

University of Colorado Denver
Nutrition Obesity Research Center
Annual Report: May 2009–April 2010
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Website: <http://medschool.ucdenver.edu/ColoradoNORC>

Organization and Goals

The major goal of the Colorado Nutrition and Obesity Research Center (NORC) at the University of Colorado Denver (UCD) is to create an environment in which researchers are able to work together to conduct high-quality research in nutrition and obesity. The Colorado NORC helps create that environment by facilitating interaction and collaboration among investigators working at different levels of basic and clinical investigation, from gene to cell to organ to animal model to human to clinical to community intervention.

The Specific Aims of the Colorado NORC are:

- To enhance an existing strong and well-funded research base in nutrition and obesity by providing an infrastructure to support ongoing nutrition and obesity research, providing measurements and expertise that would not otherwise be available to individual investigators, and providing measurements in a cost-effective manner.
- To promote interdisciplinary collaborative, vertically integrated research among members of the Colorado NORC research base.
- To strengthen several NIH-funded training programs in nutrition and obesity that provide education and research training for graduate, undergraduate, and medical students and that assist promising young M.D.s and Ph.D.s in becoming independent investigators in the field of nutrition and obesity.
- To translate basic and clinical research into programs to improve health and wellness and reduce obesity in the community.
- To improve the quality of nutrition and obesity information provided to the public.

To accomplish these specific aims, we support an Administrative Core and three Scientific Core Laboratories: Energy Balance, Metabolic, and Clinical. We also support a Pilot and Feasibility program and an enrichment program.

Core Laboratories

Administrative Core:

James O. Hill, Ph.D., Director; Robert H. Eckel, M.D., Co-Associate Director; Daniel Bessesen, M.D., Co-Associate Director; Andra Price, Administrator

Energy Balance Core:

Wendy Kohrt, Ph.D., Director; Paul MacLean, Ph.D., Co-Director; Edward Melanson, Ph.D., Co-Director

Mass Spectrometry Core: (This Core has been discontinued)

Uwe Christians, Ph.D., Director

Metabolic Core:

Jed Friedman, Ph.D., Director; Paul MacLean, Ph.D., Co-Director

Clinical Component: (This component has become a Clinical Core Laboratory)

Holly R. Wyatt, M.D., Director

External Review Board, August 2005 through July 2010:

- **David D'Alessio, M.D.**, Professor of Medicine, Albert Vontz Chair in Diabetes Research, University of Cincinnati
- **Lynis Dohm, Ph.D.**, Professor, Department of Physiology, Brody School of Medicine, East Carolina University
- **Bret Goodpaster, Ph.D.**, Assistant Professor of Medicine, The University of Pittsburgh
- **Ruth Harris, Ph.D.**, Professor, Department of Foods and Nutrition, University of Georgia
- **Eric Ravussin, Ph.D.**, Professor and Chief, Health and Performance Enhancement, Douglas L. Gordon Chair in Diabetes and Metabolism, Pennington Biomedical Research Center
- **Dale Schoeller, Ph.D.**, Professor, Nutritional Sciences, University of Wisconsin, Madison

Pilot and Feasibility Studies

The UCD NORC Pilot and Feasibility (P&F) grant program helps young investigators make the transition from beginning scientists to independently funded investigators. Here we list our active P&F recipients that received NORC funding in the budget period of August 1, 2009, to July 31, 2010.

Audrey Bergouignan, Ph.D.

A novel tracer method to determine the mechanisms of reduced dietary fat oxidation in obesity and reduced obesity. Dietary fat is preferentially trafficked away from oxidation and toward storage in obese and weight-reduced individuals. This altered partitioning of dietary fat may be caused by 1) a greater uptake of dietary fat by adipose tissue as compared to oxidative tissues (liver and muscle); 2) mitochondrial dysfunction in oxidative tissues resulting in a diversion of dietary fat to intracellular lipids (triacylglycerol, diacylglycerol, and ceramide); and/or 3) adequate mitochondrial capacity, but incomplete oxidation of dietary fat due to competition in the tricarboxylic acid cycle resulting in the generation of other lipid species (acylcarnitines) in oxidative tissues. The mechanisms responsible for this altered partitioning are, however, poorly delineated due to methodological limitations. Our objectives are 1) to combine stable isotope tracer and lipidomics techniques to develop methods for following dietary fat into lipid fractions of oxidative tissues, and 2) to apply these methods to investigate the alterations of the dietary fat trafficking in obese and reduced-obese rats. Developing a method to track the movement of dietary fat into lipid species will provide a novel tool for research in nutrition and obesity. The proposed studies in rats will be critical in developing and validating these methods before applying them to studies in humans.

Karim El Kasmi, M.D., Ph.D.

Role of KC activation in the pathogenesis of parenteral nutrition associated liver injury.

Preterm infants, infants, and children with functional and/or anatomical conditions that prevent adequate enteral feeding (including short bowel syndrome (SBS), congenital malformations, necrotizing enterocolitis [NEC], and intestinal failure [IF]) depend on total parenteral nutrition (TPN) for survival. An important complication is the development of liver injury and eventually biliary cirrhosis. This TPN-associated liver injury (TPNALI) is the most common cause of death in these infants and is the key factor contributing to the need for combined liver-intestinal transplantation. TPNALI is more common and more severe in the presence of intestinal disturbances with underlying inflammation (e.g., NEC, IF, and SBS). A novel mouse model for this disease allows us to conduct mechanistic studies. We propose that 1) during TPN liver resident macrophages, the Kupffer cells (KCs), are activated and promote an inflammatory milieu that leads to liver injury, and 2) that toll-like receptor pathways in KCs are engaged by bacterial cell wall products and bacteria that translocate into the portal circulation due to reduced intestinal motility and increased leakiness of the intestinal barrier, in conjunction with bacterial overgrowth in the gut lumen. We hypothesize that treatment with intravenous omega-3 fatty acids will alleviate liver damage and reduced KC activation.

Henry Galan, M.D.

Placental oxygen consumption heterogeneity and its impact on transport. The hypothesis of this grant proposal is that the human placenta has two distinctly different areas of oxygen consumption. In one area (stem and intermediate villi) there is a very high metabolic rate and a tissue rich in mitochondria. In another area (terminal villi) there are relatively few mitochondria and a low metabolic rate. The validity of this hypothesis and its functional implications will have very important implications in disease states such as intrauterine growth restriction (IUGR) pregnancies. Two Specific Aims are designed to test this hypothesis. The first Aim examines the distribution of carbohydrate versus amino acid transporters in these two placental areas, with the assumption that the non-energy dependent transporters (e.g., glucose transporters) will be more prominent in the low oxygen consuming area and that energy dependent transporters (e.g., CAT transporters for arginine and lysine) will be concentrated in the stem and intermediate villi. The second Aim follows on the above by testing that mitochondrial density is different in these two areas using a variety of molecular techniques for this confirmation. In summary, we believe this grant has two Aims tightly connected and will impact the design of studies of placental transport and metabolism in various disease states.

Erin D. Giles, Ph.D.

Progesterone receptor expression and obesity-associated postmenopausal breast cancer.

Obesity has been linked with an increased incidence and progression of postmenopausal breast cancer; however, the mechanisms underlying this adverse effect of obesity remain poorly understood. We have merged well-characterized models of breast cancer (methylnitrosourea), obesity (obesity-prone rats), and menopause (ovariectomy, OVX) to study obesity-associated tumor promotion after menopause. We have observed higher progesterone receptor (PR) messenger RNA (mRNA) levels in tumors from obese compared to lean. Data from the Women's Health Initiative would suggest progesterone and PR may be important for postmenopausal risk. This study will investigate if high levels of PR in obese tumors predict tumor hormone receptor status during OVX-induced weight gain (when the tumor-promoting effects of obesity emerge) and a metabolic survival advantage through a glycolytic-lipogenic gene expression profile. The

tumor expression data will be analyzed in the context of *in vivo* nutrient tracer data that already exists for these tumors, which includes tissue-specific glucose uptake and lipid trafficking. This work will provide a novel molecular and metabolic mechanism for the emergence of obesity-associated tumor promotion after OVX that may explain the link between obesity and post-menopausal breast cancer. The preliminary data derived from this work will help the investigator secure funding as in independent investigator.

Kent Hansen, Ph.D.

The effects of estrogen and exercise on adipogenesis. The proposed pilot research will partner with an ongoing randomized controlled trial (NIH R01 AG018198) studying the effects of exercise on abdominal fat in premenopausal women during 5 months of sex hormone suppression with gonadotropin releasing hormone agonist (GnRHAG). The global aim of the proposed research is to determine whether exercise can attenuate the differentiation of adipose tissue stem cells becoming mature adipocytes during the first 60 days of sex hormone suppression (GnRHAG+placebo). To meet these aims, abdominal fat biopsies will be obtained from women in the parent trial at baseline and after 30 and 60 days of concomitant exercise and GnRHAG therapy. A novel approach will be used to determine whether exercise can attenuate differentiation patterns of stem cells under sex hormone suppression with or without exercise. Briefly, women will drink daily doses of deuterium (2H)-enriched water (2H₂O) for the first 53 days of GnRHAG therapy. Adipocytes that are newly formed during this interval will be unique in that the DNA will be endogenously labeled with 2H. We will measure 2H labeling of DNA and cellularity analysis, as an index of adipogenesis and hypertrophy, and expression of adipogenic (e.g., PPAR γ and C/EBP α) mRNA in adipose tissue cell fractions.

Carrie McCurdy, Ph.D.

Regulation of obesity-induced adipose tissue inflammation by p85 α PI 3-kinase. Currently, 180 million Americans are classified as being overweight or obese. Insulin resistance, a primary disorder of obesity, increases the risk of many metabolic-related diseases, including type 2 diabetes (T2DM). Recent studies have linked obesity-induced insulin resistance to chronic low-grade inflammation, which is characterized by adipose tissue infiltration of immune cells, in particular macrophages. The mechanisms underlying these processes are incompletely understood. A key observation is that obesity leads to up-regulation of p85 α , the regulatory subunit of PI3-kinase, in parallel with reduced insulin sensitivity in metabolic tissues like muscle and adipose. Our data demonstrate that mice expressing only one functional allele of the p85 α gene become obese on a high-fat diet but demonstrate remarkably reduced macrophage infiltration and inflammatory gene expression in adipose tissue and are protected from obesity-induced insulin resistance. This observed dissociation between obesity and inflammation has led us to hypothesize that p85 α controls key pathways in adipose tissue that lead to macrophage recruitment and adipose tissue inflammation that subsequently result in insulin resistance. To address this hypothesis, we will investigate the role of p85 α in regulating adipose tissue insulin sensitivity and inflammation using transgenic mouse models and *in vitro* cell culture systems.

Makoto Miyazaki, Ph.D.

The role of stearoyl-CoA desaturase-1 in regulation of atherosclerosis. Stearoyl-CoA desaturase-1 (SCD1) is the regulated enzyme that synthesizes oleate and palmitoleate that are normally incorporated into triglycerides and cholesteryl esters. Overexpression of SCD has been linked to human cardiovascular disease. We hypothesize that overexpression of SCD1 and

overconsumption of dietary oleate can lead to the development of atherosclerosis and hypertriglyceridemia, and that the absence of SCD1 gene expression and the lack of oleate in the apoE^{-/-}SCD1^{-/-} animals attenuates the formation of cholesteryl ester and triglyceride-rich very low density lipoprotein particles that would otherwise contribute to the hypertriglyceridemia and the development of atherosclerosis in these animals. The Specific Aims of this proposal will be: 1) to test whether deletion of the SCD1 gene from ApoE^{-/-} mice attenuates the hyperlipidemia and atherosclerotic lesion development; 2) to test whether the overexpression of a constitutively active SCD1 in ApoE^{-/-} mice accelerates the hyperlipidemia and the development of atherosclerosis; and 3) to test whether dietary monounsaturated fatty acid accelerates hyperlipidemia and the development of atherosclerosis in ApoE^{-/-} mice and ApoE^{-/-};SCD1^{-/-} mice.

Andrea Salzberg, M.D.

Neuronal responses to food-related stimuli with weight loss. This pilot study is designed to investigate how neuronal, behavioral, and hormonal responses change with weight loss. We chose functional magnetic resonance imaging (fMRI) as our modality to evaluate neuronal responses to food-related stimuli because of our previous experience and because of fMRI's ability to provide insight into the complex systems controlling food intake behavior. While previous studies have shown differences in neuronal responses to food-related stimuli in obese, reduced, and lean individuals, previous investigations have not evaluated the effect of weight loss on these neuronal responses nor have they correlated behavioral or hormonal measures to the neuronal responses. Our pilot study is designed to evaluate differences in neuronal, behavioral, and hormonal responses in overweight/obese individuals at baseline and after weight loss. To recruit these subjects, we will collaborate with Dr. Holly Wyatt and enroll subjects involved in her longitudinal weight-loss study. In addition, we propose to compare our overweight/obese subjects to a lean cohort currently being studied by Dr. Salzberg's mentor, Dr. Cornier, and to follow the subjects during weight-loss maintenance.

Brian L. Stauffer, M.D.

Dietary linoleic acid to improve heart failure in humans. The general aim of this proposal is to translate findings from an animal model of heart failure, previously supported by a NORC pilot grant (G. Sparagna, PI, 7/1/06–6/30/09), into humans and determine whether cis-9, cis-12, linoleic acid (cis-LA) supplementation improves vascular endothelial and cardiac function in humans with chronic heart failure. Dietary cis-LA supplementation in a rodent model of heart failure has demonstrated remarkable results, with improved morbidity and mortality as well as stabilization of myocardial mitochondrial and cardiac systolic function through improvements in the mitochondrial phospholipid, cardiolipin. Similar to the rat, mitochondrial dysfunction in the failing human heart contributes to impaired systolic function and is associated with abnormalities in myocardial cardiolipin isoforms. The ω -6 fatty acid (a specific class of fatty acids separate from omega-3 fatty acids) supplementation proposed in the current application, while currently recommended by American Heart Association guidelines and supported by the clinical literature, is a completely novel intervention in humans with heart failure. We hypothesize that 12 weeks of cis-LA supplementation will improve the endothelial, cardiac, and mitochondrial dysfunction that are present in human heart failure. The current application is to provide the critical pilot data in humans.

Stephanie Thorn, Ph.D.

Mechanisms for fetal hepatic programming of insulin resistance and glucose production during intrauterine growth restriction. Increasing human and animal experimental evidence supports the concept that IUGR results in increased susceptibility to metabolic disease such as obesity and diabetes in later life. In animal models of IUGR the expression of key gluconeogenic genes, such as phosphoenolpyruvate carboxykinase (PEPCK), are induced during fetal development. The increase in PEPCK gene expression along with increased hepatic glucose production (HGP) appears to persist into adulthood and is a hallmark of diabetes and insulin resistance. The molecular mechanisms underlying the origin of this disorder in the fetus are largely unknown. The long-term goal of this proposal is to understand the fetal origins of hepatic insulin resistance and failure to regulate HGP at the molecular level. We intend to achieve this goal with *in vivo* and *in vitro* investigations in our sheep model of IUGR. The results of these investigations will generate important, novel concepts regarding the underlying fetal mechanisms responsible for increased susceptibility to adult metabolic disease in individuals who experienced nutrient or growth restriction *in utero*.

Adam Tsai, M.D.

A pilot trial comparing health care-based and community-based approaches to weight management. Obesity is perhaps the most common problem seen by primary care providers and one of the least likely to be treated, partly because it is not reimbursed. Debate is ongoing between those who believe that weight-loss treatment should be provided within the health care system and those who believe that weight management should not be “medicalized.” Evidence is needed to compare weight-loss strategies in different settings of care so that health care payers can make evidence-based decisions about reimbursement. The proposed study will recruit a sample of predominantly low-income, ethnic minority patients from Denver Health and will randomly assign them to attend Weight Watchers for 13 weeks or to participate in an intervention based at a primary care office. The health care intervention will include a series of visits with a nurse weight-loss counselor, consultation with a physician, and the opportunity to use meal replacements or weight-loss medication for a 13-week period. The primary outcome of the study is weight change. The health care intervention is hypothesized to be more effective than the Weight Watchers intervention. The results of the study are intended to serve as pilot data for a grant to test this hypothesis in a larger sample.

Funding Derived From Previous Pilot and Feasibility Studies

The following list itemizes grant funding derived from previous pilot and feasibility studies, outlined by title, principal investigator, affiliation, and funding project period.

- Improving Protein Balance in Postoperative Neonates Using Acids and Narcotic Analgesia. Marianne Anderson, M.D., at UCLA, Neonatal-Perinatal Medicine. 08/03–06/08
- Placental Oxygen Consumption Heterogeneity and Its Impact on Transport. Juan Arroyo, Ph.D., at University of Kansas Medical Center. 05/09–04/11
- Influence of Energy Flux on Tonic Sympathetic Support of Resting Metabolic Rate in Older Adults. Christopher Bell, Ph.D., at Colorado State University, Health & Exercise Science. 02/04–01/10

- Intramuscular Triglyceride Turnover and Insulin Action. Bryan Bergman, Ph.D., at UCD, Medicine-Endocrinology, Metabolism, and Diabetes. 07/04–06/05; 09/05–07/10; 05/10–04/12
- Cytokine Regulation of SREBP-1 in Human Skeletal Muscle. Michael Bizeau, Ph.D., at Metro State College Denver, Health Professions. 08/02–06/06
- Nutrient Regulation of Skeletal Muscle Signal Transduction in an Ovine Model of Intrauterine Growth Restriction. Laura Brown, M.D., at UCD, Pediatrics-Neonatology. 07/08–06/11
- The Effects of Short-Term Overfeeding on Insulin Sensitivity. Marc Cornier, M.D., at Denver Health and Hospital Authority and UCD, Medicine-Endocrinology, Metabolism and Diabetes. 07/01–06/07; 05/07–06/12; 09/07–08/10
- Effects of Estrogen and Raloxifene on the HPA Axis, IL-6, & Visceral Fat in Menopausal Women. Wendee Gozansky, M.D., M.P.H., at UCD, Medicine-Geriatrics. 07/03–06/06; 08/05–07/10; 06/06–04/10
- Antioxidants and Cell Signaling During Myocardial Ischemia/Reperfusion. Karyn Hamilton, Ph.D., R.D., at Colorado State University, Health and Exercise Science. 04/05–03/10
- Development of a Hepatic Lipase Assay Without the Intravenous Injection of Heparin. John Hokanson, Ph.D., M.P.H., at UCD, Colorado School of Public Health. 09/01–08/05; 12/02–11/07
- Effects of a HFD on Energetics, Metabolite Profile and Insulin Action in OP and OR Rats. Matthew Jackman, Ph.D., at UCD, Medicine-Endocrinology, Metabolism and Diabetes. 04/02–03/04
- DHEA and Exercise: Effects on Muscle IGF-1 and TNF- α . Catherine Jankowski, Ph.D., at UCD, Medicine-Geriatrics. 07/06–06/10
- Metabolic Characteristics of Protease Inhibitor-Treated Patients With Atrophy of the Subcutaneous Adipose Tissue of the Extremities. Lisa Kosmiski, M.D., at UCD, Medicine-Endocrinology, Metabolism and Diabetes. 08/00–06/06; 09/03–02/08; 03/09–02/13
- Effects of High-Fat Feeding on Skeletal Muscle Sterol Response Element Binding Protein Expression. Paul MacLean, Ph.D., at UCD, Medicine-Endocrinology, Metabolism and Diabetes. 02/05–11/07; 03/86–03/12
- Nutritional Strategy for Increased Mitochondrial Biogenesis. Benjamin Miller, Ph.D., at Colorado State University, Health and Exercise Science. 03/09–02/14
- The Role of Stearoyl-CoA Desaturase-1 in Regulation of Atherosclerosis. Makoto Miyazaki, Ph.D., at UCD, Medicine-Renal. 07/10–06/12
- Insulin Control of Fat Regulation and Exercise in Teens. Kristen Nadeau, M.D., at UCD, Pediatrics-Endocrinology. 07/04–03/11
- Evaluation of the Potential Role of Ingestion of Non-protein Amino Acids in Neurodegenerative Disease. Nichole Reisdorph, Ph.D., at UCD and National Jewish Medical and Research Center, Immunology. 09/06–08/10
- ERK1/2 Inhibition and Induction of Apoptosis by the Dietary Polyphenol EGCG in Human Colon Cancer Cells. Pamela Rice, Ph.D., at UCD, Gastroenterology. 01/04–12/06; 01/03–01/05
- Leucine Regulation of Pancreatic B-Cell Development in Fetal Growth Restriction. Paul Rozance, M.D., at UCD, Pediatrics-Neonatal/Perinatal. 09/09–08/14

- Neuronal Responses to Food-Related Stimuli with Weight Loss. Andrea Salzberg, M.D., at UCD, Medicine-Endocrinology, Metabolism and Diabetes. 07/09–06/10
- Class-Specificity of Fatty Acid Effects on Endothelial Function, Insulin Sensitivity, Cardiac Function, and Inflammation. Irene Schauer, M.D., Ph.D., at UCD, Medicine-Endocrinology, Metabolism and Diabetes. 09/07–07/12; 10/07–09/11; 2009–2010
- The Mechanism of the Prevention of Heart Failure with Linoleic Acid Supplementation. Genevieve Sparagna, Ph.D., at University of Colorado Boulder, Integrative Physiology. 05/08–04/10; 03/09–03/11
- The Role of Sphingolipids in Insulin Action. Scott A. Summers, Ph.D., at The University of Utah, Biochemistry and Medicine. 08/01–05/06; 07/04–06/07; 09/06–08/08; 03/08–02/09; 07/08–04/13
- Mechanisms for Fetal Hepatic Programming of Insulin Resistance and Glucose Production During Intrauterine Growth Restriction. Stephanie Thorn, Ph.D., at UCD, Pediatrics-Neonatology. 07/08–06/10
- Effect of Insulin and Estrogen on Whole Body and Regional Lipolysis after Menopause. Rachael Van Pelt, Ph.D., at UCD, Medicine-Geriatrics. 05/02–04/08; 09/07–08/12
- Myocardial Substrate Utilization in Patients with Dilated Cardiomyopathy. Eugene Wolfel, M.D., at UCD, Medicine-Cardiology Medicine. 01/04–12/06; 09/02–06/08
- Genetic Determinants of Long-Term Weight Loss. Holly Wyatt, M.D., at UCD, Medicine-Endocrinology, Metabolism, and Diabetes. 08/06–07/11

Scientific Advancements/Accomplishments

The overall impact of the NORC on the nutrition and obesity fields and on nutrition and obesity research within Colorado has been substantial. This impact has increased substantially over the past 5 years due to an increase in the number of researchers studying nutrition and obesity, an increase in the total funding for nutrition and obesity research, an increase in the quantity and quality of publications from our research base, an increase in collaboration among members of our research base, success in advancing the careers of our junior investigators and fellows, development of the Anschutz Health & Wellness Center, and opportunities for a new UCD research initiative in obesity.

Specific Accomplishments

Women's Health

1. At our institution we have an excellent group of researchers who study mammary gland biology, with interests in mammary gland development, lactation, and disease development. During the past grant cycle, a translational unit has emerged from this research base with the extensive support of NORC Clinical, Energy Balance, and Metabolic cores, which has an interest in the impact of obesity on mammary gland biology. While this unit has only recently emerged within our research base, we expect it to grow substantially in the next cycle of the NORC because of the number of grant applications currently in review or in preparation. Several of these researchers are part of a program project grant focused on mammary gland development (**Drs. Anderson, Neville, McManaman, MacLean, and McCurdy**). Studies

have been initiated and proposed in their recent renewal application that are focused on the impact of hyperinsulinemia and peripheral insulin resistance on the changes in epithelial cell and adipocyte biology during pregnancy through the transition to lactation. These researchers are particularly focused on the signaling pathways that coordinate the metabolic and morphological changes as the mammary gland prepares for lactation. They have joined forces with **Drs. MacLean, Schedin, Anderson, Giles, Jackman, and Thor**, who have begun working together to study the impact of energy balance and fuel metabolism on breast cancer tumor promotion. From their overlapping interests, they developed a preclinical model to study the impact of obesity and energy balance on postmenopausal breast cancer. With this model, they have observed a novel relationship between the energetic efficiency of the rapid period of weight gain after experimentally modeled menopause (surgical ovariectomy) and long term mammary tumor growth and survival. They are using the model to examine the effects of metformin therapy on tumor promotion during this time.

2. The effect of maternal obesity on the fetus and early childhood obesity is an area of intense clinical and basic science interest at the University of Colorado. Pregnancy is a critical period when an exposure may have a lifelong effect on the structure or function of an organ, tissue, or body system through biological programming. **Dr. Friedman** and collaborators at the University of Oregon and University of Utah have developed a nonhuman primate model of maternal obesity. This collaboration led to several novel observations in the fetal liver, brain, and skeletal muscle that suggest that maternal lipids in the diet result in an early obesity phenotype in the developing fetus, particularly the fetal liver. Based on this work, **Dr. Carrie McCurdy** was successful in obtaining an NIH-K12 award to study the fetal programming of insulin resistance in skeletal muscle from obese Japanese macaques. Simultaneously, Dr. Friedman developed a novel transgenic mouse model that genetically re-balances the omega-3 to omega-6 ratio on a high-fat diet, and prevents the pro-inflammatory response in diet-induced obesity. This appears to be protective of the fetal programming effects in the offspring despite maternal obesity. The effects of maternal under-nutrition on the fetus are being investigated by **Dr. Bill Hay** and colleagues using a sheep model of IUGR. The IUGR model is being used by **Dr. Laura Brown** (K12) to study skeletal muscle fetal programming, **Dr. Paul Rozance** (K01) is studying the fetal pancreas, and **Dr. Stephanie Thorn** (NIH-F32) is studying how IUGR affects hepatic insulin resistance in the fetal liver.
3. **Dr. Wendee Gozansky** is conducting clinical research to promote health in older women. Her SCALE protocol (Sex hormones, Cortisol, and Adiposity Loss via Exercise; supported by K23 AG026784, Paul B. Beeson Career Development Award in Aging, “Cortisol Metabolism & Central Adiposity After Menopause”) is investigating whether estrogens and exercise-induced weight loss modulate peripheral glucocorticoid metabolism in postmenopausal women. Dr. Karen Villalon is a collaborator on the SCALE project and has a peripheral quantitative computer tomography (pQCT) sub-study to evaluate the impact of exercise-induced weight loss on cortical and trabecular bone (F32 AG035460). They are also entertaining the possibility of adding pQCT measures to these protocols.
4. **Dr. Lynn Barbour** received funding this year through the NIH for both a 5-year R01, which studies the maternal metabolic characteristics of obese mothers who give birth to infants with excess adiposity, and a 2-year R21, which studies the optimal macronutrient diet for women with gestational diabetes.

AIDS

1. **Dr. Lisa Kosmiski (with Drs. Bessesen, Horton, and Grunwald)** is using an HIV model to study body weight and body fat regulation. The HIV lipodystrophy syndrome is characterized by extensive loss of subcutaneous fat. It is also associated with increased resting energy expenditure (REE) even after adjustment for lean body mass. In the past year, we have used dual-energy X-ray absorptiometry (DXA) modeling to better understand the organ-tissue basis of the hypermetabolism associated with this syndrome. DXA modeling can be used to predict the REE of a study population. Based on DXA modeling of five organ-tissue components, subjects with HIV lipodystrophy (LD) should have an REE similar to healthy controls. However, the measured REE of subjects with HIV LD is significantly greater than controls. Furthermore, this method has suggested that the increased REE associated with HIV LD is due to hypermetabolism in skeletal muscle.

Reduction in Health Care Costs

1. **Dr. James Hill** and colleagues are implementing science-based worksite strategies to prevent weight gain and reduce health care costs. Several pilot programs are underway and these programs include programming from America On the Move and Colorado Weigh.
2. Through an initial NIH grant **Dr. Debbie Main** and collaborators established a community-based participatory research initiative called Taking Neighborhood Health to Heart (TNH2H). This community-academic partnership involves the University of Colorado Denver and the five neighborhoods in and around the former Stapleton airport site, one of the largest urban renewal projects in the United States. Although originally conceived as a project to study the impact of the Stapleton redevelopment on surrounding neighborhoods, it has been expanded to study the impact of all neighborhood environments on health and health disparities. We have a particular focus on physical activity and healthy eating – and how both are influenced by built and social environments within very diverse neighborhoods. We have collected systematic health data on 950 adults living in five neighborhoods: objective measures of the built environments surrounding their households (including streets and parks), and audits of available resources for healthy eating and physical activity, including the availability and cost of food in all local stores. Our TNH2H coalition has expressed strong interest in decreasing risk of chronic health conditions (including overweight and obesity) through programs, policies, and environmental changes.

Obesity

1. **Dr. Cornier** and colleagues (**Drs. Bessesen, Salzberg, and Tregellas**) examined the effects of short-term overfeeding on the neuronal responses to food-related visual stimuli in 25 thin individuals. fMRI was performed after 2 days of eucaloric energy intake and after 2 days of 30% overfeeding in a counterbalanced design. fMRI was performed while the subjects were presented with visual stimuli in three different categories: neutral control objects, foods of neutral hedonic value, and foods of high hedonic value. Measures of appetite were obtained by using visual analogue scales before and after meals. In the eucaloric state, pictures of foods of high hedonic value elicited greater activation of neuronal regions than did neutrally rated foods, which is consistent with visual processing and attention (inferior temporal visual cortex, posterior parietal cortex, premotor cortex, and hippocampus) and with activation of

the hypothalamus. Two days of overfeeding led to significant attenuation of these responses. Overfeeding also resulted in reduced hunger ratings and increased satiety ratings. These findings emphasize the important role of external food-related visual cues, and suggest that there are interactions between external visual sensory inputs, energy balance status, and brain regions important in the homeostatic regulation of energy intake.

2. **Dr. Richard Johnson's** laboratory has continued to explore the potential role of excessive fructose intake in the epidemic of obesity. Some of our key findings have included the demonstration that excessive intake of fructose in adult men (200 g/d for 14 days) can induce most features of metabolic syndrome within 2 weeks, including an elevation of blood pressure. They also identified a relationship between fructose intake and blood pressure in the NHANES trial. In experimental studies they found that sucrose could induce fatty liver and T2DM in diabetes-prone rats even in the setting of caloric restriction. They also have found that uric acid can induce intracellular oxidative stress and reduce adiponectin production in cultured adipocytes.
3. **Dr. Paul MacLean** (collaborators **Drs. Higgins, Jackman, Bessesen, Giles, Hill, and Wyatt**) has studied the biology of the reduced obese rat. Weight loss is accompanied by several metabolic adaptations that work together to promote rapid and efficient regain. These adaptations include a large energy gap between appetite and expenditure needs, the preferential use of carbohydrates for energy production, the preferential trafficking of dietary fats to adipose tissue, an increase in the number of small adipocytes early in relapse, and the defense of a lower weight with the same adiposity signals. In their most recent study, they showed that regular exercise counters many of these adaptations and attenuates the rate of regain early in exercise and lowers the defended weight after relapse. These observations indicate that regimented exercise attenuates the metabolic adaptations to weight reduction in a manner that reduces the propensity to regain lost weight.
4. **Dr. Lynn Barbour** and **Dr. Jed Friedman**, along with **Dr. Dana Dabelea** in the school of Public Health, are working together on the effects of maternal obesity on the early origins of obesity in newborn infants and children. Dr. Barbour is a board certified Endocrinologist with special research interest in the medical problems of pregnancy; she is the medical director of both the high-risk pregnancy clinic and the diabetes and pregnancy clinic at the University of Colorado. Dr. Dabelea is a national/international leader in the epidemiology of diabetes and obesity and its effect in pregnancy on the life course of disease. Dr. Friedman has special research interests and expertise in maternal metabolism, particularly obesity, diabetes, and the molecular regulation of metabolism. Drs. Barbour, Friedman, and Dabelea have successfully assembled a unique cadre of investigators who work on basic, clinical, and epidemiologic aspects of pregnancy, obesity, and maternal-fetal development in humans and novel animal models (mouse, nonhuman primate, sheep). All studies are aimed toward understanding the pathophysiology of major clinical problems associated with maternal obesity/gestational diabetes and its impact on early origins of pediatric obesity in infants and children. They have established collaborations and bridges between clinical investigators in ob-gyn and research programs in Endocrinology, Neonatology, Pediatrics, and Medicine-GI that have enhanced the academic environment and research infrastructure across several departments and institutions in Denver and the State of Colorado. Dr. Friedman has close collaborations with both clinicians and basic researchers employing complementary skills in

medicine and biology, and together they have a keen interest in growing both the translational and basic scientific base with strong ties to public and community health.

5. **Dr. Ed Melanson's** research team has published several papers regarding the effect of exercise on 24-hour fat oxidation and fat balance. These studies have demonstrated that when energy balance is maintained (energy intake = energy expenditure), exercise does not induce changes in 24-hour fat oxidation. The implication of these results is that for exercise to induce an increase in fat oxidation (and therefore induce loss of fat mass), a negative energy balance must be induced. This has implications for how exercise should be prescribed for weight loss. In addition, they have completed studies to determine the effect of increasing dietary fat intake on 24-hour fat oxidation and skeletal muscle oxidative capacity. Specifically, they have performed studies to determine the effects of obesity and exercise training. The results of these studies have recently been submitted in manuscript form for peer review. The primary finding of these studies is that when dietary fat increases, fat oxidation increases, but not enough to match the increase in fat intake, resulting in positive fat balance. Additionally, when studied under energy balance conditions, there is no difference in the response in lean, obese, and endurance-trained individuals. At the skeletal muscle level, they also observed changes in oxidative capacity that are again similar in the three groups studied. Dr. Melanson is the director of the Metabolic Calorimeter Core at UCD. In this role, he oversees the operation of the room calorimeter located in the CTRC. They installed a new gas analysis system in the past year that greatly improved the quality of the data, and this led to a peer-reviewed publication in *AJP*. They are also collaborating with investigators at the NIH facility in Bethesda, Maryland, to develop new approaches to data processing with calorimeter data. These efforts are ongoing.
6. **Drs. Bergman, Perrault, Eckel, Bessesen, and Cornier** are collaborating to study how muscle lipids may influence insulin sensitivity. In particular, they are testing the hypothesis that intramuscular triglyceride synthesis is protective against insulin resistance during periods of increased lipid uptake into muscle.
7. **Dr. Hill** and colleagues (**Drs. Wyatt, Melanson, and Grunwald**) have studied the role of dietary fat in body-weight regulation. In one study published last year, they studied the relationship between dietary fat content and voluntary energy intake measured under controlled conditions. Twenty-two non-obese subjects were studied for 4 days on each of three diets, which included core foods designed to contain 26%, 34%, and 40% fat, respectively, of total calories and *ad libitum* buffet foods of similar fat content. All diets were matched for determinants of energy density except dietary fat. Subjects consumed two meals per day in an inpatient unit and were provided the third meal and snack foods while on each diet. All food provided and not eaten was measured by research staff. Voluntary energy intake increased significantly as dietary fat content increased ($p=0.008$). On the 26% dietary fat treatment, subjects consumed 23.8% dietary fat (core and ad lib foods combined) and $2,748 \pm 741$ kcal/d (mean \pm SD); at 34% dietary fat, subjects consumed 32.7% fat and $2,983 \pm 886$ kcal/d; and at 40% dietary fat subjects consumed 38.1% fat and $3,018 \pm 963$ kcal/d. These results show that energy intake increases as dietary fat content increases across the usual range of dietary fat consumed in the United States. Even small reductions in dietary fat could help in lowering total energy intake and reducing weight gain in the population.

8. In a second study, **Dr. Hill** collaborated with Dr. Joe Donnelly. Students attending The University of Kansas were randomized to *ad libitum* diets containing less than 25% of energy from fat (low fat [LF]), 28% to 32% of energy from fat (medium fat [MF]), or more than 35% of energy from fat (high fat [HF]). Participants consumed two meals per day during the week and one meal per day during the weekend in our cafeteria where amount and composition of energy intake was measured by a novel system using digital photographs of foods. All meals and snacks taken outside the cafeteria were measured by dietary recall or records. Energy intake and weight gain increased with increasing level of dietary fat and was not influenced by gender. LF gained 0.02 ± 3.1 kg, MF gained 0.7 ± 2.5 kg, and HF gained 0.94 ± 2.2 kg ($p < 0.00$ for trend). Longitudinal mixed modeling showed that the effects of dietary fat was via energy intake, and that percentage of energy from fat did not independently affect body weight. As the fat content of the diet increased, energy intake and weight gain also increased in a dose response fashion. LF diets may contribute to weight maintenance and HF diets may promote weight gain.

Recently, **Dr. Hill** collaborated with Dr. David Allison and other researchers at the University of Alabama Birmingham to review the literature on dietary fat and obesity. The review concluded that under conditions of energy deficit, high-fat diets lead to greater weight loss than low-fat diets, but under *ad libitum* feeding conditions, instructing persons to follow a low-fat diet promotes loss of body weight and body fat.

Health Promotion or Disease Prevention

1. **Dr. Kristine Erlandson** has a collaboration with Dr. Wendy Kohrt, with the Department of Infectious Diseases (with co-mentor Dr. Thomas Campbell, Chair of Infectious Diseases as well as Dr. Liz Connick and Dr. Cara Wilson), and with the Department of Biostatistics (Dr. Sam MaWhinney). Age-related alterations in body composition (loss of lean tissue, gain in adipose tissue) are associated with increased inflammatory cytokines and are strong predictors of declining physical function and frailty in non-HIV-infected individuals. In HIV-infected individuals, anti-retroviral therapy-related lipodystrophy syndrome mimics classic age-related changes in body composition, including elevated inflammatory cytokines. No prior studies have looked at the role of body composition in HIV-related frailty. In HIV-negative elderly, inadequate vitamin D is strongly associated with prevalent and incident frailty. Interestingly, in HIV-infected patients, there is a high prevalence of vitamin D deficiency, which is associated with accelerated osteoporosis and increased rates of fragility fractures. No prior studies have looked at the relationship between osteoporosis and the development of frailty in HIV-infected individuals. We hypothesize that central obesity, loss of peripheral fat mass, and low bone mineral density (as determined by DXA scan) will be associated with the development of frailty in HIV-infected individuals.
2. **Dr. Benjamin Miller** and his team are interested in the prevention of frailty in older individuals through the use of exercise and nutrition interventions. To explore whether nitrogen retention can differ on an isonitrogenous diet by changing when protein is consumed, we performed a short-term study in older individuals (64.5 ± 2.0 years) performing daily exercise while in energy balance. Energy balance was important since by itself it can change nitrogen balance. Subjects consumed an isonitrogenous-isocaloric diet with the timing of a protein or carbohydrate beverage after exercise (protein after exercise

[PRO], carbohydrate after exercise [CHO]) versus earlier in the day. Three-day mean energy balance (PRO: 202 ± 36 kcal; CHO: 191 ± 44 kcal; $p = 0.68$) did not differ between trials, but 3-day mean nutrient balance (NBAL) was significantly more positive in the PRO (1.2 ± 0.32 g N) trial than the CHO trial (0.8 ± 0.45 g N) ($p < 0.05$). Older individuals were better able to maintain nitrogen balance by simply changing when a portion of an identical amount of daily protein was consumed. This outcome stresses that timing of protein ingestion may be just as important as how much protein is ingested. It also illustrates that aerobic exercise at an intensity that approximates walking is sufficient to change anabolic stimuli in the whole body. By taking advantage of this phenomenon, one may be able to slow age-related muscle loss and frailty.

3. **Dr. Kenneth Wright** and colleagues (**Drs. Melanson and Perrault**) found the following in regard to sleep and energy balance.
 - a. Weeks of circadian misalignment, such as that which occurs in circadian sleep disorders, reduces leptin levels and therefore may have implications for appetite and energy balance.
 - b. Sleep deprivation increases energy expenditure and recovery sleep reduces energy expenditure.
 - c. There are small differences in energy expenditure due to sleep stage, whereas awakenings from sleep increase energy expenditure.
 - d. The internal circadian clock modulates the pattern of energy expenditure across the 24-hour day such that energy expenditure is lowest near the beginning of the biological night and highest near habitual wake time.

Publications Related to Accomplishments

The following publications are representative of nearly 100 peer-reviewed journal articles written by NORC researchers from May 2009 to April 2010.

1. Cornier MA, Salzberg AK, Endly DC, Bessesen DH, Rojas DC, and Tregellas JR. The effects of overfeeding on the neuronal response to visual food cues in thin and reduced-obese individuals. *PLoS ONE*. 4:e6310, 2009. PMCID: PMC2712682.
2. Kosmiski LA, Ringham BM, Grunwald GK, and Bessesen DH. Using DXA modeling to explain the increased resting energy expenditure associated with the HIV lipodystrophy syndrome. *Am J Clin Nutr*. 2009, Dec;90(6):1525-31. PMCID: PMC2777466.
3. Stroebele N, de Castro J, Stuht J, Catenacci V, Wyatt HR, and Hill, JO. A small-changes approach reduces energy intake in free-living humans. *Journal of the American College of Nutrition*. 2009. 28: 63-68. NIHMSID: NIHMS210999.
4. Hill JO, Wyatt HR, and Peters JC. Using the energy gap to address obesity: a commentary. *Journal of the American Dietetic Association*. 109:1848-1854, 2009. PMCID: PMC2796109.

5. Shikany JM, Vaughan LK, Baskin ML, Cope MB, Hill JO, and Allison DB. Is dietary fat ‘fattening’? A comprehensive research synthesis. *Critical Reviews in Food Science & Nutrition*, in press.
6. Bergman, BC, Perreault, L, Playdon, MC, Samek, A, and Eckel RH. Increased intramuscular lipid synthesis and decreased diacylglycerol saturation in endurance trained athletes. *Journal of Applied Physiology*. 2010, May. 108(5):1134-41. PMID: PMC Journal – in process.
7. Perreault, L, Bergman, BC, Hunerdosse D, and Eckel Robert. Alterations in intramuscular lipid metabolism relate to diminished insulin action in men, but not women, in the progression to diabetes. *Obesity*. 2010, Apr. PMID: PMC Journal – in process.
8. Cornier MA. The effects of overfeeding and propensity to weight gain on the neuronal responses to visual food cues. *Physiol Behav*. 97:525-530, 2009. PMID: PMC2694218.
9. Lee SM, Lee AL, Winters TJ, Tam E, Jaleel M, Stenvinkel P, and Johnson RJ. Low serum uric acid level is a risk factor for death in incident hemodialysis patients. *Am J Nephrol*. 2009;29(2):79-85. PMID: PMC2786018.
10. MacLean PS, Higgins JA, Wyatt HR, Melanson EL, Johnson GC, Jackman MR, Giles ED, Brown IE, and Hill JO. Regular exercise attenuates the metabolic drive to regain weight after long-term weight loss. *Am J Physiol Regul Integr Comp Physiol*. 2009;297: R793-802. PMID: PMC2739786.
11. Melanson EL, Gozansky WS, Barry DS, Maclean PS, Grunwald GK, and Hill JO. When energy balance is maintained, exercise does not induce negative fat balance in lean sedentary, obese sedentary, or lean endurance-trained individuals. *J Appl Physiol*. 2009;107(6): 1847-1856. PMID: PMC Journal – in process.
12. Stroebele N, Ogden LG, and Hill JO. Do calorie-controlled portion sizes of snacks reduce energy intake? *Appetite*. 2009;Jun;52(3):793-6. PMID: PMC2694140.
13. Thorn SR, Regnault TR, Brown LD, Rozance PJ, Keng J, Roper M, Wilkening RB, Hay WW Jr., and Friedman JE. Intrauterine growth restriction increases fetal hepatic gluconeogenic capacity and reduces messenger ribonucleic acid translation initiation and nutrient sensing in fetal liver and skeletal muscle. *Endocrinology*. 2009;Jul;150(7):3021-30. PMID: PMC2703533.
14. Thorn SR, Regnault TR, Brown LD, Rozance PJ, Keng J, Roper M, Wilkening RH, Hay WW, and Friedman JE. Intrauterine growth restriction increases fetal hepatic gluconeogenic capacity and reduces messenger ribonucleic acid translation initiation and nutrient sensing in fetal liver and skeletal muscle. *Endocrinology*. 2009;150:3021-30. PMID: PMC2703533.
15. Cornier MA, Salzberg AK, Endly DC, Bessesen DH, and Tregellas JR. The effects of gender on the behavioral and neuronal responses to food. *Physiol Behav*. 2010;99:538-543. PMID: PMC2826550.

16. MacLean PS, Giles ED, Johnson GC, McDaniel SM, Fleming-Elder BK, Gilman KA, Andrianakos AG, Jackman MR, Shroyer KR, and Schedin PJ. A surprising link between the energetics of ovariectomy-induced weight gain and mammary tumor progression in obese rats. *Obesity (Silver Spring)*. 2010;18: 696-703. PMID: 19798068.

Fostering Collaborations

One of the ways the UCD NORC has had its greatest impact is in facilitating collaborations among the research base. Below we provide examples of new and planned collaborations that were facilitated by the NORC.

Newly Started Collaborative Studies

1. **The impact of an environmental and curriculum change on children's obesity rates (Dr. Lois Brink, PI; Drs. Ray Browning and James Hill) (R01HD057229)**

This study employs a 2 (environmental intervention vs. no environmental intervention) x 2 (curriculum intervention vs. no curriculum intervention) factorial design at 24 inner-city ethnically diverse elementary schools with repeated measures (pre-program, mid-program, immediate post-program, and 1 year post-program) and random assignment to the curriculum intervention. Prior to randomization, 12 Learning Landscape elementary schools will be matched to 12 non-Learning Landscape elementary schools according to the percentage of students receiving free or reduced lunch, student's race and ethnicity, and school size. Six of the matched pairs will be randomly assigned to the curriculum intervention. The study aims will be evaluated with objective physical activity measures using SOPLAY (System for Observing Play and Leisure Activity in Youth) measured at baseline (year 1) and years 2, 3, and 4. The following objective and subjective assessments will also be collected: (1) A random subsample of study participants will be selected to wear accelerometers and provide weight and height measurements at baseline and years 2, 3, and 4 (n=500). (2) A random subsample of grade 4 and grade 5 students will be assessed on mediating variables and self-reported leisure time physical activity and nutrition behaviors (n=580). (3) Playground environmental characteristics (e.g., spatial proximity of activity areas and size, number, type, and quality of activity areas) will be measured at the 24 schools through GIS mapping and direct observation characteristic coding. (4) During year 5, the curriculum intervention will be disseminated to the 12 control schools and we will compare the dissemination effects to the results in the initial trial. Funded by NIH grant HD057229. This study will use Balance First, a program developed by NORC investigators, and will use the Clinical and Energy Balance Core Laboratories.

2. **Inflammatory suppression of endothelial function with aging and obesity: effects of habitual exercise (Drs. Doug Seals and Robert Schwartz)**

This project seeks to determine the role of chronic low-grade systemic and local vascular inflammation in the development of arterial endothelial dysfunction with aging and increased body fatness (overweight and obesity). Dr. Schwartz, Chief of Geriatric Medicine and a member of the NORC faculty, serves as co-investigator on the project. Overweight and obese middle-aged and older subjects are treated for 4 days with placebo or salsalate, an aspirin-like anti-inflammatory agent used in arthritis and other inflammatory clinical conditions. Arterial function and risk factors for cardiovascular disease are determined in both treatment

conditions. A second component of the study seeks to determine if a habitual aerobic exercise intervention will reverse arterial dysfunction in this group and if such benefits are mediated by reduced inflammation. Supported in part by NIH AG031141. The project uses the Energy Balance Core Laboratory.

3. Influence of energy restriction-based weight and fat loss arterial endothelial function in middle-aged and older overweight and obese adults (Drs. Doug Seals and James Hill)

This study seeks to determine the efficacy of energy intake restriction-associated weight loss for improving arterial endothelial dysfunction in middle-aged and older overweight or obese adults. Dr. Hill, Director of the Center for Human Nutrition and PI of the NORC, serves as a consultant on the project. Overweight and obese middle-aged and older adults will undergo energy intake restriction-based body weight and fat loss for 12 weeks followed by a 4-week weight-stable period. Arterial function and risk factors for cardiovascular disease are determined before and after weight loss. The physiological mechanisms by which weight loss improves arterial endothelial function also will be assessed. Supported in part by NIH AG006537. The project uses the Energy Balance Core Laboratory.

Pending Collaborative Studies

1. Reducing breast cancer recurrence with weight loss: a vanguard trial (This is a multicenter study with Dr. Cheryl Rock, from University of California San Diego, serving as overall PI and Tim Byers serving as PI from the University of Colorado; also Drs. Hill and Sedjo) (R01 CA 148791, pending)

The high incidence of breast cancer coupled with earlier diagnosis and more effective therapies has resulted in more than 2.5 million American women who now are breast cancer survivors. Excess adiposity is a key risk factor for postmenopausal breast cancer incidence, and it has been shown to be a major risk factor for breast cancer recurrence. More than 40 observational studies have examined the relationship between excess adiposity and breast cancer recurrence; recurrence rates are significantly higher among overweight women and are 78% higher among obese women (body mass index [BMI] at least 30 kg/m²) than those who are normal weight (BMI 18.5 to 24.9 kg/m²). Adiposity, therefore, is a major adverse prognostic factor after breast cancer and may be a particularly important problem among women of various minority groups. Despite this importance, there has never been a clinical trial to determine whether weight control (weight loss and maintenance of that loss) reduces recurrence risk or improves survival after breast cancer diagnosis and treatment. The biological mechanisms that tie excess adiposity, and positive energy balance via increased energy intake relative to energy expenditure, to increased risk of breast cancer recurrence are not fully understood. Endogenous circulating estrogen levels are 50% to 100% higher in postmenopausal obese women than in women who are not overweight, and high circulating estrogen levels are a documented risk factor for breast cancer recurrence. Increased adiposity also is accompanied by alterations in cytokines, hyperinsulinemia, and reduced levels of binding proteins, which may act in concert to promote the growth of breast cancer cells. In the general population, weight loss has been shown to favorably affect levels of insulin and the growth factor and sex steroid hormone axis.

We propose a 4-year trial of 800 overweight or obese (BMI greater than 27 and less than or equal to 40 kg/m²) women 21 years old and older who have been diagnosed with stage IC, II, or IIIA breast cancer. The study is designed to demonstrate the feasibility of achieving sustained weight loss in this target population. We also will examine the impact of weight loss on quality of life outcomes and include an analysis of cost effectiveness and dissemination potential. Within this trial, we will also enable future exploration of the biochemical mechanisms linking obesity to lower risk for disease-free survival and genetic modifiers of response to weight loss and increase in physical activity with concomitant reduced energy intake. Importantly, this trial is strategically designed as a vanguard component of a fully powered trial of 2,500 women who will be evaluated for breast cancer recurrence endpoints. This vanguard strategy allows the opportunity to further tailor and streamline the process and intervention for expansion into the larger trial, while at the same time accomplishing important scientific aims.

The primary specific aim of the project is to conduct a 4-year vanguard randomized controlled trial with the primary endpoint of sustained weight loss in 800 breast cancer survivors, following all subjects for 2 years post-randomization. Our primary hypothesis is that weight loss of at least 7% can be achieved and maintained in this target population.

**2. Preventing childhood obesity: 5 Alive
(Dr. Hill, PI; with Drs. Wyatt, Daniels, Kempe, Catenacci, Suh, Ogden, and Stroebele)
(U01 HL103601, pending initial review)**

This U01 application illustrates the collaborative nature of our efforts to prevent childhood obesity. We propose to develop and evaluate an innovative community intervention (“5 Alive”) aimed at preventing excessive weight gain in fifth-grade children. Using a quasi-experimental design, we will implement the intervention in Denver and Aurora, Colorado, and use Colorado Springs, Colorado, as the control community. Our intent is to make the fifth-grade year the time that children become “masters” of healthy eating and active living. This will be achieved by providing resources to fifth-grade classes to allow students to gain knowledge and skills about healthy eating and active living and offering fifth graders opportunities to learn by making healthy choices as consumers in their community. The school focus will be on healthy eating, active living, and energy balance—not on obesity. Our community partners will make healthier diet and physical activity choices more attractive by 1) reducing their cost, 2) increasing their accessibility, 3) increasing their convenience, and 4) increasing their social desirability. General efforts will be made to increase the social desirability of the program. We propose to establish fifth grade as the time when our children develop skills for lifelong weight management.

Our primary outcome will be the percent of children in the overweight or obese categories (based on BMI for age z scores). Secondary outcomes will be physical fitness, academic performance, and attendance. All of the school systems that we will be working with already collect this information on all students. We will intervene in approximately 25,000 fifth graders during 3 years and follow these children for 2 to 4 years. We hypothesize that fewer participants in 5 Alive will be overweight or obese at the 2, 3, and 4 year follow-ups.

The 5 Alive intervention is aimed at multiple behavior settings in the community; involves targeting the child, family, and community; and involves impacting behavior, the physical

environment, and the social environment. We have already engaged a large number of public and private community organizations as partners in 5 Alive and anticipate that many more will join. During years 1 and 2 we will finalize the intervention and conduct a pilot project. The full intervention will be implemented in years 3 through 6. The project is possible because of the strong existing foundation in Metropolitan Denver for addressing childhood obesity. If successful, 5 Alive could be sustainable in Denver and Aurora and translatable to other communities.

Planned Collaborative Studies

1. Dietary fructose, diabetes, and cardiovascular disease

(Drs. Johnson, Austin, Wyatt, Hill, Catenacci, and MacLean)

Dr. Rick Johnson has developed an intriguing line of research linking fructose intake to the production of uric acid and the development and progression of metabolic disease. These clinically relevant observations have spawned a number of collaborative efforts within the Center to pursue the mechanisms behind this aspect of metabolic dysregulation. In the next cycle of the NORC, they will be examining the role of adenosine tri-phosphate (ATP) depletion from fructose in the proinflammatory response, the mechanisms of urate uptake, regulation of fructokinase, and the mitochondrial effects of uric acid. They are also studying the role of nicotinamide adenine dinucleotide phosphate oxidase (NADPH oxidase) in response to fructose and uric acid, mechanisms of endothelial dysfunction, and the development of leptin and insulin resistance. Clinical studies include trials to lower fructose intake or to pharmacologically reduce uric acid as a means for preventing or treating features of the metabolic syndrome.

The following papers (research conducted before Dr. Johnson moved to Colorado) describe the development of these hypotheses:

- Feig DI, Kang DH, and **Johnson RJ**. Uric acid and cardiovascular risk. *N Engl J Med*. 2008; 359:1811-21. PMID: PMC2684330.
- **Johnson RJ**, Perez-Pozo SE, Sautin YY, Manitius J, Sanchez-Lozada LG, Feig DI, Shafiu M, Segal M, Glasscock RJ, Shimada M, Roncal C, and Nakagawa T. Hypothesis: could excessive fructose intake and uric acid cause type 2 diabetes? *Endocr Rev*. 2009; 30: 96-116. PMID: PMC2647706.
- Feig DI, Soletsky B, and **Johnson RJ**. Effect of allopurinol on the blood pressure of adolescents with newly diagnosed essential hypertension: a randomized trial. *JAMA* 2008;300(8):924-32. PMID: PMC2684336.

2. Understanding the effects of bariatric surgery on type 2 diabetes and cardiovascular risk factors

(Drs. Draznin, Schoen, Cornier, Wyatt, Melanson, Bessesen, and Polsky)

In the last decade, numerous observations have clearly demonstrated that certain bariatric operations are curative for patients with T2DM. Substantial improvement in glucose metabolism occurs rapidly after operations involving a malabsorptive component, before any weight loss is recorded. Furthermore, the most recent findings of improvement and resolution of T2DM in patients with BMI less than 30, albeit in very few patients, have opened additional avenues for investigation.

The challenge in this field is to answer the following questions:

- a. What are the safest and the most efficacious bariatric procedures in patients with T2DM and BMI of less than 35 as compared to intensive medical therapy?
- b. What is the best study design to evaluate safety and efficacy of bariatric operations in patients with T2DM and BMI of less than 35 that can be used in a large multicenter randomized clinical trial? Because it is imperative to understand the mechanism(s) whereby bariatric operations improve and/or resolve T2DM, we propose to incorporate mechanistic aims into this feasibility study that can be employed in a future large multicenter trial. Future trials must be able to examine and differentiate the roles of caloric restriction (negative energy balance), weight loss, and the impact of altering gastrointestinal anatomy (with resulting changes in enteroinsular physiology) in the mechanism of resolution of T2DM after bariatric operations. We will assess the ease of recruitment, patient compliance, logistics of scheduling and medication washout, rates of dropout, and patients' and providers' satisfaction with the study design and implementation.
- c. We propose to randomize patients with T2DM and BMI between 30 and 35 into three groups: a) intensive medical therapy, b) laparoscopic Roux-en-Y bypass surgery (most commonly used mixed restrictive-malabsorptive operation), and c) laparoscopic gastric banding (a commonly used restrictive surgery). In brief, in a 6-month follow-up (the most one can accomplish in a short 2-year study), we will be able to estimate the initial safety and efficacy of these procedures in patients with T2DM and BMI less than 35. In addition to safety and efficacy, the proposed study design will allow us to estimate the impact of negative energy balance before the weight loss, acute post-operative weight loss, and chronic 6-month weight loss. All patients will be studied four times: a) at baseline, b) after 3 days of very low calorie diet (VLCD) to assess the impact of negative energy balance before any changes in weight might be observed, c) after the 15 pounds weight loss either post-operatively or in the course of continuous VLCD therapy (medically treated group) to evaluate the impact of the initial acute weight loss, and d) at 6 months of follow-up, near the peak of weight loss, to estimate the influence of chronic weight loss. At each time points we will perform a hyperinsulinemic-euglycemic clamp; hyperglycemic clamp; DXA scan for body composition; mixed meal test for GLP-1, GIP, and ghrelin; measurements of energy expenditure and substrate utilization; and blood tests for glucose, insulin, C-peptide, free fatty acids, lipid profile, adiponectin, TNF-alpha, RBP4, IL-6, hsCRP, and HbA1c.

Educational Activities/Accomplishments

These activities are supported and will continue to be supported through the Center for Human Nutrition, University of Colorado Denver School of Medicine.

1. Increasing Awareness of Nutrition Within the Universities

A major goal of the UCD NORC is to increase awareness of nutrition within Colorado's

academic institutions. This includes promoting nutrition research by presenting seminars, grand rounds, and lectures to groups of non-nutrition investigators within each university.

2. **Medical School Education**

In the previous cycle of funding, a Nutrition Academic Award (K07 DK002976 Krebs, PI; Bessesen, Co-Inv) enabled the establishment of a vertically integrated nutrition curriculum within the UCD School of Medicine. Four years ago, following a period of study, the School of Medicine began an entirely reorganized medical school curriculum that was built around system-based blocks. Dr. Bessesen was a part of the Curriculum Reform Committee and serves as co-director of the Digestive, Metabolic, and Endocrine Systems Block, which is given in the Fall of the second year of medical school. The Metabolism portion of this block includes 23 hours of lecture and small groups on topics in nutrition. These range from didactic sessions on dietary fat, vitamins, malnutrition, and obesity to discussions of international and hospital-based nutrition. Nutrition elements are also incorporated into the Introduction to Clinical Medicine (Foundations of Doctoring) course, which is a longitudinal part of the core curriculum given during the first 2 pre-clinical years. Laura Primak, R.D., is charged with incorporating nutritional topics into the Pediatrics and General Internal Medicine clerkships taken by third-year students. There are also two nutrition electives offered in the fourth year. These are 2 weeks in duration and have been taken by more than 25% of medical students. Nutrition core concepts have also been incorporated into residency training for both Pediatrics and General Internal Medicine. This consists of three weeklong elective blocks in the General Internal Medicine primary care track that emphasize nutritional aspects of management of T2DM, hyperlipidemia, and obesity. Nutrition topics are also incorporated into the longitudinal didactic curriculums of both the Pediatric and General Internal Medicine residency programs.

3. **Graduate School Education**

Graduate Training in Nutrition at Colorado State University (CSU)

The Department of Food Science and Human Nutrition at CSU provides Masters and Ph.D. degrees in nutrition. It is possible for graduate students at CSU to engage in research at UCD. We hope to stimulate increased CSU student participation within the NORC.

Graduate Training at UCD

The UCD has a Graduate Program in Clinical Science (CLSC). This program is intended for individuals with a health sciences graduate degree or a health care professional degree (e.g., physicians, Masters in Public Health graduates, biostatisticians, epidemiologists, nurses, pharmacists, physical therapists, and dentists) who are interested in obtaining either a Ph.D. or Masters degree in Clinical Science (Clinical Investigation, Health Information Technology, or Health Services Research). Several members of the NORC are mentors for students in this program.

Graduate Training in Exercise Science at CU and CSU

The University of Colorado Boulder offers Masters and Doctoral degrees in Integrative Physiology, which includes exercise science. The Department of Health and Exercise Science at CSU offers Master's and Doctoral degrees. Both departments have a heavy emphasis on nutritional aspects of physical activity. Students in both programs can work directly with members of the Colorado NORC.

4. **Seminar Series**

The UCD NORC has combined efforts with the Department of Medicine's Division of Endocrinology, the Department of Pediatrics, and the University of Colorado Cancer Center to host a unified Research Conference every Wednesday at 11 am and two additional, less formal, interest groups. Given the growing overlapping interest between obesity, nutrition, metabolism, and cancer researchers, this move was initiated to help promote their interaction and collaboration. The specific benefits to the NORC include:

- Improving our collaborative interaction with the Cancer Center, the Endocrine Division, and the Department of Pediatrics
- Improving interaction and collaborations with more external speakers
- Increasing the NORC's presence and status by increasing interaction with researchers not normally affiliated with the NORC
- Increasing the quality and impact of NORC seminars and interest groups
- Encouraging better attendance for interest groups, where trainees and junior faculty can present and lead discussions, informal brain storming sessions can occur, and new collaborative relationships can develop
- The resources to provide lunch for attendees of both Research Conference seminars and of the Metabolism Interest Group, which particularly raises attendance of graduate students and fellows
- The resources and facilities to begin videoconferencing Research Conferences to interested NORC members at Colorado State University

The Research Conference has two seminar series that fill the schedule and provide a modest lunch for attendees: Hormones and Related Malignancies and the Nutrition & Metabolism Seminar Series. Many times the seminar is listed for both seminar series, because the topic is relevant to both themes. Attendance ranges from 40 to 80 scientists and trainees.

The Western Dairy Council has been a long-time supporter of the NORC and its seminar series. Their support is used to bring in external speakers throughout the year. During the past year, we have obtained more funding from the University of Colorado Cancer Center's program in Hormones and Related Malignancies, which will increase the support for external speakers in the coming years. Throughout the year, the Division of Endocrinology and the Center for Human Nutrition facilitate application for educational grants from private industry, which also allows us to bring in external faculty.

NORC also sponsors the Metabolism Interest Group. This interest group is run by Drs. Irene Schauer, Rocio Pereira, and Kristin Nadeau, and provides a venue for trainee presentations, grant reviews, methodological seminars, and journal article reviews. It originally began as a bimonthly meeting but now is held weekly. The group meetings are attended by 20 to 30 people each week.

5. **Centers for Obesity Research and Education (CORE)**

The UCD NORC is part of the national coordinating center for the Centers for Obesity Research and Education (CORE). CORE consists of eight academic medical centers committed to providing hands-on training in weight management to health care professionals. CORE was established in 1999 to provide practical information about weight management

and obesity to primary care physicians and other health care professionals. Obesity experts from the following academic medical centers created CORE:

- Columbia-St. Luke's' Roosevelt Hospital (New York)
- Harvard University (Boston)
- Mayo Clinic (Rochester)
- Northwestern Medical School (Chicago)
- Pennington Biomedical Research Center (Baton Rouge)
- University of California, Los Angeles
- University of Colorado Denver (Denver)
- University of Minnesota (Minneapolis)

6. Providing Accurate Nutrition and Obesity Information to the Public

The public is hungry for information about nutrition and obesity but this information is often obtained from unreliable sources. It is our goal to ensure that accurate and timely nutritional information is provided to the population of the Rocky Mountain Region. Several programs are underway to accomplish this goal:

- **Local Media Appearances:** We have been very effective in positioning the Center for Human Nutrition as the first place local (and national) journalists go for nutrition and obesity information. Many members of our research base enjoy interacting with the media and we are able to refer journalists to the most appropriate person to contact for their specific topic. With increased media attention on obesity, Colorado investigators are increasingly active in providing information to journalists.
- **Public Lectures:** We provide a number of lectures about nutrition and obesity each year to the public. The UCD organizes several public lectures each year, and obesity is a very hot topic for the public. Drs. Hill, Wyatt, Eckel, and Bessesen have provided lectures in this series. Additionally, the Center for Human Nutrition provides at least one public lecture on obesity each year. This generally draws several hundred members of the public and is often attended by members of the media. It is our intent to provide more of these public lectures in the future.
- **“Talk to the Doc”:** Dr. Hill and Ms. Cathy Walsh of Channel 4 host a local physician each month to talk about an issue related to health. This is featured live on the channel 4 website.
- **New Health & Wellness Center:** A major mission of the new Health & Wellness Center will be to provide information about diet, physical activity, and weight management to health professionals and to the public. The facility will have space to bring these individuals in for lectures or conferences, and the technological capacity to provide information via the Internet or via satellite transmission.

7. Translating Science to Improve the Health of the Community

- **America On the Move:** Drs. James Hill, Holly Wyatt, and several other NORC members were influential in establishing America On the Move (AOM) in 2003 as a national

weight gain prevention initiative. AOM is based on research showing that the energy gap for prevention of excessive weight gain is small and that a small-changes approach to changing behavior has the potential to prevent excessive weight gain in individuals and in the population. AOM aims to translate the small-changes research into programs that can help reduce obesity in the population. Since its inception, America On the Move has reached more than 4 million people nationwide through its website, through school programs, and through media messages. AOM is used in communities throughout Colorado and around the country as a community approach to addressing obesity. Two communities in metropolitan Denver – Broomfield and Aurora – received funding during the past 5 years from Livewell Colorado to implement America On the Move in their communities. The program is based on research conducted within the Colorado NORC, and research in this area continues (Dr. Hill, NIH and USDA funding) within the Colorado NORC. Dissemination of AOM has been funded by the Colorado Department of Public Health and Environment and by Livewell Colorado, a nonprofit devoted to reducing obesity in Colorado. With funding from the USDA, Dr. Hill and colleagues are evaluating the feasibility of using the AOM family program with USDA extension agents. If successful, this would be an effective way of providing weight-gain prevention information to communities.

- Colorado Weigh: Drs. Holly Wyatt and James Hill have maintained a behavioral weight-loss program, Colorado Weigh. This program is available to anyone within the community for a small fee. The program translates research from the National Weight Control Registry, the Diabetes Prevention Program (DPP), and the Look AHEAD study. Data are collected on all participants, which is now more than 1,500 individuals. The success of Colorado Weigh has led to interest from many other NORC investigators in having access to participants before and after weight loss to investigate various research questions. Thus Colorado Weigh will be used in our Clinical Core as a means of producing weight loss. Colorado Weigh will also be a part of the new University of Colorado Anschutz Health and Wellness Center. We offer Colorado Weigh at the UCD and also throughout the community.
- Fit 4 Colorado: Fit 4 Colorado is a collaboration between America On the Move and CBS Channel 4 Television. The program promotes small changes in diet and physical activity through news stories, public service activities, and recognizing schools for making innovative changes in diet and/or physical activity. One school is chosen each month and receives a \$1,000 prize. Dr. Hill and a Channel 4 on-air personality visit the school and film the children in an assembly. The film is shown during the weather segment of the local evening news.
- Balance First: Balance First is an energy balance curriculum developed as part of America On the Move by NORC researchers to teach elementary school children about energy balance. The curriculum aims to teach the integration of energy intake and energy expenditure, and is aimed to empower children by teaching them energy balance skills. Balance First has reached more than 2.5 million elementary school children. We are revising and expanding Balance First through support from a new grant, with Dr. Lois Brink as Denver PI.

- **Learning Landscapes:** Dr. Lois Brink, a new member of the NORC research base, has established the Learning Landscapes program. Learning Landscapes is a forward-thinking program at the UCD that connects the design and construction of urban public spaces with healthy initiatives. Since 1998, in partnership with the Denver Public Schools, Learning Landscapes has transformed 48 neglected public elementary schoolyards into attractive and safe multi-use parks tailored to the needs and desires of their neighbors and communities.
- **Improving School Nutrition:** Dr. Nanette Stroebele has been working with schools and children to alter the food and physical activity environments in order to improve nutrition and reduce obesity. In particular, Dr. Stroebele worked with the Denver Public Schools to translate the small-changes approach in modifying school lunches. She identified three popular foods that were also high in fat and energy density – pizza, chicken nuggets, and macaroni and cheese. Working with the food manufacturers, she was able to develop versions of these foods with lower energy density and lower fat content. She demonstrated that the modified versions of these products were very acceptable to the students. Dr. Stroebele is currently working with three school districts in metropolitan Denver to help improve the food and physical activity environments.
- **Metro Denver Health and Wellness Commission (MDHWC):** Dr. James Hill was one of the founders of the Metro Denver Health and Wellness Commission, established with the goal of making metropolitan Denver the healthiest community in the country. The MDHWC consists of key community leaders from government, academia, and the private sector. Barbara O'Brien, the Lieutenant Governor of Colorado, serves as the MDHWC chair and Dr. Hill serves as chair of the MDHWC Executive Committee. The MDHWC has developed a strategic plan and a set of metrics to measure success (<http://www.mdhwc.org>).
- **Livewell Colorado:** Livewell Colorado is a new nonprofit organization aimed at reducing obesity in Colorado by creating a convergence of organizations and individuals addressing obesity. Dr. James Hill was a founding member of the Livewell Colorado Board of Directors and has established a close working relationship with Livewell Colorado and the University of Colorado NORC. With funding from the Kaiser Permanente Foundation and the Colorado Health Foundation, Livewell Colorado provides strategic direction and funding for efforts to reduce obesity in Colorado. Livewell currently funds more than 25 Colorado communities in efforts to address obesity.
- **Anschutz Health & Wellness Center (HWC):** The new Health & Wellness Center will greatly facilitate the translation of research into clinical practice and into the community. The HWC will offer lifestyle programs (e.g., weight management, increasing physical activity) for referred patients and for the public. Additionally, the HWC will promote and support translation of research into community programs. This will be accomplished by facilitating community research and by working in collaboration with groups such as Livewell Colorado and the Metro Denver Health and Wellness Commission.

Benefits and Interactions Resulting From the Existence of the NORC

First, we have successfully identified funding for a new UCD program in Health & Wellness. This was made possible with an initial gift of \$15 million from the Anschutz Foundation and was matched by approximately \$21 million from UCD. A new 100,000-square-foot facility is under construction to house the Health & Wellness Center. This facility will have state-of-the-art space for clinical research in diet, physical activity, and obesity. It will house our Clinical Core Laboratory and the clinical trials program in obesity as well as new clinical programs in weight management, lifestyle prevention of chronic disease, and programs for improving diet and physical activity.

The location of the NORC Clinical Core Laboratory within the HWC will allow it to serve as a conduit for clinical researchers who are recruiting human subjects for intervention studies. Additionally, we hope to develop a “patient registry” and a repository of phenotype and genotype information as well as tissue samples from patients and study subjects coming to the HWC. The HWC will also house community programs aimed at improving lifestyle. The administrative offices of the NORC will be located in the new HWC building. Additionally, several NORC clinical researchers will be housed in the building. The HWC will substantially strengthen our ability to provide education and training in nutrition and obesity to health care professionals, students (medical and graduate), and the public. The HWC will have facilities for workshops and classes (including cooking and exercise classes) for health professionals and for the public.

Second, the success of our research program enabled us to propose a new research initiative in obesity within the UCD School of Medicine. In Summer 2009 the School of Medicine (SOM) issued an announcement that they were looking for research programs of excellence to invest in for the future. We submitted an application to establish a research program in obesity. We were chosen as one of six programs to compete for substantial SOM funding. We spent 4 months developing our proposal and it was recently presented at the SOM Research Retreat. We have received notification that our proposal will be funded, although the exact amount will be negotiated with the Dean of the School of Medicine.

Third, we have experienced explosive growth (54%) in our research base during the last 5 years and many of the new researchers are interested in obesity. We decided it was appropriate to engage the NORC research base in developing the obesity initiative proposal and to use the process to reevaluate the organizational structure of the NORC. Through this process it was decided that:

1. The NORC research base was too broad for the focus on Nutrient Utilization and Function, and in fact we felt that our major theme was promotion of translational research.
2. We reorganized our research base around six focus areas: regulation of food intake, weight management, energy metabolism in health & disease, obesity and metabolic dysregulation, maternal-fetal origins of chronic disease, and obesity cell biology.
3. We identified a strong need for a Clinical Core Laboratory. In order to achieve this we have discontinued the mass spectrometry core laboratory and created a new clinical core. The

clinical core will provide assistance with investigators conducting clinical research, especially those doing obesity intervention studies.

Examples of Basic Science – Clinical Investigations That Have Occurred

As the Colorado NORC grows and our resources stay the same, we face a challenge in how best to support our research base. We have found the use of scientific cores is one way to support the research base, but that bringing people together to facilitate interaction is another way to support our research base. As we look for additional sources of support for our research base we believe there are potential opportunities from creating collaborative, multidisciplinary teams within our research base. A strength of our research base is that we have a large number of investigators studying nutrition and obesity at different levels – epidemiology, basic science, clinical science, and community/population research – who actually interact. We believe that bringing these teams together on a regular basis can lead to higher quality, collaborative research studies and can generate program project applications and/or responses to multidisciplinary requests for applications. We believe these teams will also facilitate translation of the science to the public. We therefore chose “A Translational Approach to Nutrition and Obesity” as the new focus of our NORC. We have identified four areas in which a research question is already being addressed at multiple levels and a translational team is in place:

1. Weight-Loss Maintenance
2. Maternal-Fetal Programming of Obesity
3. Menopause, Obesity, and Disease Risk
4. Obesity and the Mammary Gland

Within each of these translational teams, researchers working in basic research are collaborating with investigators working in clinical research and in translational research.

Translational Team 1: Weight-Loss Maintenance

Colorado NORC investigators have been involved in weight-loss maintenance research for many years. **Drs. Hill and Wyatt** have studied individuals in the National Weight Control Registry (NWCR), a registry of successful reduced-obese individuals, for many years. Based on this research, they proposed that weight loss and weight-loss maintenance are different processes requiring different strategies. Their research suggested that very high levels of physical activity were essential for weight-loss maintenance. Drs. Hill and Wyatt coined the term “energy gap” to quantify the amount of behavior change required to prevent weight gain and to maintain weight loss. **Dr. Vicki Catenacci** obtained a K23 grant to continue studying physical activity patterns in the National Weight Control Registry. Her grant involved measurement of energy expenditure and substrate utilization in a whole-room calorimeter, in collaboration with **Dr. Ed Melanson**. Based on work with the NWCR, Dr. Wyatt was successful in obtaining an R01 grant to study exercise in weight-loss maintenance. Simultaneously, **Dr. Paul MacLean** has been studying the weight-reduced state using a rodent model of dietary obesity. He has identified several physiological and behavior factors that occur during the weight-reduced state that appear to make the animals susceptible to weight regain. This includes factors in the brain and periphery.

Drs. Wyatt and MacLean began collaborating and investigating whether the factors present in the reduced obese rodent were present in the reduced obese human.

Because Dr. Wyatt was producing and studying weight reduced subjects, other investigators became interested in studying these subjects before and after weight loss. These included **Dr. Marc Cornier**, who was interested in using fMRI to study the brain response to food intake before and after weight loss. **Dr. Dan Bessesen** was interested in studying dietary fat trafficking before and after weight loss. **Dr. Rick Johnson** is interested in examining the relationship between uric acid and metabolic rate in the reduced obese state.

Dr. James Hill, the PI for the Denver Look AHEAD site, is very interested in strategies to increase weight-loss maintenance. The Look AHEAD study aims to maintain weight loss over many years and has begun to use a series of challenges and contests among participants to stimulate continued adherence to diet and physical activity goals.

Recently, **Drs. Boris Draznin** and **Jonathan Schoen** developed a project to study the metabolic effects of bariatric surgery. Understanding how bariatric surgery helps with weight-loss maintenance may eventually help us improve our lifestyle strategies for weight-loss maintenance.

Finally, Drs. Hill and Wyatt offer a behavioral weight-loss program—Colorado Weigh—to the public. They continue to study the effectiveness of this community-based weight management program on long-term weight loss. Additionally, they have studied the impact of this program in other communities.

We believe that providing an opportunity for these investigators to meet regularly and interact will stimulate higher quality, multidisciplinary research. We also believe this area is ripe for generation of program projects.

Special Populations: National Weight Control Registry; Colorado Weigh participants
Cores to be Used: Clinical, Metabolic, Energy Balance

Translational Team 2: Obesity and the Mammary Gland

At our institution we have an excellent group of researchers who study mammary gland biology, with interests in mammary gland development, lactation, and disease development. During the past grant cycle, a translational unit emerged from this research base – with the extensive support of NORC Clinical, Energy Balance, and Metabolic cores – with an interest in the impact of obesity on mammary gland biology. While this unit has only recently emerged within our research base, we expect it to grow substantially in the next cycle of the NORC because of the number of grant applications currently in review or in preparation.

Several of these researchers are part of a program project grant focused on mammary gland development (Drs. Anderson, **Neville**, McManaman, MacLean, and McCurdy). Studies have been initiated and proposed in their recent renewal application that are focused on the impact of hyperinsulinemia and peripheral insulin resistance on the changes in epithelial cell and adipocyte biology during pregnancy through the transition to lactation. These researchers are particularly focused on the signaling pathways that coordinate the metabolic and morphological changes as

the mammary gland prepares for lactation. They have joined forces with **Drs. Paul MacLean and Carrie McCurdy** in several pending grants to perform hyperinsulinemic-euglycemic clamps in pregnant insulin-resistant and insulin-sensitive transgenic mice.

Drs. Jim McManaman, Paul MacLean, and Matt Jackman are currently using the Energy Balance and Metabolic Cores to perform 24-hour dual tracer experiments in pregnant dams (mice), assessing energy balance, fuel utilization, and the trafficking of ingested fuel. Their ongoing studies with these NORC-supported tools have extended into lactation, to obtain a comprehensive examination of the impact of obesity on energy balance and nutrient metabolism in lactating dams and their nursing litters. Their graduate student, **Jessica Wahlig** (Drs. MacLean and McManaman, co-mentors), acquired a Colorado Clinical and Translational Sciences Institute (CCTSI) predoctoral fellowship related to this obesity/lactation project, and is also involved in the concurrent clinical pursuits of this group on lactation impairments in obese women. Jessica is working with **Dr. Maya Bunik**, a physician who deals with lactation impairments in the clinic. Dr. Jim McManaman, Dr. Maya Bunik, and Dr. Jessica Wahlig plan to study lactation defects in obese women.

The study of the impact of obesity on normal mammary gland function has provided an exceptional foundation for studying the impact of obesity on disease in this tissue. **Dr. Pepper Schedin** has a long-standing interest in the how mammary gland development affects breast cancer. During the last cycle of this award, she and **Dr. Paul MacLean** used seed money from the American Cancer Society to develop a model of postmenopausal obesity. Observations from the model have generated a significant amount of collaborative pursuits within this translational unit. Additional funding from the Komen Foundation has been obtained (Drs. Anderson, MacLean, Schedin, and Giles) to examine the therapeutic impact of metformin on obesity-associated tumor promotion, and preliminary findings from these observations have generated enough interest that **Dr. Erin Giles**, postdoctoral fellow with Dr. MacLean, obtained a Thorkildenson Fellowship and has just been awarded another fellowship from the American Institute for Cancer Research to perform tracer studies in the model. **Dr. Steve Anderson** is working with **Dr. Ann Thor**, Head of Pathology, to translate their observations of metformin for breast cancer therapy into a clinical pursuit, examining the efficacy of metformin therapy on breast cancer risk. Related to this work, **Dr. Robert Eckel** and his postdoctoral fellow, **Dr. Isabel Schlaepfer**, have recently initiated a collaboration with breast cancer researchers (Drs. Carol Sartorius and Kate Hortwitz) to study the role of Munc18c as mediators of lipid homeostasis in obesity and cancer phenotypes. **Dr. Peggy Neville** and her colleagues are collaborating with Dr. MacLean to study the role of hyperinsulinemia and Her-2 status in obesity-related tumor promotion in preclinical models. They have grants pending with the Komen Foundation to support this effort.

Both preclinical and clinical studies have been initiated and submitted by members of our research base to study the effect of exercise and calorie restriction on mammary tumor promotion (Drs. MacLean, Schedin, Anderson, Higgins, Jackman, and Giles; pending) and breast cancer outcomes (Drs. Byers and Hill; pending), and Dr. Catherine Jankowski is studying the impact of breast cancer therapy on the ability of patients to exercise regularly (P20). Many of these researchers are involved in a consortium within the university; this consortium is working on an NCI-sponsored SPORE application that will have nutritional and exercise components. This consortium, led by Dr. Anthony Elias, is just beginning to make links with the NORC

researchers and Core facilities. Making the obesity-cancer connection at our institution is an emerging priority, and we expect that coordinated efforts of the NORC and the University of Colorado Cancer Center will continue to grow and expand into clinical, epidemiological, and community outreach programs.

All of these efforts have been supported extensively by the NORC Cores. Given the emerging interest in obesity on mammary gland development, function during lactation, and breast cancer, this translational unit is likely to contribute significantly to the continued growth and success of the Colorado NORC in coming years.

Cores to be Used:	Clinical, Energy Balance, Metabolic
Access to Special Populations:	Breast Cancer Survivors, individuals participating in a community behavior-based weight-loss program

Translational Team 3: Maternal-Fetal Programming of Obesity

Basic Science

The effect of maternal obesity on the fetus and early childhood obesity is an area of intense clinical and basic science interest at the University of Colorado. Pregnancy is a critical period when an exposure may have a lifelong effect on the structure or function of an organ, tissue, or body system through biological programming. **Dr. Friedman** and collaborators at the University of Oregon and University of Utah have developed a nonhuman primate model of maternal obesity. This collaboration led to several novel observations in the fetal liver, brain, and skeletal muscle that suggest that maternal lipids in the diet result in an early obesity phenotype in the developing fetus, particularly the fetal liver. Based on this work, **Dr. Carrie McCurdy** was successful in obtaining an NIH K12 award to study the fetal programming of insulin resistance in skeletal muscle from obese Japanese macaques. Simultaneously, Dr. Friedman developed a novel transgenic mouse model that genetically re-balances the omega-3 to omega-6 ratio on a high-fat diet, and prevents the pro-inflammatory response in diet-induced obesity. This appears to be protective of the fetal programming effects in the offspring despite maternal obesity. The effects of maternal under-nutrition on the fetus are being investigated by **Dr. Bill Hay** and colleagues using a sheep model of IUGR. The IUGR model is being used by **Dr. Laura Brown** (K12) to study skeletal muscle fetal programming, **Dr. Paul Rozance** (KO1) is studying the fetal pancreas, while **Dr. Stephanie Thorn** (NIH F32) is studying how IUGR affects hepatic insulin resistance in the fetal liver.

Clinical Investigation

To investigate the clinical implications of these observations, Dr. Friedman and Dr. Barbour obtained an NIH R01 grant, entitled Regulation of Maternal Fuel Supply and Neonatal Adiposity. This is a collaboration among **Dr. Linda Barbour, Endo, PI; Dr. Jed Friedman, Co-I; and colleagues Dr. Wendy Kohrt, Medicine; Dr. Patti Thureen, Pediatrics; Dr. Rachel Van Pelt, Medicine; Dr. Kristin Nadeau; and Dr. Nancy Krebs, Pediatrics**. These studies will address how pregnant lean, obese, and gestational diabetes mellitus (GDM) women handle glucose and lipids over 48 hours, including maternal adipose tissue biopsies and maternal inflammation in early and late gestation. This study also examines the relationship between

maternal metabolism and fetal adiposity using a specially designed body composition device (a Peapod), and analysis of neonatal insulin resistance at birth and at 1 year old. One exciting finding thus far is the strong linear relationship identified between the change in maternal triglycerides and fetal adiposity ($r=0.89$). **Dr. Friedman and Dr. Barbour also submitted an R-21** designed to study the role of lipids versus carbohydrate in the maternal GDM diet on fetal adiposity, inflammation, and cardiovascular risk. **Dr. Krebs, Dr. Barbour, and Dr. Friedman**, are mentoring **Dr. David Brumbaugh** who is pursuing pilot funding to evaluate the effects of maternal diabetes and obesity on the abdominal and liver fat content in newborn babies using magnetic resonance spectroscopy. Working with clinical investigators in the Department of Ob-Gyn and Women's Health at the UCD, **Dr. Kristen Boyle** is utilizing a novel incubated human muscle fiber system suitable for the study of muscle metabolism and signal transduction *in vitro*. We have demonstrated that obese women with GDM have a unique insulin receptor defect that may underlie future risk for the development of T2DM. We identified specific intracellular serine-threonine kinases as possible candidates for inhibitors of insulin signal transduction and a novel pathway for insulin resistance.

Epidemiology

Dr. Dana Dabelea in the School of Public Health is doing exciting epidemiological research exploring the Fetal Origins Hypothesis in Diverse Youth (EPOCH). She has an R01 focusing on over-nutrition (exposure to diabetes *in utero*) and under-nutrition (IUGR) and is examining the consequences on childhood obesity phenotypes. **Dr. Dabelea and Dr. Friedman** are also collaborating on a newly funded R01 epidemiological evaluation of more than 2,000 lean and obese women designed to explore the relationships between maternal body size and behaviors during pregnancy, intra-partum fuels, markers of inflammation and insulin resistance, and infant body size and fatness. This study is relying heavily on the Metabolic Core Laboratory and the Energy Balance Laboratory in Pediatrics. Capitalizing on these studies, **Dr. Nancy West** received support from the CCTSI to examine the environmental factors in early life, including DNA methylation, on the fetal origins of diseases across the lifespan.

Population/Community

To help translate these studies into the community, **Dr. Dabelea** was funded recently to conduct the National Children's Study (NCS) in Colorado, the largest and most comprehensive long-term study of children's health and development ever attempted in the United States. The Colorado NCS team is based on a multidisciplinary partnership between the UCD School of Public Health (SPH) and Departments of Pediatrics, and Obstetrics & Gynecology in the School of Medicine. The aim of the study is to recruit 250 pregnant women in Douglas County per year for 4 years. The women will be recruited from the community and monitored closely throughout pregnancy by a team of investigators. During pregnancy and after birth, information will be collected on many exposures and outcomes. The children may be followed for up to 21 years.

Dr. Jennifer Leiferman received a K01 to investigate the effects of antenatal physical activity intervention on maternal stress, biomarkers, and preterm delivery. These are collaborative interdisciplinary projects with clinical investigators who have expertise in epidemiology, ob-gyn, pediatrics, endocrinology, and neonatology.

Dr. Nancy Krebs is conducting metabolic studies comparing three different complementary feeding regimens in U.S. breastfed infants on body composition, and a pilot study examining the feasibility of an office-based electronic system to enhance providers' assessment of obesity and cardiovascular disease risk in children.

This cadre of investigators has been very successful thus far in capitalizing on their particular strengths in maternal-fetal research. By forming a working group focused on maternal-fetal programming of obesity, we will provide a framework within which researchers utilizing human physiology, animal modeling, epigenetics, and epidemiology will be able to find and develop collaborative approaches to understanding the basis for how obesity develops, beginning in fetal life and early infancy, and the effects on the population over the lifespan. We also hope to develop novel approaches that can prevent childhood obesity beginning in the womb, and to continue to train and mentor promising junior investigators in this field.

Access to Special Populations:	Pregnant women, neonates
Cores to be Used:	Clinical, Metabolic, Energy Balance

Translational Team 4: Translational Unit on Menopause, Obesity, and Disease Risk

Menopause-related changes in health are poorly understood, largely because the menopause is a not an event but rather a process that occurs over many years. This makes it challenging not only to isolate the biological changes that are the direct result of the menopause (i.e., sex hormone withdrawal) but also to study the mechanisms of change. It has long been known that the withdrawal of estrogen increases bone loss and risk for osteoporosis. There is now a solid base of evidence that the menopausal transition is also associated with an increase in fat accrual, a redistribution of fat to abdominal regions, and a loss of lean mass other than bone. Such changes likely increase risk for metabolic dysfunction, cardiovascular disease, and premature disability. This recently formed translational unit of basic, clinical, and community investigators is studying the mechanisms underlying these changes and intervention strategies to mitigate the unfavorable consequences of the menopause on health.

Basic Science

Dr. Erin Giles is working with Drs. Paul MacLean and Matt Jackman to characterize the impact of obesity on energy intake, energy expenditure, and fuel utilization throughout the normal estrous cycle and after surgical ovariectomy in rodents. They have observed that obese rats have a blunted response to the reduction in food intake on diestrous day 2 and proestrous, and a more substantial energy imbalance on their estrous day. In addition, obese rats preferentially used carbohydrate for energy production and had a blunted ovariectomy-induced positive energy imbalance. Early in this period of rapid weight gain, obese rats exhibit an increase in the relative frequency of small adipocytes (less than 20 μm), and *in vivo* tracers have been used to show that these small adipocytes preferentially take up dietary fat when compared to adipocyte of medium (greater than 40 μm ; less than 80 μm) or large (greater than 80 μm) size. This manuscript is currently being prepared for publication.

Clinical Science

A major area of focus of the IMAGE (Investigations in Metabolism, Aging, Gender, and Exercise) research group, directed by **Dr. Wendy Kohrt**, is on metabolic actions of estrogens. Dr. Kohrt currently has an R01 on Estrogen Deficiency and Mechanisms of Fat Accumulation that uses a pharmacologic model of menopause to study estrogen-related modulation of energy balance. Premenopausal women undergo 5 months of sex hormone suppression via GnRHag, with placebo or estradiol add-back therapy. The primary hypothesis is that estrogen deficiency results in a suppression of metabolic rate, thereby increasing the propensity for fat gain. The second hypothesis is that estrogen deficiency alters the regulation of hypothalamic-pituitary-adrenal (HPA) axis activity, such that there is an exaggerated cortisol response to stress in the estrogen-deficient state. A companion hypothesis is that estrogen alters peripheral metabolism of glucocorticoids in a manner that results in increased local production of cortisol in certain adipose tissue depots. This work is being conducted by **Dr. Wendee Gozansky** (K23, R21). The global hypothesis is that cortisol-related mechanisms contribute to the redistribution of fat toward abdominal regions after the menopause. Adipose tissue is obtained in these studies to identify tissue-level mechanisms of action; these are done in collaboration with **Drs. Ro Pereira, Paul MacLean, and Jed Friedman**. In a companion pilot study, **Dr. Kent Hansen** (NORC and CCTSI pilot awards), a postdoctoral fellow working with Dr. Kohrt, is investigating whether the expected increase in fat mass in response to GnRHag therapy occurs via adipocyte hypertrophy or hyperplasia, and whether this can be prevented by either estrogen add-back therapy or exercise. Hyperplasia is being assessed by cell sizing (i.e., shift in size distribution toward more small cells), by the incorporation of deuterium into nucleosides of new adipocytes (daily dosing with deuterium during the first 50 days of GnRHag therapy), and by the evaluation of markers of inflammation, macrophage infiltration, and early adipocyte differentiation. The clinical and bench work is done in collaboration with **Drs. Gozansky, Pereira, MacLean, Jackman, and Friedman**.

Dr. Rachael Van Pelt has found that adipose tissue in lower body regions has favorable associations with certain risk factors for cardiovascular disease in women. After adjusting for either trunk fat mass or intra-abdominal fat area, she found that leg fat mass is inversely associated with serum triglyceride level. The hypothesis is that adipocytes in peripheral regions avidly sequester triglycerides. She is now studying (R01) whether reducing lower extremity fat mass increases serum triglycerides or alters other cardiovascular disease risk factors. Pre- and postmenopausal women undergo lower-extremity lipectomy and are followed for 18 months. Post-prandial lipid kinetics, using an isotope-labeled meal, are evaluated before and at several time points after surgery. Adipose tissue biopsies are obtained to determine the fate of dietary fats. The hypothesis is that post-prandial lipemia will be exaggerated after lower extremity lipectomy in both pre- and postmenopausal women, but that it will normalize after 18 months in premenopausal women because they will re-accumulate lower extremity adipose tissue. In contrast, it is hypothesized that postmenopausal women will re-accumulate fat in central body regions and this will be associated with persistent adverse changes in post-prandial lipemia. These studies are conducted in collaboration with **Drs. Kohrt, Pereira, and Bessesen**.

A factor that may contribute to the increased propensity for fat gain in menopausal women is the loss of lean mass. A number of ongoing studies (**Drs. Kohrt, Barry, Villalon, and Jankowski**) are focused on novel strategies to prevent the well-described effects of estrogen deficiency on

bone metabolism. However, a more novel focus of this research is on skeletal muscle metabolism. **Dr. Jankowski** is studying the potential anti-catabolic effects of estrogens in muscle (Department of Medicine pilot). Premenopausal women undergo 14 days of GnRH antagonist therapy (GnRHant), to acutely suppress sex hormones, with or without estrogen add-back therapy. During the first 9 days of GnRHant, one leg is immobilized (knee brace in 135 degrees of flexion, use of crutches); remobilization plus exercise occurs during the final 5 days of GnRHant. Muscle biopsies (v lateralis) are performed before and 9 and 14 days after GnRHant for evaluation of signaling factors associated with muscle anabolism (Akt, mTOR, p70s6k) and catabolism (FOXO, atrogen1). The primary hypothesis is that estrogen will prevent the immobilization-related increase in catabolic signaling factors. Collaborators on this project include **Drs. Kohrt, Gozansky, Friedman, and MacLean**.

There is a solid core of translational investigators (**Drs. Moreau, Reusch, Schauer, and Regensteiner**) studying the role of estrogen in vascular function in menopausal women. **Dr. Kerrie Moreau** (R01) is challenging the current ‘timing hypothesis’ that estrogens have different biological effects in early versus late menopausal women. Pre-, peri- (early and late), and postmenopausal women undergo GnRHant therapy with placebo or estrogen add-back therapy. A complement of tests is performed to evaluate vascular function. The major hypothesis is that estrogen will have favorable effects on vascular function in pre- and perimenopausal women, but not postmenopausal women. A novel translational aspect of the study involves the collection of vascular endothelial cells from an antecubital vein for *in vitro* experiments of factors that influence endothelial cell function. Collaborators on these studies include **Drs. Kohrt, Schwartz, Gozansky, and Reusch**. **Dr. Irene Schauer** (BIRCWH K12, CWHR pilot) and her mentors, **Drs. Regensteiner and Reusch**, are exploring the role of serum fatty acids in insulin resistance and vascular dysfunction in adults with type 1 diabetes (T1D). Serum fatty acid levels are either elevated (fat emulsion plus heparin) or suppressed (acipimox) during euglycemic-hyperinsulinemic clamps. Preliminary data support the hypothesis that elevated fatty acids contribute to insulin resistance and vascular dysfunction in T1D. However, there appear to be sex differences, such that the consequences of elevated fatty acids are exaggerated in women. The role of estrogen as a mediator of this response is not clear. **Drs. Van Pelt, Gozansky, Schwartz, and Kohrt** have demonstrated that, in healthy menopausal women, even acute exposure to estrogen can increase the glucoregulatory action of estrogen; years since menopause may be an important determinant of this response (i.e., older women may be less responsive).

The members of this Translational Unit on Menopause, Obesity, and Disease Risk are anxiously awaiting the arrival at UCD of **Dr. Nanette Santoro**, who was recently appointed Chief of the Department of Obstetrics and Gynecology. Dr. Santoro’s major research interests include the reproductive endocrinology of premature, peri- and postmenopause, infertility, and the physiology of gonadotropin-releasing hormone secretion. She has been involved with numerous industry- and government-supported clinical trials, including the Study of Women’s Health Across the Nation (SWAN), the Kronos Early Estrogen Prevention Study (KEEPS), and the Reproductive Physiology of Ovarian Failure.

How the Center Has Contributed to Bringing New Investigators to Nutrition Research

Facilitating Development of Junior Investigators

We continue to be especially proud of our success in facilitating the career development of our young investigators. Since the establishment of the NORC, an extraordinarily high proportion of our new investigators have obtained NIH funding and a large number of new collaborations among established investigators have led to new NIH grants. Currently, 11 members of our research base have K01, K02, K08, K23, or career development awards from the NIH.

Creating an Atmosphere of Collaboration

In Colorado, we have created an exceptional environment for nutrition and obesity research. The environment consists of: 1) well-funded senior scientists with strong mentoring skills; 2) an atmosphere that encourages collaboration, with research programs funded by multiple grants from multiple investigators and supporting junior and senior investigators; 3) a stream of talented junior investigators attracted and supported through institutional training grants, facilitated by NORC pilot project funding, and mentored successfully in their career advancement; 4) scientific core laboratories with specialized equipment and sophisticated, cost-effective techniques that are not possible in individual laboratories; 5) an enrichment program that brings people together and encourages interactions around nutrition and obesity issues; and 6) substantial support from our university. However, the environment created by the NORC is truly more than the sum of these parts. We have created a research and outreach continuum that extends from the patient's nutritional problem(s) to the research laboratory and back to fulfillment of the patient's need(s). In this environment, nutrition and obesity researchers are limited only by their ideas and energy, not by the environment.

Leveraging the Colorado NORC To Create New Programs

There have been two major and exciting developments related to obesity and nutrition during the past year.

1. Last year we officially established the Anschutz Health & Wellness Center. This was made possible by \$15 million given by the Anschutz Foundation. The UCD contributed approximately \$25 million additional to create the new program and to fund a new building of approximately 100,000 square feet. Groundbreaking for the new building is anticipated for August or September 2010. The program will focus on 1) research in diet, physical activity, and weight management; 2) development of clinical programs to improve lifestyle to optimize health and prevent chronic disease; 3) translating science into the community; and 4) education for students, health care professionals, and the public.
2. We were successful in securing support for a new University of Colorado School of Medicine initiative in obesity. This program, Colorado Obesity Research Initiative, will link and support more than 100 researchers at the University of Colorado who are working in the area of obesity. We are finalizing the specific amount of support available but it is anticipated to be in the range of \$5 million to \$8 million over 3 years.