Insulin Resistance and Metabolic Adaptability in Obese Cats: Two Unlikely Partners

Margarethe Hoenig, Drmedvet, PhD
University of Illinois
College of Veterinary Medicine
Department of Veterinary Clinical Medicine
Urbana, IL
mhoenig@illinois.edu

Abstract
Obese cats show marked insulin resistance of peripheral tissues, but they remain euglycemic despite long-term marked obesity. They are able to maintain hepatic insulin sensitivity and lower endogenous glucose output to sustain euglycemia. Progression to diabetes involves the loss of hepatic insulin sensitivity, beta-cell mass and function.

Glossary of Abbreviations
EGP: Endogenous Glucose Production
EHC: Euglycemic-Hyperinsulinemic Clamp
HOMA: Homeostasis Model Assessment
IVGTT: Intravenous Glucose-Tolerance Test
PEPCK: Phosphoenolpyruvate Carboxykinase

Insulin resistance is a condition associated with obesity in which the beta cells of the endocrine pancreas still produce insulin, but the cellular action of insulin is impaired. The term is often focused on the removal of glucose from the circulation, i.e., glucose clearance, and is frequently applied to the body as a whole, though it has been known for several years that insulin signaling and metabolic pathways can be controlled in a tissue-specific manner.[1] Because both impaired insulin secretion and tissue resistance to the effect of insulin are prerequisites for the development of Type 2 diabetes, differences in tissue insulin sensitivity and insulin secretion may explain why a smaller percent of obese cats progress to diabetes compared to obese humans. People and cats have approximately the same incidence of obesity, yet about 0.5 to 1% of cats and over 8% of obese people develop diabetes. We will examine how we can evaluate the differential tissue regulation by insulin and discuss what is known about the role of different tissues in the ability of obese cats to maintain euglycemia and adapt their metabolic needs. A comparison among different species of several aspects of this review has been published.[2,3]

Several tests have been applied to examine the presence of insulin resistance on a whole body level. They have various degrees of accuracy. In dogs and cats, dynamic tests such as the intravenous glucose-tolerance test (IVGTT) are often used in the research setting. In lean, obese and diabetic cats, the so-called "high-dose" IVGTT is frequently used to test the response to >0.8 g glucose/kg body weight,[4,5] with insulin and glucose concentrations measured at various time points. The frequently sampled IVGTT has also been applied in cats,[6,7] though only in one study has it been compared to the gold standard method, the euglycemic-hyperinsulinemic clamp (EHC), and discrepant results were found between the two tests.[7]

In dogs, the intravenous glucose dose, which is maximally stimulating insulin secretion, has been reported to be 0.6 g/kg body weight.[8] The IVGTT measures insulin sensitivity only in part because the clearance of glucose from blood is not only a measure of tissue responsiveness to insulin but largely depends on the secretory ability of the beta cells. Because it is a dynamic test and secretion and action of insulin are difficult to separate, the IVGTT does not allow precise calculations of tissue responsiveness to insulin as optimal tests for insulin sensitivity are conducted under steady state or near steady state conditions.

Intravenous glucose tolerance has been rarely examined in people because it is too work intensive to apply to a large cohort. If an IVGTT is performed, a much lower dose of glucose is used (0.3 mg/kg), which leads to much lower glucose concentrations and indicates abnormalities at a more advanced stage than would be seen with the high-dose test.[9]

Insulin sensitivity in cats also has been examined by a few investigators using the insulin-tolerance test with a dose of 0.1 unit/kg body weight.[10,11] This dosage of insulin, however, leads to hypoglycemia, and the counter-regulatory response in a healthy animal makes interpretation of the insulin tolerance test difficult.

The most frequently used test in people as a surrogate measure of insulin resistance is the oral glucose-tolerance test. Insulin secretion is not usually measured during this test, thereby it is not a direct index of beta-cell function and one cannot readily distinguish the cause for changes in glucose clearance. A dose of 75 g glucose is administered orally to adult humans, and well-defined parameters have been set for a normal or abnormal response.[12] The 75 g glucose dose leads to blood-glucose concentrations similar to those observed after normal food intake. This may explain why results from

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tests in humans are more predictive of progression to diabetes as the abnormality is already present at lower levels of glucose, i.e., levels that are actually present in a person under normal physiological conditions. In general, this test relies on the principle that with increasing insulin resistance, insulin concentrations will either increase to compensate for the lower insulin sensitivity normalizing blood glucose concentrations or the secreted concentrations fail to do so. The latter case results in glucose intolerance or overt diabetes.

Oral glucose-tolerance testing, unfortunately, has not been shown to be a useful test for cats and dogs because a large glucose dosage is needed and results have been quite variable.[13,14] Authors of some publications have proposed applying the same criteria for a "normal" response in postprandial and/or fasted people directly to cats. However, until published data support similar standards, such interspecies comparisons should be used cautiously, if not discouraged.

Other measures of insulin sensitivity such as homeostasis model assessment (HOMA) have been used in cats.[12] However, these tests in cats have not been examined for their validity. In dogs, HOMA is not a good marker to detect changes in insulin resistance, especially in cases when pancreatic function is compromised.[13]

The gold-standard method to evaluate insulin sensitivity is the euglycemic-hyperinsulinemic clamp, which has been performed in humans, dogs and cats. The EHC is used to measure insulin sensitivity because at steady state the amount of glucose infused by tissues equals the amount of glucose disposed. This test provides even more information when a tracer is added to the infusion.[17] Addition of d-[3-3 H]-glucose, which is metabolized to [^{3}H]-H_2O through the glycolytic pathway, allows the calculation of whole-body glycolysis and total-body glycogen synthesis.[18,19] This test is usually performed in unanesthetized humans or pets; however, we also have shown that tranquilization with tiletamine/zolazepam did not alter the results in cats[20] compared to those that were studied in the awake state.[7] We found that in cats the development of obesity leads to a decrease in glucose effectiveness, i.e., a decrease in insulin-independent glucose uptake, and an increase in insulin resistance[21] similar to what has been shown in people and dogs.[22,23] We calculated that each kg of body weight increase was associated with an increase in insulin resistance by approximately 15 to 30%. Despite the insulin resistance, insulin output increased and fasting plasma glucose remained normal during the development of obesity even when the fat mass increased by approximately 100% and more.[21] This observation highlights that peripheral insulin resistance is not a good indicator of glucose tolerance and by itself is not a predictor for the development of diabetes.

The question as to why obese cats are able to maintain euglycemia despite marked peripheral insulin resistance can be answered when metabolic pathways in the liver are examined. The liver plays a crucial role in the disposal of exogenous glucose and the control of endogenous glucose production (EGP). Using a noninvasive triple tracer technique ($^{2}H_2O$; [U-$^{13}C_3$]propionate; and [3,4-$^{13}C_2$]glucose), we were able to follow pathways involved in glycolysis, gluconeogenesis and the Krebs cycle in both the fasted and postprandial state. We found that endogenous glucose production was decreased in obese cats in the fasted and postprandial state, counteracting the effect of insulin resistance on peripheral glucose uptake and resulting in the maintenance of euglycemia in these long-term obese animals.[24,25] The lower EGP in the obese cats was due to a lower glycogenolysis and/or gluconeogenesis. There was a strong negative correlation between plasma insulin concentrations and endogenous glucose production and between endogenous glucose production and girth, as well as body mass index, suggesting that the decreased endogenous glucose production in our long-term obese cats was due to the higher insulin concentrations of obesity.

We found that neutered obese female cats were metabolically different from neutered obese males, suggesting an early programming of sex differences because these cats had been neutered at an early age. Obese neutered female cats had reduced glucose oxidation, glycogenesis and lipogenesis but increased fat oxidation during the EHC compared to lean female and male obese cats.[20] Interestingly, we found using triple tracers that female obese cats had approximately 1.5 times higher fluxes through phosphoenolpyruvate carboxykinase (PEPCK) and citrate synthase accompanied by a 1.5 times increased rate of pyruvate cycling compared to male obese cats. This suggests that the hepatic Krebs cycle is significantly more active in female obese than male obese cats. Despite that, EGP and gluconeogenesis were not increased because pyruvate cycling was also increased in neutered females compared to neutered male cats.

Pyruvate cycling plays a protective role against overproduction of EGP in the liver by modulating gluconeogenesis from the Krebs cycle. The "futile" pyruvate cycling would decrease hepatic oxidative stress and could be a protective mechanism for female obese cats against the development of diabetes mellitus. We speculated that this mechanism may explain why fewer female cats develop diabetes than male cats. Metabolism was closely correlated with insulin concentrations suggesting that hepatic sensitivity was well maintained in obese cats. It is well known from people that loss of hepatic insulin sensitivity is a prerequisite for the development of diabetes.

The findings that obese cats maintain normal glucose homeostasis despite peripheral insulin resistance were confirmed by results from two other studies. When we examined glucose fluctuations in obese and lean cats during their normal daily routine with a continuous glucose monitoring system, we found that there was no difference in the average glucose

and fructosamine concentrations.[26] Day-to-day glucose variations were more pronounced than intra-day variations, indicating that food intake and other routine behaviors of cats had little influence on glucose concentrations. Only one of the long-term obese cats in that study had a profile with higher overall glucose concentrations and might be considered prediabetic.

In the second study, we examined a large number of healthy cats with normal, overweight and obese body conditions as well as naïve and treated diabetics. There was a clear separation between nondiabetic (all body conditions) and diabetic cats regarding glucose and fructosamine concentrations.[27] The fact that fructosamine concentrations were not higher in any obese cats, which are at greater risk to develop diabetes, compared to lean cats suggests that cats switch rather quickly from the euglycemic to the hyperglycemic state. We would have expected a sliding scale of glucose concentrations if obese cats slowly develop hepatic insulin resistance and subsequent diabetes.

To our knowledge, glucose turnover and metabolic fluxes have not been evaluated in diabetic cats.

There are other differences in obese cats compared to obese humans, which may contribute to the fact that fewer obese cats than people progress to diabetes. In contrast to observations in obese humans, inflammatory cytokines were not found to be increased in plasma or urine in obese cats, and adipocytes lacked infiltration of macrophages.[21]

Conclusion

Obese cats show marked peripheral insulin resistance but are able to maintain euglycemia because of their ability to adapt their hepatic glucose metabolism and to regulate endogenous glucose output. Progression to diabetes involves the loss of hepatic insulin sensitivity coupled with a change in beta-cell mass and function.

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