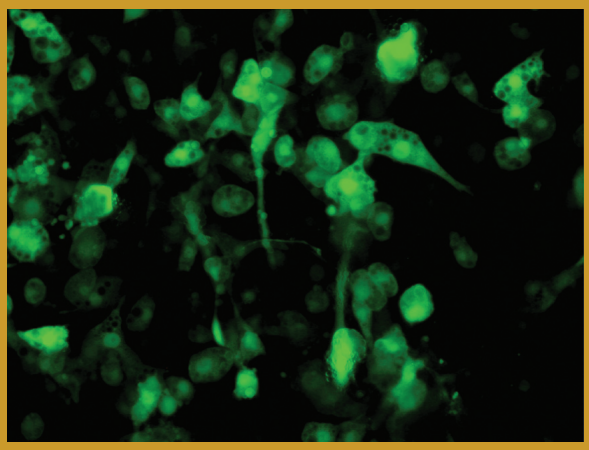

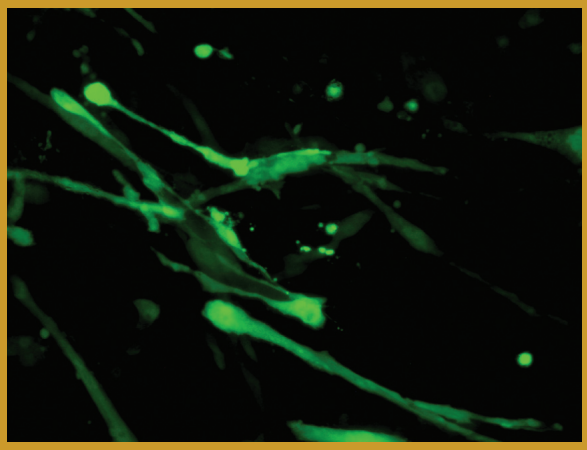

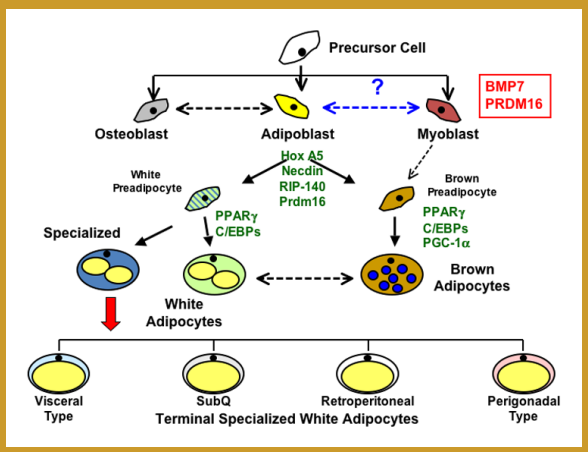

NIDDK-supported research highlighted in this chapter sheds new light on the origin of different types of fat in the body. For example, researchers have discovered a new role for a protein, called PRDM16, as a master switch between the development of brown fat cells and muscle cells. Brown fat cells burn fat molecules to generate heat. The other type of fat, white fat, tends to store excess calories, and is associated with obesity-related diseases. To gain insight into how brown fat cells are formed, scientists recently assessed the effects of depleting the protein PRDM16 from mouse cells that ordinarily develop into brown fat cells. With normal levels of PRDM16, these cells developed into brown fat cells (top panel). Unexpectedly, when depleted of PRDM16, these cells did not develop into fat cells, as predicted, but rather became muscle cells (middle panel). The bottom panel is a diagram summarizing the complexity of steps and factors involved in fat cell development. In the diagram, PRDM16 and another factor called BMP-7, which is also discussed in this chapter, regulate this process at an early step in development, when stem cells have the potential to become precursor cells for bone (osteoblast), fat (adipoblast), or muscle (myoblast). The diagram also shows the steps and factors that determine whether fat precursor cells develop into white or brown fat. This chapter contains additional new information regarding the formation of fat cells, and the potential implications of this research for treating obesity.

Images of cells are courtesy of Dr. Bruce M. Spiegelman and reprinted by permission from Macmillan Publishers Ltd: Nature, 454: 961-967, copyright 2008. Diagram of fat cell development courtesy of Dr. C. Ronald Kahn, Joslin Diabetes Center and Harvard Medical School.

Obesity

Obesity has risen to epidemic levels in the U.S. Individuals who are obese may suffer devastating health problems, face reduced life expectancy, and experience stigma and discrimination. Obesity is a strong risk factor for type 2 diabetes, fatty liver disease, and many other diseases and disorders within the NIDDK's mission.

Approximately one-third of U.S. adults are considered obese based on body mass index (BMI), a measure of weight relative to height.[1,2] Furthermore, while obesity and overweight have risen in the population in general, the greatest increases observed over approximately the past two decades have been in the prevalence of extreme obesity; those who are severely obese are most at risk for serious health problems.[3] Levels of childhood overweight and obesity have also escalated in the past several decades. Obesity affects approximately 16 percent of children and teens ages 2 through 19.[1,4,5] These children are at risk for developing serious diseases both during their youth and later in adulthood. Overweight and obesity also disproportionately affect racial and ethnic minority populations, and those who are socio-economically disadvantaged.

The increased prevalence of obesity in the U.S. is thought to result from the interaction of genetic susceptibility with behavior and factors in the environment that promote increased caloric intake and sedentary lifestyles. Thus, the NIDDK supports a multidimensional research portfolio on obesity, ranging from basic studies to large clinical trials. For example, researchers are elucidating the hormones and other signaling molecules that influence appetite, satiety, and energy expenditure, and that link obesity to type 2 diabetes and other adverse health conditions. With imaging technology, scientists have explored changes in brain activity elicited by the sight of food, and how this brain activity is affected by weight loss. Research on body fat has led to surprising new findings about the origins and formation of different types of fat tissue—not only "white fat," which stores fat molecules and is associated with obesity, but also "brown fat" tissue, which burns fat molecules to generate heat. Research is also revealing molecular links between metabolism, appetite, and the circadian rhythm. Investigators are continuing to develop and test behavioral and environmental interventions to prevent or treat obesity in children. Other research addresses potential medical intervention strategies, including observational studies to evaluate the risks and benefits of bariatric surgery as a treatment for severe obesity. The NIDDK additionally supports studies of eating disorders that are associated with obesity in some people.

Highlights of recent advances from NIDDK-supported research on obesity are provided in this chapter. To help bring the results of research to the public and health care providers, the NIDDK also sponsors education and information programs. Given the importance of the obesity epidemic as a public health problem, and its relevance to the mission of the NIDDK, the Institute continues to play a leading role in the NIH Obesity Research Task Force. Co-chaired by the Directors of the NIDDK and the National Heart, Lung, and Blood Institute, the Task Force includes representatives from these and numerous other NIH Institutes, Centers, and Offices. With extensive input from external scientists and the public, the Task Force developed the *Strategic*

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[1] *Statistics Related to Overweight and Obesity. http://win.niddk.nih.gov/statistics/index.htm*

[2] *Ogden CL, et al: JAMA 295: 1549-1555, 2006; National Center for Health Statistics. Obesity Among Adults in the United States—No Significant Change Since 2003-2004. Data Brief Number 1. Hyattsville, MD: Public Health Service. 2007.*

[3] *Flegal KM, et al: JAMA 288: 1723-1727, 2002; Flegal KM and Troiano RP: Int J Obes Relat Metab Disord 24: 807-818, 2000; Freedman DS, et al: JAMA 288: 1758-1761, 2002.*

[4] *Ogden CL, et al: JAMA 299: 2401-2405, 2008.*

[5] *For children and adolescents, this document uses the terms overweight and obesity interchangeably to refer to a BMI at or greater than the 95th percentile on growth charts (which are based on previous national surveys).*

Plan for NIH Obesity Research, published in August 2004 (http://obesityresearch.nih.gov/About/strategic-plan.htm). The NIH is currently supporting a spectrum of research studies consistent with the recommendations of the *Strategic Plan*.

MOLECULAR CONTRIBUTORS TO OBESITY

In the Eye of the Beholder—The Sight of Food Elicits Different Brain Responses after Weight Loss, Due to Changes in the Hormone Leptin: Scientists have gained new insight into why food can appear particularly irresistible to someone struggling to maintain a reduced body weight, and their research has implications for obesity treatment. Building on the knowledge from previous studies that weight and fat loss reduces leptin levels, the scientists used brain imaging to assess whether weight reduction also modulates brain responses to the sight of food, and whether any such changes can be reversed by the administration of replacement doses of leptin. In the first phase of the study, using a special liquid formula diet, the scientists helped six volunteers who were obese lose 10 percent of their body weight. They then had the volunteers maintain their reduced weight on a carefully-monitored diet. During the weight maintenance phase of the study, the volunteers received leptin injections for several weeks and placebo injections for several weeks so that the effects of the hormone as compared to placebo could be assessed for each person. The scientists used functional magnetic resonance imaging, or fMRI, which provides an image of the brain that identifies regions of high activity. The researchers looked at the activity in different parts of the volunteers' brains as they viewed foods, including fruits, vegetables, grains, and sweets; and, as a control, non-food objects like a cell phone. The sight of the food affected activity in several parts of the volunteers' brains. Comparing images taken before and after weight loss, the scientists found that weight loss altered the mental response to food, resulting in an increase in brain activity in some areas and a decrease in others. Prior research had shown that the affected areas of the brain are associated with a variety of emotional and sensory responses to food, decision making, and other behaviors related to food. The scientists also discovered that leptin administration reversed many of the weight loss-induced changes in brain activity. Thus, food is perceived differently depending on the eye—or in this case, the leptin levels—of the beholder. This research advances knowledge of the many biologic processes that conspire to make the body regain lost fat: earlier studies had shown that weight loss leads to increased hunger and also causes the body to expend less energy and muscles to work more efficiently, so that fewer calories are burned. The identification of leptin as having a key role in all of these processes augments the evidence that leptin replacement therapy may help people maintain weight loss.

Rosenbaum M, Sy M, Pavlovich K, Leibel RL, and Hirsch J: Leptin reverses weight loss-induced changes in regional neural activity responses to visual food stimuli. J Clin Invest 118: 2583-2591, 2008.

Rev-ing Up Metabolism and Fat Cell Development: Scientists have recently uncovered intriguing new roles in metabolism for a key protein controlling the circadian clock. In animals and humans, the internally driven circadian clock regulates many behaviors and bodily processes—including sleep/wake cycles, changes in blood pressure, and body temperature fluctuations—to harmonize these activities with daily, rhythmic changes in the environment, most notably day/night cycles. Not surprisingly, metabolic functions, such as the synthesis of glucose (sugar) and fats and the release of glucose into the blood, are also controlled by the circadian clock—but how that is accomplished at the molecular level has been poorly understood. Now, researchers have evidence that a protein called Rev-erb-alpha links the circadian clock and metabolism. Rev-erb-alpha is a key regulator of the circadian clock, controlling in a rhythmic fashion how much protein is made by a "master" clock gene. In their experiments, the researchers found that Rev-erb-alpha is highly dependent for its regulatory activity on another molecule called heme. The heme molecule is integral to many metabolic pathways, and its concentrations in the cell oscillate in a circadian rhythm. Through a series of biochemical tests and experiments in cells, they observed that, if heme was present, Rev-erb-alpha functioned normally. However, if they removed heme or used a mutant Rev-erb-alpha protein that could not bind heme, the protein's activity was diminished. At the same time, the researchers discovered that Rev-erb-alpha regulates not only a

master circadian clock gene, but also two key genes involved in glucose and fat metabolism—and that this activity also depends upon Rev-erb-alpha's ability to bind to heme. Thus, the interaction of heme with Rev-erb-alpha is a strong candidate for a coordinator of the circadian clock and metabolism. In a related study, researchers from the same laboratory demonstrated that Rev-erb-alpha is also important to the maturation of fat cells from their precursor cells. Through studies using fat cell precursors grown in the laboratory, the researchers found that the Rev-erb-alpha protein is present in high amounts at the onset of maturation, but its levels quickly drop as maturation proceeds. The initial increase and sudden decrease in levels of Rev-erb-alpha appears to be necessary to kick-start fat cell maturation and then allow it to move forward. From these studies, Rev-erb-alpha thus appears to play a key role not only in the circadian clock, but also in metabolism and in cellular development. Further study of these newly-discovered roles for Rev-erb-alpha will help investigators better understand the interrelationships between the circadian clock, metabolism, and cellular differentiation and how they might influence each other in both health and disease.

Yin L, Wu N, Curtin JC, Qatanani M, Szwergold NR, Reid RA, Waitt GM, Parks DJ, Pearce KH, Wisely GB, and Lazar MA: Rev-erb-alpha, a heme sensor that coordinates metabolic and circadian pathways. Science 318: 1786-1789, 2007.

Wang J and Lazar MA: Bifunctional role of Rev-erb-alpha in adipocyte differentiation. Mol and Cell Biol 28: 2213-2220, 2008.

Rats Susceptible To Developing Obesity from a High-Calorie Diet Exhibit Reduced Density of Brain Cell Connections: Scientists have shown that rats susceptible to obesity from a high-calorie diet have fewer brain cell connections than control rats in a key part of the brain that regulates body weight, known as the hypothalamus. This reduced density of brain cell connections was seen in both young and adult animals, indicating that the defect occurs early in life and persists into adulthood. Specifically, the scientists were interested in the brain cell connections that form in response to the hormone leptin. Leptin plays a key role in the development of brain cell connections in the hypothalamus and, in addition, serves as a sensor to the brain regarding appetite regulation whereby increased leptin levels depress appetite and promote satiety. Interestingly, most people who are obese have high leptin levels but are resistant to leptin signaling, including its influence on appetite. Similarly, in this rat model of diet-induced obesity, the obese rats also are resistant to leptin signaling. The scientists hypothesized that the leptin-resistant state of the rats may be due to abnormal development of the hypothalamic brain cell connections, specifically from the arcuate nucleus of the hypothalamus (ARH). Using a fluorescent tracer, the scientists were able to visualize the connections from the ARH to another part of the hypothalamus also known to be important in body weight regulation. The scientists compared these connections in brains of diet-induced obese rats and in other rats termed "diet-resistant" because they do not gain excess weight when fed the same high-calorie diet. The brain cell connections between these two parts of the hypothalamus were two- to four-fold less dense in diet-induced obese rats as compared to the diet-resistant animals. The initial experiment was conducted in rats 12-16 days old, but the same pattern was observed in adult rats. Furthermore, maternal diet did not influence the density of the connections. This result indicated that the genetic predisposition of the offspring held greater influence over the density of the connections than the environmental effects of the mother's diet. To investigate the role of leptin in the formation of these connections, the scientists added leptin to samples of brain tissue from the rats. They found that leptin caused the brain cells to form fewer connections in the brain tissue from rats prone to diet-induced obesity, in comparison to diet-resistant rats. The combined results of these and other experiments suggest that the leptin-resistant state of diet-induced obese rats may begin with decreased brain cell connections formed during neonatal development of the ARH, a key area that relays leptin signaling to other parts of the hypothalamus. Therefore, with fewer connections to transmit the leptin signal, the brain is less able to respond to leptin levels, even in very high amounts. The research presented here provides new avenues to investigate the complex brain pathways involved in obesity, eating behavior, and the regulation of body weight using emerging technologies in brain imaging and related fields.

Bouret SG, Gorski JN, Patterson CM, Chen S, Levin BE, and Simerly RB: Hypothalamic neural projections are permanently disrupted in diet-induced obese rats. Cell Metab 7: 179-185, 2008.

The Power of Hunger—the Hormone Ghrelin Harnesses Brain Cells' Energy Machines To Increase Appetite: Scientists have discovered that the hormone ghrelin induces hunger through a process involving mitochondria, components of cells that generate energy for biologic processes. In particular, the researchers found that ghrelin acts via a mitochondrial protein called UCP2. Ghrelin is produced in the gut; its levels rise just before meals to induce food intake, and then fall just after a meal. When ghrelin travels from the gut to the brain, it latches onto specific brain cells and sets in motion an elaborate set of molecular signaling pathways that not only increase hunger, but, as shown in this new study, also ensure that the target brain cells will have ample energy to keep sending their message to "start eating," even though the message needs to be sent at times when the body may be comparatively low on energy. Prior research indicated that UCP2 is present in the same brain cells that serve as docking sites for ghrelin, and both UCP2 and ghrelin levels rise during fasting. Based on this guilt-by-association, the researchers explored whether UCP2 may somehow assist ghrelin. To do this, they assessed ghrelin's actions in normal mice and in mice that lacked UCP2 due to a genetic mutation. They found that ghrelin increases the number of mitochondria in brain cells, which in turn contributes to an increase in overall mitochondrial activity. Focusing specifically on a group of brain cells known to promote increased food intake, the scientists next observed that ghrelin causes these cells to fire off more signals, increase production of molecules associated with hunger, and inhibit another type of brain cell known to dampen appetite. All of these effects of ghrelin were dependent upon UCP2, as they were greatly attenuated in the mutant, UCP2-deficient mice. Other experiments showed that UCP2 affects how much food the mice consume.

Having elucidated the importance of mitochondria and UCP2 to ghrelin's effects on appetite, the scientists next sought to determine what UCP2 may be doing to assist ghrelin. Increased mitochondrial activity can lead to increased production of a destructive metabolic byproduct called reactive oxygen species (ROS, often referred to as "free radicals"), which UCP2 has previously been found to help eliminate. The scientists compared the levels of ROS in the brains of normal and UCP2-deficient mice. In normal mice, although ghrelin increased mitochondrial use of fatty acids as fuel, ROS levels did not increase. In mice lacking UCP2, however, ghrelin administration led to higher levels of these toxic byproducts. The scientists reasoned that UCP2 may enable appetite-inducing brain cells to keep working by eliminating toxic byproducts which would otherwise inhibit them. They also found that ghrelin boosts UCP2 production in normal mice, enabling further scavenging of ROS. Through these and other experiments, the scientists have shed new light on how ghrelin induces food intake. Ghrelin causes certain brain cells to increase their appetite-promoting activities, and increases mitochondrial burning of fatty acids to provide the cells with energy to drive these activities. UCP2 helps rid the cell of the resulting toxic byproducts, to ensure that their adverse effects do not interfere with the cell signaling that increases appetite and, ultimately, food consumption. These novel insights may lead to new approaches to help reduce overweight and obesity.

Andrews ZB, Liu Z-W, Walllingford N, Erion DM, Borok E, Friedman JM, Tschöp MH, Shanabrough M, Cline G, Shulman GI, Coppola A, Gao X-B, Horvath TL, and Diano S: UCP2 mediates ghrelin's action on NPY/AgRP neurons by lowering free radicals. Nature 454: 846-851, 2008.

A Role for the Immune System in Protecting Against Obesity and Insulin Resistance: One of the processes associated with obesity and the insulin resistance that leads to type 2 diabetes is inflammation of highly metabolic tissues such as liver and muscle, and other metabolic tissues such as fat. In this process, immune system cells called macrophages are activated to migrate to these tissues and release signals that induce inflammation and promote insulin resistance. However, not all macrophages have this effect. Those that do not are said to be "alternatively activated," and are more common in lean animals than in those that are obese. Now, researchers have identified key molecular triggers of the alternative activation pathway, and identified a subset of liver macrophages called Kupffer cells which, when alternatively activated, have a key role in preventing insulin resistance in over-fed mice. Further investigation will be required to understand the genetic and environmental signals that trigger the pathway to alternatively activate macrophages and the precise mechanism by which these cells modulate insulin sensitivity. If these results prove to be relevant to human tissues as they are in mice, therapeutic

stimulation of the alternative activation program might be an effective means to treat obesity and prevent insulin resistance.

Odegaard JI, Ricardo-Gonzalez RR, Red Eagle A, Vats D, Morel CR, Goforth MH, Subramanian V, Mukundan L, Ferrante AW, and Chawla A: Alternative M2 activation of Kupffer cells by PPARdelta ameliorates obesity-induced insulin resistance. Cell Metab 7: 496-507, 2008.

DETERMINANTS OF "BROWN" FAT FORMATION

Brown or Brawn—Brown Fat and Muscle Cells Share Common Origins, with Fate Determined by PRDM16: In a surprising finding, scientists have discovered that skeletal muscle cells and brown fat cells can arise from the same precursors. They also identified the biologic switch, a protein called PRDM16, that directs the precursors to develop into brown fat. Previously, it was thought that the two major types of fat tissue, brown and white, shared the same developmental origins. However, in many ways they are quite different. White fat cells serve as the body's energy reserves by storing fat, and they additionally secrete a variety of signaling molecules. Excess white fat tissue is associated with obesity and related diseases. By contrast, brown fat cells burn fat molecules to dissipate heat, and might thus protect against obesity. Brown fat cells also share similarities with muscle cells; for example, both burn fat in mitochondria, the components of the cell that generate energy to power biologic processes or to provide body heat.

To understand how brown fat cells are formed, scientists recently assessed the effects of depleting PRDM16 from mouse cells that were thought to be precursors of brown fat. Unexpectedly, brown fat precursors depleted of PRDM16 did not morph into white fat cells, as predicted, but rather became muscle cells. The scientists then hypothesized that brown fat and muscle cells might have the same precursors. To determine whether this was the case, they designed a way to visualize cells that arose from these precursors. Their approach was to genetically engineer mice so that their muscle cell precursors contained a readily visible marking that would be inherited during development by cells that derived from these precursors. Once the mice had grown, the scientists found that the brown fat and skeletal muscle cells—but not the white fat cells—contained this marking. Finally, the scientists found that excess production of PRDM16 in muscle cell precursors could divert their development from muscle cells into brown fat. Thus, the researchers concluded that both brown fat and muscle cells share a precursor and that PRDM16 plays an important role in determining whether the precursor gives rise to brown fat or muscle.

Scientists also recently gained new understanding of how PRDM16 can both turn on brown fat-specific genes and turn off genes important for white fat cells. It does so by associating with different sets of additional regulatory proteins. Interestingly, brown fat cells can arise within white fat tissue under certain conditions, such as extended exposure to cold or stimulation of some nervous system responses, and these types of brown fat cells appear to have a different origin, unrelated to muscle. These studies provide novel insights into the development of brown fat, and may inform the development of a new intervention strategy for obesity: generating more brown fat cells to burn excess calories.

Kajimura S, Seale P, Tomaru T, Erdjument-Bromage H, Cooper MP, Ruas JL, Chin S, Tempst P, Lazar MA, and Spiegelman BM: Regulation of the brown and white fat gene programs through a PRDM16/CtBP transcriptional complex. Genes Dev 22: 1397-1409, 2008.

Seale P, Bjork B, Yang W, Kajimura S, Chin S, Kuang S, Scimè A, Devarakonda S, Conroe HM, Erdjument-Bromage H, Tempst P, Rudnicki MA, Beier DR, and Spiegelman BM: PRDM16 controls a brown fat/skeletal muscle switch. Nature 454: 961-967, 2008.

Building Brown Fat with BMP-7: A new discovery in fat cell research may point the way to new therapeutic options for obesity. In mammals, not all fat, or adipose tissue, is the same. White adipose tissue stores extra calories as fat for later use and is the tissue associated with obesity, while brown adipose tissue, or brown fat, actually burns fat to generate heat, keeping an animal warm and slim. Until recently, it was thought that in humans, only newborns had brown fat, but new findings suggest that adults actually retain some as well. In a recent study, researchers sought out factors that determine the generation of brown fat from

precursor cells. Working with laboratory-grown fat precursor cells from mice, they found that treatment with a molecule called bone morphogenetic protein 7, or BMP-7, is sufficient to drive precursor brown fat cells to develop into active brown fat cells. BMP-7 treatment suppressed cellular factors that normally inhibit brown fat cell development and induced production of key molecules that drive brown fat cell maturation—including UCP1, a signature protein found in brown fat cells that is essential for generating heat. Precursor brown fat cells treated with BMP-7 also increased by five-fold the number of their mitochondria, the cellular powerhouses that enable them to burn fat. In contrast, precursor white fat cells from mice were not affected by BMP-7 treatment. Demonstrating that BMP-7 is important to brown fat tissue development in animals, the researchers found that mice genetically engineered to have no BMP-7 had very little brown fat and produced little or no UCP1 protein as compared with normal, BMP-7-producing siblings. Increasing the amount of BMP-7 in mice had the opposite effect. The researchers found that administering extra BMP-7 to mice via a genetically engineered virus not only increased their brown fat mass, but also significantly increased whole body energy expenditure and basal body temperature—leading to a significant reduction in weight gain when compared with control mice. Although these experiments were all performed with mouse cells and in mice, now that brown fat has been found in humans, the findings suggest that BMP-7 may prove to be a therapeutic target to help counteract obesity in humans in the future.

Tseng Y-H, Kokkotou E, Schulz TJ, Huang TL, Winnay JN, Taniguchi CM, Tran TT, Suzuki R, Espinoza DO, Yamamoto Y, Ahrens MJ, Dudley AT, Norris AW, Kulkarni RN, and Kahn CR: New role of bone morphogenetic protein 7 in brown adipogenesis and energy expenditure. Nature 454: 1000-1004, 2008.

LIFESTYLE INTERVENTION TO REDUCE OBESITY IN CHILDREN

Strategy To Reduce Screen Time for Young Children—Beneficial Effects on Body Weight:

With a combination of advanced technology, stickers, and other incentives, researchers have developed a new strategy to help parents limit the hours their young children spend transfixed in front of a television or computer game—and thus reduce their risk for gaining excess weight. Increasing evidence has linked television viewing with childhood obesity. The researchers focused on reducing screen time in children ages 4 to 7 in part because intervening early may help prevent later health problems. At the start of the study, the children were at or above the 75th percentile for body mass index (BMI, a measure of weight relative to height) for their age and gender, and they spent 14 hours or more each week watching television or playing computer games. Families were randomly assigned to either the intervention or control group. For the intervention group, the researchers attached a "TV Allowance" device to all televisions and computers in the home, and provided each family member with an individual code to permit screen time. The devices were programmed to allow only a certain amount of screen time for the participating child, and to reduce the child's weekly screen time by half over the first several months of the study. Parents also gave the children stickers and praise for their progress, as well as other incentives. In the control group, children did not have these types of limits on screen time; instead, their families received newsletters with tips on parenting, activities, and recipes.

The study showed that television and computer use can be reduced substantially in young children, and that these reductions can beneficially affect BMI. After 6 months and 12 months of the study, children in the intervention group had lower BMIs than those in the control group. Among the children from families of lower socioeconomic status, the intervention was particularly beneficial: the researchers observed a substantial difference in BMI between the intervention and control groups, and the improvement in BMI lasted for the full 2 years of the study. This result is encouraging given the increased risk for obesity in socioeconomically disadvantaged children. The researchers also found that reduced screen time correlated with the children's eating fewer calories, although not with changes in physical activity. The intervention's overall effect on BMI was not large. However, by inducing even modest reductions in BMI and television watching behavior at a young age, this type of relatively inexpensive intervention, in combination with other efforts, may ultimately help reduce risk for health problems associated with childhood obesity.

Epstein LH, Roemmich JN, Robinson JL, Paluch RA, Winiewicz DD, Fuerch JH, and Robinson TN: A randomized trial of the effects of reducing television viewing and computer use on body mass index in young children. Arch Pediatr Adolesc Med 162: 239-245, 2008.

POTENTIAL MEDICAL INTERVENTION STRATEGIES TO COMBAT OBESITY AND ITS ASSOCIATED CONDITIONS

Drugs Boost Exercise Endurance in Mice: Researchers have identified two drugs that, in mice, seem to confer many of the healthful benefits of long-term exercise, giving them more fat-burning muscle and better endurance. These findings might eventually lead to better treatments for certain muscle disorders, frailty, obesity, and other conditions in which exercise is known to be helpful but not always practical.

Scientists have long searched for drugs that mimic or enhance the effects of exercise and its known therapeutic benefits. Several years ago, scientists identified a potential drug target—a signaling protein known as PPARdelta—which regulates several fat-burning genes in muscle cells. The researchers created genetically-engineered mice that produced high levels of PPARdelta in their muscles. The animals' running endurance nearly doubled, with their muscles developing more fat-burning, fatigue-resistant muscle fibers than normal mice. In a new study, the researchers used drugs, rather than genetic engineering, to enhance the effects of PPARdelta and another exercise-related molecule, AMPK. The scientists first gave oral doses of a PPARdelta-activating drug to mice for several weeks. Initial results were disappointing. The drug by itself had no impact on the animals' running endurance in a treadmill test. However, when the scientists added exercise training to the mix—having the mice run on a treadmill for nearly an hour daily for a month—the treated mice were able to run up to 75 percent farther than mice that received exercise training alone. The treated mice developed nearly 40 percent more fatigue-resistant muscle fibers than untreated animals. In addition, gene activity in their muscle cells was strikingly similar to the genetically-engineered mice in previous studies.

The researchers also tested the effects of the protein AMPK, which is known to be activated by exercise and is involved in regulating many other genes. The mice received daily doses of the drug AICAR, which activates AMPK. Surprisingly, 4 weeks of AICAR treatment alone activated exercise-related genes and enhanced running endurance by 44 percent in sedentary mice. In fact, the drug allowed sedentary animals to run longer and farther than animals that had received weeks of exercise training. Either drug alone activated a unique subset of exercise-related genes. However, the greatest rise in gene activity occurred when the PPARdelta-activating drug was combined with exercise training, which activates AMPK. This drug-exercise pattern of gene activity—dubbed an "endurance gene signature"—led to the greatest improvement in endurance. Because the muscles of humans and mice use similar genetic pathways, these findings could eventually lead to improved therapies. However, the drugs' effects on human muscles and endurance must still be tested.

Narkar VA, Downes M, Yu RT, Embler E, Wang Y-X, Banayo E, Mihaylova MM, Nelson MC, Zou Y, Juguilon H, Kang H, Shaw RJ, and Evans RM: AMPK and PPARdelta agonists are exercise mimetics. Cell 134: 405-415, 2008.

Reprinted, in slightly modified form, from NIH Research Matters*; original article by Vicki Contie published August 11, 2008.*

Encouraging Metabolic Results in Test of a Candidate Therapeutic for Cardiovascular Disease Risk: Scientists have recently shown that a compound that mimics some of the effects of a naturally-occurring hormone has metabolic benefits in people with high cholesterol, without causing adverse effects on the heart in a short-term study. The compound is designed to work like thyroid-stimulating hormone (TSH), a key metabolic regulator. People who make too little of their own TSH have numerous health problems, including obesity, high cholesterol, osteoporosis, and impaired kidney function. TSH may thus appear to be an attractive therapy to counteract some of these conditions, even in people who do not have low TSH levels. However, administering too much TSH can cause dangerously elevated heart rates and arrhythmias, as well as other health problems. Therefore, researchers have worked to develop

compounds with some of the salutary effects of TSH, without its potentially serious side-effects. Such TSH-like molecules have been successfully tested in overweight animals, where they proved to induce, without toxicity, weight loss and improvements in blood lipids. In the current study, scientists tested one of the TSH-like compounds in a small number of people with elevated cholesterol. They found that it significantly lowered LDL ("bad")-cholesterol, without causing the obvious heart problems typically observed in people with elevated TSH. The treatment did not result in weight loss or other obvious bodily changes during the 2 week test. Statins are popular and effective medications that also lower LDL-cholesterol, but they are poorly tolerated by some people. The TSH-like compound works by a different mechanism than statins, and therefore may prove to be an option for people who cannot take those medications. Although the results obtained in this study are very preliminary—long-term tests will be required to determine whether this potential therapeutic provides enduring benefits without simultaneously causing other health problems—these findings are exciting because they provide hope for a new therapy to combat heart disease.

Berkenstam A, Kristensen J, Mellström K, Carlsson B, Malm J, Rehnmark S, Garg N, Andersson CM, Rudling M, Sjöberg F, Angelin B, and Baxter JD: The thyroid hormone mimetic compound KB2115 lowers plasma LDL cholesterol and stimulates bile acid synthesis without cardiac effects in humans. Proc Natl Acad Sci USA 105: 663-667, 2008.

Dr. Rudolph Leibel and Dr. Natasha Leibel: Combating Obesity and Diabetes Is a Family Affair

Dr. Rudolph Leibel and Dr. Natasha Leibel (Photo credit: Charles Leduc)

When it comes to furthering the understanding and treatment of diabetes and obesity, it is difficult to imagine a more passionate and devoted team than that of Dr. Rudolph Leibel and his daughter, Dr. Natasha Leibel, at the Naomi Berrie Diabetes Center at Columbia University Medical Center.

Dr. Rudolph Leibel (referred to as Rudy in this profile) is the Co-Director of the Naomi Berrie Diabetes Center and Professor of Pediatrics and Medicine at Columbia University. He has dedicated his research career to studying the regulation of body weight in rodents and humans and the genetic basis of type 2 diabetes and obesity. He has had research support from the NIDDK for over 25 years and has served as a member of the NIDDK Advisory Council and the NIDDK Clinical Obesity Research Panel. Dr. Natasha Leibel (referred to as Natasha in this profile) is a pediatric endocrinologist specializing in the treatment of diabetes in children. Together, father and daughter exemplify the concept of translational research at its best. Rudy's work in the laboratory is leading to a greater understanding of the causes of obesity and type 2 diabetes and is also providing insight into potential treatments. Natasha is at the forefront of putting this new research knowledge into practice as she cares for children with diabetes.

Dr. Rudolph Leibel: At the Forefront of Research on Obesity and Diabetes

While this father and daughter duo work in the same field—and only a few floors apart in the same building—they each arrived at their career through their own distinct path. Rudy, a world-renowned scientist who has made many contributions to the fields of diabetes and obesity research, started out his medical school training thinking that he would have a very different career path. "I wanted to be a psychiatrist," he states.

However, at the end of medical school, he decided to enter pediatrics—a decision that would eventually lead to his interest in laboratory research.

Following his general pediatric residency, Natasha's birth, and 2 years in the U.S. Army, Rudy started an NIH fellowship in endocrinology (the study of diseases related to the glands and hormones of the body) at Massachusetts General Hospital in Boston. Endocrinology was very exciting to him. As he recalls, "There was plenty of opportunity to study the origins of clinical diseases and to make new discoveries—very little was known about the hormones involved in many of the endocrine diseases. Assays to measure these hormones were just becoming available."

Rudy recalls one particularly memorable experience in the clinic over 30 years ago, when he received a visit from an obese child and his mother—a visit that would dramatically change his career path. At that time, obese children were frequently referred to pediatric endocrinologists because most people assumed that childhood obesity was associated with thyroid or adrenal problems, which is rarely the case.

Rudy could not find anything wrong with the child and, therefore, counseled the mother on healthier eating and physical activity. The mother was unimpressed with what

she determined was Rudy's lack of knowledge about her son's condition and she made a point of telling him so with some creative language.

Rudy says, "I was shocked for a number of reasons, as you might imagine, but, as I thought about it, I really had to agree with her. There was indeed a collective lack of understanding among scientists and clinicians about the causes of obesity." Rudy decided, "If I were going to remain in this field and help obese children, I needed to do something to move the field forward and advance knowledge about the regulation of body weight and the underlying causes of obesity."

A short time after this experience in the clinic, Rudy made an impromptu visit to the laboratory of Dr. Jules Hirsch, an expert in obesity and the properties of fat tissue in the body, at Rockefeller University in New York City. This visit further inspired Rudy to pursue the regulation of body weight in a research setting. The entire family supported Rudy's interest in research and moved to New York—even if it meant moving from a big house in Boston to a small apartment in New York.

Rudy states, "I stayed in research much longer than I might have because of a real passion for the work and a desire to follow it to completion…I originally went to Rockefeller on a 2 year leave of absence; however, this leave has now lasted 35 years."

Rudy's dedication and passion have led to the identification of several genes involved in the regulation of body weight and diabetes. For example, his lab collaborated in the discovery of the leptin gene. This discovery is just one of many key contributions that he has made to increasing understanding of the underlying causes of obesity and diabetes.

Dr. Natasha Leibel: At the Forefront of Treating Children with Diabetes

"I made the conscious decision to go to medical school while I was in college," Natasha replies, when asked when she first was interested in medicine.

Her father, however, thinks that her interest in medicine and her passion for helping others came about at an earlier age. "When we lived in Boston, I was practicing in a hospital and seeing a lot of patients. Because I was so busy, patients would often call my home asking for medical advice, and Natasha would listen to me talking to them," Rudy remembers. "As a little girl of 6 or 7 years old, Natasha would answer the phone and provide advice to patients about the treatment of acute conditions such as diarrhea and high blood sugar," he says. Although Rudy felt, "she gave rather solid advice," he stresses that his wife would always intervene and explain, "That was not the Dr. Leibel you were looking for." Rudy states, "I'm not saying that led her to a career in medicine but I don't think she was dissuaded by any of it."

Growing up, Natasha was very much aware of her father's career and the importance he placed on science as an essential part of education. She also observed his compassion and kindness toward others and, from this, knew that she also wanted to be in a profession where she could make a real difference in the lives of other people. "I decided that medicine was a good fit for me because of the many opportunities within the discipline and also because it would provide me with a chance to give back to society," said Natasha.

During medical school, Natasha felt especially drawn to pediatrics because, as she states, "I really enjoyed working with families. Children, in particular, are so vulnerable, especially when they are sick." Like her father, she was fascinated by the field of endocrinology; however, she is facing a new set of challenges. While Rudy is on the forefront of research, Natasha is on the forefront of clinical care where she specializes in caring for children with diabetes, including children with type 2 diabetes. Type 2 diabetes was previously called "adult-onset" diabetes because it primarily affected older adults. However, the disease is increasingly being diagnosed in children—a trend associated with the rising rates of pediatric obesity.

Natasha states, "The development of type 2 diabetes in children is so new that there are many questions regarding the optimal treatment of diabetes in this young population." To answer these questions, Natasha is involved in the NIDDK's Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) study. The TODAY study seeks to identify the best treatment approach for type 2 diabetes in children and teens.

Natasha also cares for patients with type 1 diabetes, an autoimmune disease that most often occurs in infancy, childhood, and adolescence. Therefore, whether treating

a child with type 1 or type 2 diabetes, Natasha has the difficult task of caring for children living with a chronic and burdensome condition. Natasha says, "I get to be a part of these families' lives, which is a wonderful gift as a doctor." She also knows how difficult living with diabetes is for these children. Natasha is passionate about helping her patients and making a difference in their lives.

Bringing the Clinic and Laboratory Together for Patients

Some of Natasha's patients have the opportunity to meet and work with the researchers, including Rudy, who also are working in the laboratory to advance understanding of diabetes causes and treatment. Natasha says, "It is amazing to bring the children who we take care of to the laboratory to talk with the scientists. It is very encouraging for the children to see that there are people working so hard to try to understand their disease."

In addition, the Naomi Berrie Diabetes Center hosts a summer diabetes camp where Natasha notes, "The high point of the week is bringing the children upstairs to the laboratory." While Rudy and Natasha do not advertise their father/daughter relationship during these visits, the children often figure it out. Rudy and Natasha feel that some of the children are especially heartened to learn that a child of someone who has been working in the field of diabetes research for so long has gone on to pursue clinical training in this area. In fact, some of the participants in the diabetes camp are so inspired by the scientists and clinicians that Rudy and Natasha feel that they may be stimulating a future generation of pediatric endocrinologists. The Diabetes Center also sponsors summer students who are in high school, college, or medical school, to work in the research laboratories. Some of these students are also patients in the Diabetes Center. This program gives them an opportunity to contribute to a better scientific understanding of their disease.

Following Your Passion

Since Rudy and Natasha both pursued medical training in the same subspecialty—and Rudy was even one of Natasha's teachers in medical school—it may seem as if Rudy's career path had a major influence on Natasha's choice of profession. On the contrary, she explains, the way her father has lived his life has had a much bigger impact on Natasha than his medical and scientific pursuits.

"Growing up," Natasha explains, "my sister Alexis, who is also a physician, and I were always encouraged to find what we were passionate about and what we enjoy doing. Certainly, that was the example we saw him live by," she says of Rudy, "which was to follow his passion and to work really hard." Natasha continued, "He has been my role model because of the way he has lived his life and the challenges he has taken on. His enthusiasm and dedication are a constant source of inspiration."

Being in the same area of medicine does provide its advantages though, for both Natasha and Rudy. Natasha states, "It is an honor to work with him, and a great bond we have being in the same profession. Although I may have 'helped' him as a young girl with his patients, I certainly go to him all the time with questions about my patients. He is a phenomenal clinician." Likewise, Rudy is extremely proud of Natasha and her accomplishments in the profession they share.

Working only a few floors apart, Natasha says that, "People always ask us if we eat lunch together." She cautions that this is not the case because she and her father have fundamentally different views about lunch. Rudy skips lunch, while Natasha makes lunch a priority. In fact, when Natasha started at the Naomi Berrie Diabetes Center in 2005, Rudy joked that he would take her to lunch in 2007. Natasha laughs and says, "He did, but I ended up paying because he forgot to bring money." It looks like the next lunch date is scheduled for sometime this year.

In the meantime, both father and daughter are working tirelessly in the laboratory and in the clinic to combat obesity and diabetes. Even if they don't meet every day for lunch, Rudy and Natasha do see each other and talk about their work. Natasha confided, "We do talk frequently about science and he is a great source of knowledge. Mostly now he likes to talk about his grandchildren, though." And, she adds about her inspiring physician-scientist father, "He is a great babysitter!"

STORY OF DISCOVERY

Leptin as a Treatment for Lipodystrophy: A Translational Success Story

This story begins with an obese mouse and ends with a medical treatment for people who may lack fat tissue altogether. The common link that ties together these two very different entities is a hormone called leptin. Identifying this link was a result of the collaboration among many investigators over several years, including NIDDK-supported scientists at universities, scientists in the NIDDK Intramural Research Program, industry researchers, and many others. This translational success story is a demonstration of how exciting discoveries in the laboratory are used to improve the health of people.

The Obese Mouse and the Discovery of Leptin

In 1950, scientists identified a new mouse model that was extremely obese. They called the unknown gene causing the obesity "*ob*." By the 1980s, the identity of the *ob* gene was still unknown, but it was becoming more and more apparent that research on genetic contributors to obesity was critically important to pursue. Therefore, the NIDDK sought to support research to identify obesity-related genes in rodents, including the *ob* gene. The Institute sponsored a workshop on this topic and developed an initiative to solicit research applications. In 1989, the NIDDK awarded a grant to Dr. Jeffrey Friedman through this initiative. Dr. Friedman's subsequent pioneering research led to the 1994 discovery of the mouse *ob* gene. The hormone produced by this gene was named "leptin," a term that derives from a Greek word meaning thin. Because the *ob* mutant mouse was obese, the scientists realized that the normal *ob* gene—and the hormone it encodes—must contribute to leanness.

The landmark discovery of leptin unleashed a wave of new research advances in fat biology and metabolism. Researchers found that leptin is secreted by fat cells and released in proportion to the amount of fat. These observations drastically altered the former view of normal fat tissue as simply a passive "fat storehouse." Research fueled by this 1994 discovery also led to the identification of a number of other substances that, like leptin, are secreted by fat cells and influence appetite and metabolism.

Studies demonstrated that obese animals deficient in leptin, including mice carrying the mutant form of the *ob* gene, lost weight when given the hormone. Therefore, researchers postulated that leptin treatment might also be useful for human obesity. There are, in fact, very rare instances of complete deficiency of leptin in humans that result in morbid obesity from infancy. Leptin treatment in these individuals caused substantial weight loss, providing hope for improved quality of life and longevity.

Unfortunately, in clinical studies done at that time, leptin administration was not effective in treating the vast majority of cases of human obesity, which are not due to leptin deficiency. In most cases, obesity results from a complex interaction among genetic variation (potentially involving many genes not yet identified) and the environment. Obese individuals, in fact, usually have very high levels of leptin, probably a consequence of the many fat cells secreting it. The inability of the high levels of leptin to decrease body weight suggests that the more common forms of obesity are associated with a resistance to leptin's actions. Although these results were disappointing, scientists did not give up in their quest to use this new knowledge to benefit people.

Testing Leptin as a Treatment for Lipodystrophy

Scientists in the NIDDK's Intramural Research Program had broad experience with respect to

STORY OF DISCOVERY

studying people with various forms of insulin resistance. Using this experience and knowledge, they identified a patient population—people with lipodystrophy—who could potentially benefit from leptin treatment.

Lipodystrophy is actually a group of disorders with disparate origins but with a common set of characteristics. Individuals with lipodystrophy lack fatty tissue in the face, neck, or extremities. They sometimes have central obesity and sometimes lack fat tissue altogether. While lipodystrophy is characterized by the loss of fatty tissue in certain areas of the body, tissues such as liver and muscle exhibit significant abnormal accumulation of fat, which impairs metabolic activity. These patients also exhibit resistance to the effects of insulin and are thus at high risk of developing diabetes. They may also have a range of lipid abnormalities. Treatment of lipodystrophy has included the administration of insulin, oral hypoglycemic (blood glucose lowering) agents, and lipid-lowering drugs. In spite of treatment, patients with lipodystrophy continue to have severely high levels of triglycerides, leading to recurrent attacks of acute inflammation of the pancreas; severe problems controlling blood glucose levels, posing risks of diabetic eye and kidney disease; and fat accumulation in the liver, which can result in cirrhosis and liver failure.

Because many people with lipodystrophy have low leptin levels, and because research had demonstrated beneficial effects of leptin on insulin sensitivity and fat metabolism in a number of tissues, researchers in the NIDDK Intramural Research Program and their collaborators investigated whether leptin treatment could ameliorate conditions associated with lipodystrophy. In two small clinical studies of individuals with lipodystrophy treated for short periods of time (3-8 months), leptin therapy had dramatic benefits. In one study of female patients with different forms of lipodystrophy, most of whom also had type 2 diabetes, leptin therapy improved blood glucose levels, lowered triglyceride levels, and decreased liver fat content. In another study, leptin therapy markedly improved insulin sensitivity, lowered lipid levels, and decreased liver fat content in individuals with severe lipodystrophy who also suffered from poorly controlled type 2 diabetes. Patients in these studies were able to reduce or discontinue their diabetes medications.

Seeing such dramatic results, the researchers next examined the effect of long-term leptin therapy (12 months) in patients with severe forms of lipodystrophy and poorly-controlled diabetes. Long-term leptin therapy had similarly remarkable results. Patients had improved blood glucose and blood lipid levels, and decreased fat in their livers. The patients also reported a dramatic reduction in their appetite, which led to moderate reductions in their weight. In addition, patients were able to discontinue or reduce their diabetes medications. These exciting results suggested that leptin was an effective treatment for severe lipodystrophy.

The scientists also examined the effect of leptin on other metabolic abnormalities associated with lipodystrophy. For example, female patients often have irregular or absent menstrual cycles. Leptin treatment was found to be corrective of that condition—eight of eight female patients achieved normal menstrual function following leptin therapy. In a study of 10 patients, leptin effectively improved liver function and reduced liver fat content in people with lipodystrophy and nonalcoholic steatohepatitis, a progressive metabolic liver disease. In a study of 25 patients with lipodystrophy, researchers found that a surprisingly high number had some form of kidney disease. Leptin treatment was found to improve their kidney function. Thus, leptin corrected a broad range of metabolic defects associated with lipodystrophy.

Lipodystrophy can either be inherited or acquired, and can be complete (near total lack of fat) or partial (fat loss in certain parts of the body). Clinical trials

conducted by scientists in the NIDDK Intramural Research Program and their collaborators examined leptin treatment for various forms of lipodystrophy and found that leptin effectively treated all forms tested. These results suggest that leptin is generally effective for treating lipodystrophy, independent of the underlying cause.

Testing Leptin for Treating Lipodystrophy: A Team Effort

The clinical trials testing leptin therapy for lipodystrophy conducted by the NIDDK Intramural Research Program required numerous collaborators, and spawned new collaborations. Leading this effort was Dr. Phillip Gorden, a former NIDDK Director who returned to the laboratory to continue his research. Because leptin was manufactured by industry, the Intramural Research Program and the NIDDK Office of Technology Transfer and Development worked with industry to obtain the leptin needed for the studies. In addition, because lipodystrophy affects the liver and kidneys, scientists in the Intramural Research Program with expertise studying those organs were valuable contributors to the studies. Furthermore, collaborators external to the NIDDK have studied the genetic underpinnings of different forms of inherited lipodystrophy; several genes have now been identified. Finally, many of the patients were evaluated and treated at the NIDDK's Metabolic Clinical Research Unit, a new facility in the NIH Clinical Center that enables scientists to make precise metabolic measurements. It was only through the contributions of all of these collaborators that this translational success story came to fruition.

Looking to the Future

Looking to the future, scientists are continuing research on leptin and exploring approaches for its use in treating other diseases and disorders. As described in this story, knowledge gained from studying a common condition, obesity, led to the discovery of leptin and a treatment for a very rare disease, lipodystrophy. Scientists are now coming full circle by building on the successful clinical studies with leptin in lipodystrophy and applying it to research on common diseases. For example, the NIDDK Intramural Research Program is conducting studies to examine leptin's effects on treating people with other forms of severe insulin resistance and other common metabolic conditions. If leptin proves effective in these cases, these studies would be an example of how research on rare diseases may additionally benefit people with more common diseases and syndromes. The discovery of leptin has led—and continues to lead—to a cascade of exciting and unexpected findings with broad implications for improving health.

Food Intake and Body Weight: Regulation by Apo A-IV in the Brain

Dr. Patrick Tso

Dr. Patrick Tso is Professor of Pathology, Associate Director of the Cincinnati Obesity Research Center, and Director of the Center for Lipid and Atherosclerosis Research at the University of Cincinnati College of Medicine. Additionally, he is the Director of the Cincinnati Mouse Metabolic Phenotyping Center, funded by NIDDK. Dr Tso is a highly respected researcher in the area of lipid (fat) metabolism, a field in which he has worked for over 20 years. At the September 2008 meeting of the NIDDK Advisory Council, Dr. Tso shared insights from his exciting research on how food intake and body weight are regulated by apolipoprotein A-IV (apo A-IV). The following are highlights of his presentation.

Feeling Full after a High-Fat Meal: The Discovery That Apo A-IV Regulates Food Intake

How does a high-fat meal make one feel full? Dr. Tso described his laboratory's research to understand what causes satiety and the insights that have emerged from these studies about the role of a small biologic factor called apo A-IV, which is made in humans and animals. Intrigued by the dramatic increase in intestinal apo A-IV production known to occur after ingestion of dietary fat, Dr. Tso and one of his post-doctoral fellows, Dr. Kazuma Fujimoto, investigated a potential role for apo A-IV in satiety. The researchers conducted their experiments in rodents, and began with a focus on a body fluid called lymph, which varies in composition depending upon what food has been ingested. After a fatty meal, lymph from the abdomen contains an abundance of apo A-IV along with fat absorbed from the food. To assess whether this fluid might curtail food intake, they compared different samples of lymph, some with fat and some without. After administering the lymph samples intravenously into fasting rats, they assessed how much the rats subsequently ate. In describing this study, Dr. Tso shared an anecdote about the experimental design. He recounted that Dr. Fujimoto was concerned that the rats might be too "worried" to eat if a person was nearby. So, Dr. Fujimoto decided to speak to each rat for 15 minutes every day (in his native Japanese) to help the animals feel comfortable around him. This procedure evidently worked, as the rats did eat—but those who had been given the fat-containing lymph ate significantly less. Thus, something in the lymph was signaling that a relatively small meal would be sufficient. The researchers then investigated which component of the lymph might be causing this satiety effect: the fat, or the apo A-IV. After a series of additional experiments, they discovered that it was apo A-IV.

Site of Action: The Brain

Dr. Tso next sought to discover where in the body apo A-IV exerts its effect to reduce food intake. Although apo A-IV was originally found to be produced in the intestine, substantial regulation of appetite occurs in the brain. Thus, Dr. Tso, with another post-doctoral fellow, Dr. Koji Fukagawa, explored whether apo A-IV could reduce food intake when infused directly into the brain. The answer was yes, as determined by further studies in rats.

Building on this research, Dr. Tso and another of his post-doctoral fellows, Dr. Min Liu, found that apo A-IV is not only produced in the intestine, but it is also synthesized in a part of the brain, the hypothalamus, known to play a crucial role in the control of food intake and body weight. In further experiments in rats, they demonstrated that excess apo A-IV in the brain,

from infusions, reduces body weight in parallel to its effect on satiety.

From Feeding to Fullness: Elucidating Biologic Pathways in the Brain

Having illuminated the role of brain apo A-IV in regulating food intake, Dr. Tso and his colleagues next asked: What regulates apo A-IV? They first tested whether brain apo A-IV, like apo A-IV in the intestine, is affected by feeding and fasting. From studies in rats, the researchers found that apo A-IV levels in the brain substantially decreased after fasting, a result that is consistent with their findings regarding the role of apo A-IV in satiety; an animal that had not eaten for a day should not feel full. The researchers next explored the effects of different types of food on apo A-IV levels in the brain. When the rats, after fasting, were given their standard "chow," apo A-IV levels in the brain did not change significantly. If the animals instead ate high-fat food, their brain apo A-IV levels greatly increased.

Dr. Tso and his laboratory also discovered that apo A-IV levels in the brain fluctuate with the circadian rhythm—the day/night cycles. Levels of this satiety-inducing factor were lowest at night, when rats typically eat, and peaked during the day, when rats normally do not eat. Thus, the daily rise and fall of apo A-IV levels mirrored the animals' feeding patterns. The researchers then explored whether the changes in apo A-IV levels were caused by the cycles of light and dark per se, or by the concomitant cycles of feeding and fasting. When they shifted the rats' meal times to the daylight hours (by providing food only during the day), the researchers found that apo A-IV levels changed, too. Dr. Tso concluded that it was the cycles of feeding and fasting that affected apo A-IV levels, rather than daylight and darkness. That is, under normal conditions, apo A-IV levels are low at night (when food is typically available), and the rats are thus able to eat. Their ingestion of food causes apo A-IV levels to rise by the morning hours, which in turn makes the rats too full to eat during the day. After not eating for a while, the apo A-IV levels fall again so that by night time, the rats become hungry and eat.

To further explore the pathway by which apo A-IV causes satiety, Dr. Tso and his research team investigated whether apo A-IV interacts with the hormone leptin. Mice deficient in leptin are strikingly obese, and this hormone also plays a critical role in body weight regulation in humans. The researchers measured apo A-IV levels, and the effects of fasting and feeding, in normal mice and mice that lacked leptin (as a result of a genetic mutation). In the leptin-deficient mice, levels of apo A-IV in the brain were lower than in the normal mice. Additionally, when leptin-deficient mice were given a high-fat meal, the levels of brain apo A-IV did not increase as in normal mice. The scientists then injected leptin into the deficient mice, and found that this led to a restoration of normal levels of apo A-IV. From these and other experiments, the researchers concluded that leptin regulates apo A-IV, and that leptin and apo A-IV interact to reduce food intake and body weight.

Dr. Tso's research team then turned their attention to other factors in the brain known to regulate food intake, collectively referred to as the melanocortin system, to determine whether these factors interact with apo A-IV. Again using rodents as a model system, the researchers found that apo A-IV and a major component of the melanocortin system, called POMC, are present in the same brain cells, and both apo A-IV and POMC levels are low during fasting. Administering apo A-IV led to an elevation in POMC levels as well. This research, together with additional studies, demonstrated that apo A-IV also interacts with the melanocortin system to inhibit food intake.

Conclusions—Apo A-IV

Dr. Tso's research on apo A-IV has yielded novel insights into the regulation of food intake and satiety. In concluding his presentation, Dr. Tso noted that apo A-IV has other functions as well, related to fat metabolism and other biologic processes. By

shedding light on the regulation of satiety, this research will also advance understanding of what could go awry in obesity, with implications for potential intervention approaches.

Dr. Tso acknowledged the contributions of the scientists who worked with him on these studies when they were post-doctoral fellows in his laboratory: Drs. Kazuma Fujimoto, Koji Fukagawa, and Min Liu. Additionally, Dr. Tso thanked his long-time collaborator on this research, Dr. Stephen Woods, who is also a Professor at the University of Cincinnati.