction (55% of wild-type intake), body weight of ob/ob mice was similar to that of wild-type mice, while body weight of ad libitum-fed ob/ob mice was clearly higher.

*Figure 3.* Growth curves of ad libitum-fed ob/ob mice (obese ob/ob), food restricted ob/ob mice (lean ob/ob) and ad libitum-fed wild-type mice (wt). At 2 months of age ob/ob mice were food restricted to 55% of ad libitum wild-type food intake to keep them lean. Values represent the mean ± SD of at least 5 mice per group.
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**Body composition.** Body weight, lean body mass and the amount of water and lipid of lean ob/ob, obese ob/ob and wild-type mice are shown in Table 2. Although body weight of lean ob/ob mice was similar to that of wild-type animals, their body composition (lipid and lean mass expressed as percentage of body weight) was more like that of ad libitum fed ob/ob counterparts. Lean body mass of lean ob/ob mice was significantly lower than that of obese ob/ob and wild-type animals. Absolute amounts of TG in lean ob/ob mice were significantly higher than in wild-type -, but significantly lower than in obese ob/ob mice.

**Tissue TG.** Hepatic TG content of lean ob/ob mice was very low as compared to obese ob/ob mice and similar to wild-type mice (35.3 ± 16.0, 141.7 ± 51.4, 47.1 ± 35.3 μmol/g protein, respectively; P<0.05 lean ob/ob vs. obese ob/ob, Fig. 4A). TG content of muscle tended to be lower in lean ob/ob mice compared to obese ob/ob mice, but was significantly higher (P<0.05) compared to wild-type mice (173.2 ± 90.2, 257.7 ± 86.7 and 59.0 ± 37.0 μmol/g protein for lean ob/ob, obese ob/ob and wild-type mice respectively, Fig. 4B).

**Plasma parameters.** Plasma glucose, FFA and insulin concentrations under basal and hyperinsulinenic euglycemic clamp conditions are shown in Table 3. In the basal state plasma parameters of lean ob/ob mice were not different from obese ob/ob mice. Both groups of ob/ob mice were hyperinsulinemic as compared to wild-type animals. Obese ob/ob mice had significantly higher plasma glucose concentrations as compared to wild-type mice, while circulating glucose levels in lean ob/ob mice were in between those in obese ob/ob and wild types.

In steady state clamp conditions, circulating insulin levels were significantly higher in ob/ob mice than in wild-type mice, despite equal insulin infusion rates. Glucose levels in steady state were not significantly different between lean and obese ob/ob mice, but glucose levels in wild-type mice were significantly lower than glucose levels in both groups of ob/ob mice. FFA concentrations were diminished during hyperinsulinenemia (as compared to baseline) by ~25% in obese ob/ob, ~50% in lean ob/ob and ~70% in wild-type mice.
Glucose turnover. Hyperinsulinemia suppressed hepatic glucose production to a different extent in the three groups. The hepatic insulin sensitivity index was significantly higher in lean ob/ob mice compared to obese ob/ob mice, but significantly lower compared to wild-type mice (11.8 ± 8.9, 1.3 ± 1.1 and 56.6 ± 13.0 %·nmol⁻¹ for lean ob/ob, obese ob/ob and wild-type mice respectively; P<0.05, Fig. 5).

Insulin infusion enhanced glucose disposal in all groups. The glucose disposal index was significantly reduced in obese ob/ob mice as compared to wild-type, while the index of lean ob/ob mice was in between those of obese ob/ob and lean wild-types (37.5 ± 21.4, 25.1 ± 14.6 and 59.6 ± 17.3 µmol·min⁻¹·kg⁻¹·nmol⁻¹ for the lean ob/ob, obese ob/ob and wild-type mice, respectively; P<0.05 wild-type mice vs. ob/ob mice, Fig. 6).

Figure 4. Triglyceride levels in liver (A) and muscle (B) determined in obese ob/ob, lean ob/ob and wild-type (wt) mice. Values represent the mean ± SD of at least 5 mice per group. * P < 0.05 vs. wt; † P < 0.05 vs. obese ob/ob.
Glucose production

Figure 5. Percentual inhibition of the endogenous glucose production (EGP) per nmol insulin as determined by hyperinsulinemic clamp in obese ob/ob, lean ob/ob and wild-type mice. Values represent the mean ± SD of at least 5 mice per group. * P < 0.05 vs. wt.; † P < 0.05 vs. obese ob/ob.

Glucose disposal

Figure 6. Insulin mediated glucose disposal (Rd) per nmol insulin as determined by hyperinsulinemic euglycemic clamp in obese ob/ob, lean ob/ob and wild-type mice. Values represent the mean ± SD of at least 5 mice per group. * P < 0.05 vs. wt.
Table 1. Plasma parameters in ob/ob mice that received an i.c.v. infusion of vehicle or leptin under basal and hyperinsulenic conditions. Values represent the mean ± SD of at least 5 mice per group. * P < 0.05 vs. vehicle.

<table>
<thead>
<tr>
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<th>Glucose (mmol/l)</th>
<th>FFA (mmol/l)</th>
<th>Insulin (nmol/l)</th>
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<tr>
<td></td>
<td>Basal</td>
<td>Hyperinsulenic</td>
<td>Basal</td>
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<tr>
<td>Vehicle</td>
<td>11.5 ± 4.5</td>
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<td>1.2 ± 0.4</td>
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<td>Leptin</td>
<td>11.2 ± 3.9</td>
<td>9.3 ± 2.2</td>
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Table 2. Body composition of obese ob/ob, lean ob/ob and wild-type (wt) mice. Values represent the mean ± SD of 9 obese ob/ob mice, 12 lean ob/ob mice and 12 wild-type mice. * P < 0.05 vs. wt; † P < 0.05 vs. obese ob/ob.

<table>
<thead>
<tr>
<th></th>
<th>BW (g)</th>
<th>Lean bm (g)</th>
<th>Water (g)</th>
<th>(% of BW)</th>
<th>Lipid (g)</th>
<th>(% of BW)</th>
</tr>
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<tr>
<td>Obese ob/ob</td>
<td>50.3 ± 2.8*</td>
<td>23.8 ± 2.4</td>
<td>14.7 ± 1.6*</td>
<td>29.3 ± 3.1*</td>
<td>26.5 ± 3.8*</td>
<td>52.5 ± 5.6*</td>
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<tr>
<td>Lean ob/ob</td>
<td>29.7 ± 2.5</td>
<td>16.5 ± 2.9*†</td>
<td>10.6 ± 1.4*†</td>
<td>35.9 ± 5.6*†</td>
<td>13.2 ± 2.5*†</td>
<td>44.6 ± 8.0*†</td>
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<td>Wt</td>
<td>26.3 ± 4.6</td>
<td>24.4 ± 3.5</td>
<td>17.7 ± 2.8</td>
<td>67.5 ± 3.6</td>
<td>1.9 ± 1.5</td>
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Table 3. Plasma parameters in obese ob/ob, lean ob/ob and wild-type (wt) mice under basal and hyperinsulinemic conditions. Values represent the mean ± SD of at least 5 mice per group. * P<0.05 vs. wt.

<table>
<thead>
<tr>
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<th>Glucose (mmol/l)</th>
<th>FFA (mmol/l)</th>
<th>Insulin (nmol/l)</th>
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<tr>
<td></td>
<td>Basal</td>
<td>Hyperinsulinemic</td>
<td>Basal</td>
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<tr>
<td>Obese ob/ob</td>
<td>6.6 ± 1.2*</td>
<td>7.2 ± 1.4*</td>
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<tr>
<td>Lean ob/ob</td>
<td>5.5 ± 1.8</td>
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<td>0.8 ± 0.1</td>
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<td>Wt</td>
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Discussion

Here we show that intracerebroventricular administration of minute amounts of leptin acutely ameliorates insulin resistance in $ob/ob$ mice. Chronic calorie restriction, despite curtailing body weight to that of wild type controls, barely affects body composition of these animals. However, it does considerably blunt lipid accumulation in muscle and liver and also clearly reinforces insulin action on glucose production and disposal. As the mice were pair-fed for 3 days prior to the experimental procedures, we believe that the metabolic benefits we report here are primarily due to the constraint of adipose mass and tissue lipid storage, and not to calorie restriction per se. We infer that both leptin deficiency and the obese phenotype contribute to insulin resistance in $ob/ob$ mice. Finally, circulating insulin levels during insulin infusion were consistently higher in $ob/ob$ animals than in normal weight controls and i.c.v. leptin administration acutely reduced these levels, which suggests that leptin promotes (hepatic) insulin clearance via activation of cerebral leptin receptors.

The current literature pertaining to the acute effects of leptin on glucose metabolism is rather confusing. Nevertheless, our finding that i.c.v. leptin administration acutely facilitates insulin action in $ob/ob$ mice corroborates and complements various previous reports. Kamohara and coworkers were the first to show that both intravenous (i.v.) and i.c.v. leptin acutely enhance glucose turnover in wild-type C57/BL6 mice $^{10}$. Subsequently, Sivitz et al demonstrated, that i.v. leptin administration for 48 hours enhances the glucose infusion rate necessary to maintain euglycemia during insulin in normal weight Sprague Dawley rats $^{19}$. Rossetti et al reported that i.v. leptin for 6 hours enhances insulin's ability to inhibit endogenous glucose production, whereas it does not affect peripheral insulin action in lean Sprague-Dawley rats $^{20}$. They went on to show that i.c.v infusion of leptin redirects intrahepatic glucose flux, whereas it does not significantly impact on insulin's capacity to suppress HGP in these animals $^{21}$. In apparent contrast, Cusin et al. showed that subchronic (4 days) i.c.v. leptin treatment reinforces insulin action on glucose disposal, but not production, in lean heterozygous Zucker FA/FA rats $^{22}$. However, leptin treated animals had lost weight at the time hyperinsulinemic clamps were performed in their experiment, which probably explains the metabolic impact of the intervention as pair-fed animals, which were not treated with leptin, exhibited similar
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metabolic features. We now demonstrate that i.c.v. leptin infusion acutely reinforces insulin action on glucose production and disposal in leptin deficient ob/ob mice. We believe that this model is ideally suitable for the study of acute metabolic effects of leptin, as exogenous leptin inevitably competes with endogenous peptide in other models. The mechanism that is responsible for the acute metabolic impact of i.c.v. leptin remains to be established. However, a single i.c.v. injection of leptin acutely activates AMPK in skeletal muscle of FVB mice \textsuperscript{23} and downstream biochemical corollaries can stimulate glucose uptake \textsuperscript{24}.

As far as we are aware, the impact of curtailing body weight by calorie restriction on insulin sensitivity has never been studied before in ob/ob mice. However, a wealth of data supports the position that calorie restriction prevents the onset of insulin resistance in various animal species \textsuperscript{25}, and weight loss promotes insulin action in humans (\textsuperscript{26} and references herein). The size of adipose depots, in particular the visceral fat mass, has an inverse correlation with insulin sensitivity, and the magnitude of beneficial change of insulin action in response to weight loss correlates with the percent decrease of visceral fat in humans.\textsuperscript{26} Moreover, (visceral) obesity is often accompanied by accumulation of lipids in tissues that are not designed to store triglycerides (i.e. muscle and liver) and this may impair insulin action in these tissues \textsuperscript{27}. Calorie restriction precluded lipid storage in liver and muscle to a large extent in our study. Thus, calorie restriction facilitates insulin action on glucose production and disposal in ob/ob mice, probably because it constrains the growth of adipose depots and largely prevents lipid storage in muscle and liver. Notably, all mice were pair-fed for 3 days prior to the experiments. Therefore, we believe that the metabolic benefits we report here are not due to calorie restriction \textit{per se}.

Our data showing that prolonged calorie restriction does not alter body composition (despite a reduction of body weight) corroborate previous reports \textsuperscript{28-30}. They strongly support the notion that leptin not just controls energy balance, but also directs fuel flux away from adipose tissue. In keeping with this, subchronic leptin administration specifically reduces (visceral) fat content in rats \textsuperscript{4}. Insulin clearance from the circulation may be controlled by leptin through activation of cerebral receptors. Ob/ob animals had consistently higher circulating insulin concentrations than wild-type controls during insulin infusion at an equal rate. This observation corroborates earlier papers, reporting that insulin clearance is reduced in
obese individuals. Insulin, when released by beta cells into the portal vein, is primarily cleared by the liver. The kidney is largely responsible for clearance of (exogenous) insulin from the systemic circulation. If insulin is not cleared by liver or kidney, basically all insulin sensitive tissues can internalize and degrade insulin, where muscle has a primary role. Elevated lipids and free fatty acids hamper insulin processing at the cellular and whole body level, perhaps through inhibition of insulin degrading enzyme. Thus, storage of excess TG in insulin degrading tissues (e.g. liver and kidney) may diminish insulin clearance in obesity.

We were surprised to find that i.c.v. administration of leptin significantly reduced circulating insulin concentrations during continuous insulin infusion. This finding strongly suggests that leptin controls insulin clearance through activation of its receptors in the brain.

What are the potential (patho)physiological implications of our findings? Growing evidence implies that obesity is a state of cerebral leptin resistance in many individuals. Disrupted leptin signal transduction in the brain may 1) produce obesity and favor accumulation of adipose tissue rather than lean body mass; 2) hamper insulin action to inhibit glucose production and stimulate glucose disposal; and 3) restrain insulin clearance. If so, drugs designed to impact signals downstream the leptin receptor conveying its metabolic messages (i.e. neuropeptide Y, pro-opiomelanocortin) may be useful tools in the battle against obesity and type 2 diabetes.

In summary, we here show that leptin deficiency per se is deeply involved in the genesis of the metabolic phenotype of ob/ob mice. It is not just responsible for hyperphagia and obesity, but critically determines body composition, insulin action and insulin clearance from the circulation in these animals. The data suggest that putative cerebral leptin resistance in obese humans may underlie various features of the metabolic syndrome and therefore molecular messengers downstream the leptin receptor may be useful targets for the treatment of obesity and type 2 diabetes.
Acknowledgements

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Reference list

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