Project Grow-2-Gether: A Study of the Genetic and Environmental Influences on Child Eating and Obesity

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"Project Grow-2-Gether" is a child nutrition study of same-sex, 3- to 7-year-old monozygotic and dizygotic twin pairs. The study attempts to bridge two bodies of literature that have rarely interfaced with respect to obesity and ingestive behavior: the first being behavioral genetic approaches to obesity-related traits, and the second being developmental approaches focusing on parent–child relationships. The overarching aim of Project Grow-2-Gether is to disentangle genetic from potential home-environmental influences on child eating behavior and body fat. This paper reviews the rationale for Project Grow-2-Gether, its procedures, and core phenotypic measurement battery. A focus of the study is acquisition of controlled food intake measurements obtained in the laboratory, measurement of specific home environmental variables, and multi-method evaluation of parent–child feeding relations. Future directions may involve longitudinal assessment of child growth and molecular analyses for specific genes that influence child eating behavior.

Background

Childhood obesity is a complex disorder influenced by multiple genetic and environmental factors (Faith et al., 1999; Rosenbaum & Leibel, 1998; Segal & Allison, 2002). To better identify these factors, genetically informative studies have phenotyped subjects using state-of-the art metabolic, physiologic, and body composition technologies to pinpoint specific obesity-promoting genes (Pérusse et al., 2001). However, obesity results from elevated energy intake relative to energy expenditure (i.e., "positive energy balance"), and obese children eat more calories on average than non-obese children. The study of individual differences in eating behavior is therefore critical to understanding etiological pathways of childhood obesity. Yet few genetics studies have been built around behavioral phenotyping of food selection, dietary patterns, or energy intake measures in children (Allison et al., in press; Faith et al., 1997; Faith et al., in press).

This paper reviews the overarching aims and methods of "Project Grow-2-Gether" — a same-sex twin study designed to decompose genetic and environmental influences on the eating patterns and body composition measures of young children. The emphasis of this study concerns objectively measured food intake phenotypes obtained under controlled laboratory conditions. The study also obtains multiple measures of maternal feeding practices to test the impact of this home-environmental variable on child eating regulation and body fat. As described below, certain developmental theories speculate that maternal control during feeding may disrupt a child's ability to recognize satiety signals and hence be a risk factor for childhood obesity (Birch & Fisher, 1998). Project Grow-2-Gether explores this hypothesis in the context of a twin study, to better control for potential genetic influences on the child's eating behavior and body fat.

Evidence for Genetic Influences on Child Eating Behavior

Evidence for genetic influences on child *body fat* is extensive. The broad heritability of human obesity seems to fall in the range of 65–70% (Allison et al., 1996; Segal & Allison, 2002). Each year, the list of candidate genes for human obesity increases (Pérusse et al., 2001). Select data suggest that differences in human food preferences and eating patterns are also partially genetically influenced (Keller et al., 2002). Classical studies in adults using food records to measure ad libitum energy intake in the free-living environment, found that total energy intake, macronutrient-specific selection, and issues related to meal timing and frequency all had a heritable component (DeCastro, 1999). In a study that measured ad libitum food intake in the laboratory, we found that genetic varia-

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tions accounted for approximately one third the variance in adult's total energy intake (Faith et al., 1999). Other studies have reported a small but significant heritable component for particular food preferences (Reed et al., 1997). Numerous animal studies demonstrate a genetic influence on food intake (Allison et al., in press). Collectively, these data support one of Project Grow-2-Gether's central hypotheses: that there should be a significant heritable component to children's energy intake phenotypes.

Evidence to Suggest Maternal Feeding Style Influences on Child Eating Behavior

In classical experiments by Davis (1939), young children demonstrated an ability to self-select appropriate levels of total and macronutrient intake to support growth when given access to an array of healthy foods. These results, in conjunction with findings by Fomon (1993), and Johnson & Birch (1994), suggest that young children may have an ability to detect internal hunger and satiety cues and adjust their eating accordingly. Consistent with this theory are certain data showing that children will adjust energy intake at meals proportional to energy consumed approximately 30 minutes earlier as a snack — a phenomenon called "caloric compensation" (Birch & Deysher, 1985). Cross-sectional data suggest that increased parental control during feeding or feeding prompts are associated with poorer child caloric compensation (Birch & Fisher, 1998), increased child eating in the absence of hunger (Fisher & Birch, 1999), and increased child body weight (Klesges et al., 1983). These data have led to the hypothesis that increased parental feeding control may impede children's ability to learn caloric regulation and, indirectly, be a risk factor for childhood obesity. Although not all studies support this hypothesis (Saelens et al., 2000), the theory is provocative and additional studies are needed to distinguish whether correlations between parent and child phenotypes represent causal effects.

Project Grow-2-Gether Methods

Overview

The main goals of this study are: (1) To test whether there is a significant genetic contribution to caloric compensation and total energy intake in preschool children, and (2) To test whether certain parental feeding styles promote poorer caloric compensation and increased body weight in preschool children, controlling for concurrent genetic influences on child eating and weight.

Subjects

Project Grow-2-Gether recruits same-sex twins, of all ethnic backgrounds, who are between 3 and 7 years old. Families are recruited by general newspaper advertisements, target mailing, word-of-mouth, and web page (http://cpmcnet.columbia.edu/dept/obesectr/NYORC/twins.html). Parents are told that they will participate in a study looking at possible genetic influences on young children's earliest food preferences and eating patterns and parental feeding attitudes. We have collected data from 67 families as of 7.29.02. The ethnicity breakdown of these families to date is approximately 40% Non-Hispanic White, 25% African-American, 25% Hispanic-American, and 10% Asian-American.

Overview of Procedural Design

Table 1 summarizes the main procedural components that occur over four days and the data yielded. Days 1 and 2 are primarily devoted to collection of food intake data necessary for computation of child's "caloric compensation" ability (described below). Questionnaire data pertaining to home environmental and reported feeding style variables are also collected on these visits. Days 3 and 4 are dedicated to obtaining replicate measures of video-recorded mother–child feeding interactions taken at ad libitum buffet lunch meals. Preloads are not provided on visits 3 and 4; hence,

Table 1

Overview of Four Laboratory Visits

Approximate Times	Day 1	Day 2	Day 3	Day 4
11:30–12:00	Family arrives at Child Feeding Lab; Consent obtained. Children play and become familiar with staff.	Family arrives. Mother completes questionnaires. Children play.	Family arrives. One twin eats in an isolated room with the mother. Mother–child interactions during lunch are videotaped.	Family arrives. One twin eats in an isolated room with the mother. Lunch is videotaped.
12:00–12:30	Children drink high or low calorie preload drink, followed by play time until lunch.	Children drink high or low calorie preload drink followed by play time until lunch.	The other twin eats lunch with the mother. Mother–child interactions during lunch are videotaped.	The other twin eats lunch with the mother. Lunch is videotaped.
12:30–1:00	Children eat lunch while staff member reads to them.	Children eat lunch while staff member reads to them. Children are given cheek swab for zygosity tests.	Child body composition assessment.	Children get pictures taken to display in lab.
1:00–1:30	Children receive prize; Instructions given for next visit; Family is escorted out by staff.	Children receive prize. Instructions given for next visit. Family is escorted out by staff.	Children receive prize. Instructions given for next visit. Family is escorted out by staff.	Children receive prize. Family is escorted out by staff.

Table 2

Overview of Phenotypic Measurements from Project Grow-2-Gether

Phenotype Category	Description
Eating Measures	
"Caloric Compensation"	Ability to adjust energy intake at laboratory multi-item lunches following consumption of a low-calorie or high-calorie "preload" drink. Behaviorally measured under controlled laboratory conditions. Twins eat at same table, but not allowed to eat from each others' plates.
Ad Libitum Total and Macronutrient Intake	Ad libitum consumption of self-selected foods served at a buffet lunch. Measures taken on 2 occasions to allow for replicate measures. Behaviorally measured under controlled laboratory conditions. Twins eat separately, such that independent measurements can be obtained.
Body Composition	
Anthropometrics	A long-standing method for evaluating growth and body fatness. This technique uses callipers to grasp a skinfold to provide a measurement (mm) for a double fold of skin and subcutaneous fat.
Bioimpedance Analysis (BIA)	A quick and simple method to estimate total body water, which is translated into fat-free (lean) mass by standard equations.
Dual Energy X-Ray Absorptiometry (DXA)	Laboratory method to estimate fat, lean and bone mineral content of the body
Maternal Feeding Style	
Questionnaires	Variety of self-report instruments assessing maternal feeding style, general parenting style, eating attitudes, maternal body image, and other aspects of the home environment. Mothers rate each child individually, when possible, to yield independent ratings for each child.
Video-taped Mother–child Dyadic Interactions	Observational assessment of mother–child interactions during lunch buffet sessions. Mothers interact separately with each child. A variety of feeding behaviors are coded including "Encouragement" and "Discouragement." Child behaviors also recorded (e.g., "Request" and "Refusal").

these measures represent total and macronutrient-specific intake rather than caloric compensation. Finally, body composition data are collected on Day 4.

Phenotypic Measurements

Table 2 summarizes the main phenotypic measurements collected in Project Grow-2-Gether. These include:

Compensation lunches. During visits 1 and 2, children consume a "preload" liquid fruit drink that contains 150 kcal (628 kJ) or 3 kcal (12.5 kJ), respectively (the exact order is randomized). After drinking the preload and following a 25 minute interim period, children consume a multi-item lunch including macaroni and cheese, canned string beans, string cheese, graham crackers, green grapes, baby carrots, and whole milk. By pre- and post-weighing foods and converting to calories, we compute the difference in energy intake following the low- and high-calorie preloads. This difference score is scaled in such a way to represent each child's caloric compensation ability.

Ad libitum food intake. During the third and fourth visits, each child is separately presented with a buffet lunch in a videotaped room. Each twin eats alone with the mother, who is present but is not consuming food. The mother is instructed to act as she normally would at home during mealtime. The following foods are presented: carrots, raisins, apple sauce, yogurt, peas, corn, string cheese, cheese crackers, bologna sandwich with white and wheat bread, turkey sandwich with white and wheat bread, chicken nuggets, mustard, mayonnaise, ketchup, coke, whole milk, apple juice, orange juice, fruit punch, chocolate milk, Oreo cookies, fruit rollup, chocolate pudding, jello, Hershey kisses, graham crackers, ring dings, granola bar, potato chips, banana, and fruit cocktail. These foods were chosen because they are all familiar to and liked by young children. Foods are pre- and post-weighed for calculation of energy intake.

During lunches, we video-record dyadic interactions between mother and each respective twin. We conduct a frame-by-frame coding and analysis of tapes using the Noldus Observer Video-Pro 4.0 software (www.noldus.com) to code the following maternal feeding behaviors: Encouragement To Eat, Discouragement To Eat, Manners, and Probing Child Satiety. We code child behavior for: Food Request and Food Refusal.

Questionnaires. To gather information on self-reported maternal feeding styles, eating attitudes, and body image, mothers complete a comprehensive questionnaire battery including: the Child Feeding Questionnaire (Birch et al., 2001), the Food Neophobia Scale (Pliner, 1994), and the Eating Disorders Examination Questionnaire (Wilfley et al., 1997).

Body Composition Measures

Anthropometrics. Established methods are used for evaluating growth and subcutaneous fat (Lohman et al., 1988) including weight (kg), height (cm) and various skinfold thicknesses and waist circumference.

Bioimpedance analysis (BIA). BIA provides an estimate of total water, which is transformed into fat free mass (lean) based on the concept that tissues rich in water and electrolytes are much more resistant to the passage of an imperceptible electrical current than lipid-rich adipose

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tissue. This is a quick, safe and reasonably accurate procedure for children (Hymsfield et al., 1998).

Dual energy x-ray absorptiometry (DXA). DXA provides estimates of fat, lean soft tissue and bone mineral content of the body. DXA can also assess regional body composition (i.e., trunk, arms, and legs). The method is based on the differential attenuation of two photon beans as they are absorbed by various body tissues. DXA requires minimal cooperation from the participant, is relatively quick (approximately 10 minutes) for the total body scan measurements, and has minimal radiation exposure that presents no practical health risk to subjects (Goran et al., 1996; Pietrobelli et al., 2001).

Genetic Information

We collect buccal swabs for zygosity determination based on analysis of 10 highly polymorphic markers.

Data Analyses

We will use the full range classical twin modeling methods to test the relative impact of genetic, environmental, and maternal feeding patterns on child eating and body composition measures (Neale & Cardon, 1992).

Future Directions

Future plans for Project Grow-2-Gether involve longitudinal follow-up assessment of children as they grow, and collection for DNA samples for potential molecular analyses of specific quantitative trait loci that influence child eating behavior. Projects with external collaborators are welcome.

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References

Allison, D. B., Kaprio, J., Korkeila, M., Koskenvuo, M., Neale, M. C., & Hayakawa, K. (1996). The heritability of BMI among an international sample of monozygotic twins reared apart. *International Journal of Obesity, 20,* 501–506.

Allison, D. B., Pietrobelli, A., Faith, M. S., Fontaine, K. R., Gropp, E., & Fernandez, J. R. (in press). In R.H. Eckel (Ed.), *Obesity*. New York, NY: Lippincott Williams & Wilkins.

Birch, L. L., & Deysher, M. (1985). Conditioned and unconditioned caloric compensation: Evidence for self-regulation of food intake by young children. *Learning and Motivation, 16,* 341–355.

Birch, L. L., & Fisher, J. O. (1998). Development of eating behaviors among children and adolescents. *Pediatrics, 101,* 539–549.

Birch, L. L., Fisher, J. O., Grimm-Thomas, K., Markey, C. N., Sawyer, R., & Johnson, S. L. (2001). Confirmatory factor analysis of the Child Feeding Questionnaire: A measure of parental attitudes, beliefs and practices about child feeding and obesity proneness. *Appetite, 36,* 201–210.

Davis, C. M. (1939). Results of the self-selection of diets by young children. *Canadian Medical Association Journal, 41,* 257–261.

DeCastro, J. M. (1999). Behavioral genetics of food intake regulation in free-living humans. *Nutrition, 7/8,* 550–554.

Faith, M. S., Johnson, S. L., & Allison, D. B. (1997). Putting the "behavior" into the behavior genetics of obesity research. *Behavior Genetics, 27,* 423–439.

Faith, M. S., Pietrobelli, A., Nuñez, C., Heo, M., Heymsfield, S. B., & Allison, D. B. (1999). Evidence for independent genetic influences on fat mass and body mass index in a pediatric twin sample. *Pediatrics, 104,* 61–67.

Faith, M. S., Rha, S. S., Neale, M. C., et al. (1999). Evidence for genetic influences on human energy intake: Results from a twin study using measured observations. *Behavior Genetics, 29,* 145–154.

Faith, M. S., Tepper, B. J., Hoffman, D. J., & Pietrobelli, A. (in press). Genetic and environmental influences on childhood obesity. *Clinics Family Practice*.

Fisher, J. O., & Birch, L. L. (1999). Restricting access to foods and children's eating. *Appetite, 32,* 405–419.

Fomon, S. J. (1993). *Nutrition of normal infants.* St. Louis, MO: Mosby-Year Book.

Goran, M. I., Driscoll, P., Johnson, R., Nagy, T., & Hunter, G. (1996). Cross validation of body composition techniques against dual-energy X-ray absorptiometry in young children. *American Journal of Clinical Nutrition, 63,* 299–305.

Heymsfield, S. B., Nunez, C., & Pietrobelli, A. (1998). Bioimpedance analysis: What are the next steps? *Nutrition in Clinical Practice, 12,* 201–203.

Johnson, S. L., & Birch, L. L. (1994). Parent's and children's adiposity and eating style. *Pediatrics, 94,* 653–661.

Keller, K. L., Pietrobelli, A., Must, S., & Faith, M. S. (2002). Genetics of eating and its relation to obesity. *Current Atherosclerosis Reports, 4,* 176–182.

Klesges, R. C., et al. (1983). Parental influences on children's eating behavior and relative weight. *Journal of Applied Behavioral Analysis, 16,* 371–378.

Lohman, T. G., Roche, A. F., & Martorell, L. (1988). *Anthropometric standardization reference manual.* Champaign, IL: Human Kinatics.

Neale, M. C., & Cardon, L. R. (1992). *Methods for genetic studies of twins and families.* Boston, MA: Kluwer Academic Publishers.

Pérusse, L., Chagnon, Y. C., Weisnagel, J., et al. (2001). The human obesity gene map: The 2000 update. *Obesity Research, 9,* 136–168.

Pietrobelli, A., Heymsfield, S. B., ZiMiang, W., & Gallagher, D. (2001). Multi-component body composition models: Recent advances and future directions. *European Journal of Clinical Nutrition, 55,* 69–75.

Pliner, P. (1994). Development of measures of food neophobia in children. *Appetite, 23,* 147–163.

Reed, D. R., Bachmanov, A. A., & Beachamp, G. K. (1997). Heritable variation in food preferences and their contribution to obesity. *Behavior Genetics, 27,* 373–87.

Rosenbaum, M., & Leibel, R. L. (1998). The physiology of body weight regulation: Relevance to the etiology of obesity in children. *Pediatrics, 101,* 525–539.

Saelens, B. E., Ernst, M. M., & Epstein, L. H. (2000). Maternal child feeding practices and obesity: A discordant sibling analysis. *International Journal of Eating Disorders, 27,* 459–463.

Segal, N., & Allison, D. B. (2002). Twins and virtual twins: Bases of relative body weight revisited. *International Journal of Obesity, 4,* 437–441.

Wilfley, D., Schwartz, M. B., Spurrell, E. B., & Fairburn, C. G. (1999). Assessing the specific psychopathology of binge eating disorder patients: Interview or self-report? *Behaviour Research and Therapy, 35,* 1151–1159.

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