Final Progress Report for Research Projects Funded by Health Research Grants

Instructions: Please complete all of the items as instructed. Do not delete instructions. Do not leave any items blank; responses must be provided for all items. If your response to an item is "None", please specify "None" as your response. "Not applicable" is not an acceptable response for any of the items. There is no limit to the length of your response to any question. Responses should be single-spaced, no smaller than 12-point type. The report **must be completed using MS Word**. Submitted reports must be Word documents; they should not be converted to pdf format. Questions? Contact Health Research Program staff at 717-783-2548.

1. **Grantee Institution:** Monell Chemical Senses Center

2. **Reporting Period (start and end date of grant award period):** 01/01/10-12/31/10

3. **Grant Contact Person (First Name, M.I., Last Name, Degrees):** Gary K. Beauchamp, PhD

4. **Grant Contact Person's Telephone Number:** 267-519-4700

5. **Grant SAP Number:** 4100050917

6. **Project Number and Title of Research Project:** *Relationship between Parental Smoking, Dietary Habits, Salt and Sweet Preferences and Blood Pressure among Children*

7. **Start and End Date of Research Project:** 01/01/10-12/31/10

8. **Name of Principal Investigator for the Research Project:** Julie Mennella, PhD

9. **Research Project Expenses.**

   9(A) Please provide the amount of health research grant funds spent on this project for the entire duration of the grant, including any interest earned that was spent:

   $240,527.93

   9(B) Provide the last names (include first initial if multiple individuals with the same last name are listed) of **all** persons who worked on this research project and were supported with health research funds. Include position titles (Principal Investigator, Graduate Assistant, Post-doctoral Fellow, etc.), percent of effort on project and total health research funds expended for the position. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project	Cost
Mennella	Member	15%	$20,470.45
Reed	Member	10%	$10,797.39
Finkbeiner	Senior Technician	100%	$33,796.17
Castor	Research Analyst	50%	$13,694.35
Duke	Senior Technician	25%	$ 6,463.87
Hwang	Technician	25%	$ 2,229.97
Lysenko	Technician	25%	$ 182.70
Khoshnevisan	Technician	25%	$ 321.75

9(C) Provide the names of **all** persons who worked on this research project, but who *were not* supported with health research funds. Include position titles (Research Assistant, Administrative Assistant, etc.) and percent of effort on project. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project
Miller	Postdoctoral Fellow	10%
Lipchock	Postdoctoral Fellow	5%
Silva-Garcia	Graduate Student	5%
Daniels	Student Research Apprentice	5%
Crowley	Student Research Apprentice	5%
Burdick-Wills	Student Research Apprentice	5%

9(D) Provide a list of **all** scientific equipment purchased as part of this research grant, a short description of the value (benefit) derived by the institution from this equipment, and the cost of the equipment.

Type of Scientific Equipment	Value Derived	Cost
None		

10. Co-funding of Research Project during Health Research Grant Award Period. Did this research project receive funding from any other source during the project period when it was supported by the health research grant?

Yes_____ No ____X_____

If yes, please indicate the source and amount of other funds:

11. Leveraging of Additional Funds

11(A) As a result of the health research funds provided for this research project, were you able to apply for and/or obtain funding from other sources to continue or expand the research?

Yes____x_____ No_____

If yes, please list the applications submitted (column A), the funding agency (National Institutes of Health—NIH, or other source in column B), the month and year when the application was submitted (column C), and the amount of funds requested (column D). If you have received a notice that the grant will be funded, please indicate the amount of funds to be awarded (column E). If the grant was not funded, insert "not funded" in column E.

Do not include funding from your own institution or from CURE (tobacco settlement funds). Do not include grants submitted prior to the start date of the grant as shown in Question 2. If you list grants submitted within 1-6 months of the start date of this grant, add a statement below the table indicating how the data/results from this project were used to secure that grant.

A. Title of research project on grant application	B. Funding agency (check those that apply)	C. Month and Year Submitted	D. Amount of funds requested:	E. Amount of funds to be awarded:
Efficacy of Bitter Blockers on Flavor Acceptance in Pediatric Populations	X NIH ☐ Other federal (specify:_____) ☐ Nonfederal source (specify: _____)	February, 2010	$1,865,083	$1,585,321

11(B) Are you planning to apply for additional funding in the future to continue or expand the research?

Yes___x____ No_____

If yes, please describe your plans:

During the upcoming year, we plan to submit two grants to NIH (one a National Research Service Award from a postdoctoral fellow in Dr. Mennella's lab- Dr. Stacie Miller and another a R01) on the effect of parental smoking on health of children, including impact on taste and odor preferences and chemical sensitivity.

12. Future of Research Project. What are the future plans for this research project?

We are currently writing a manuscript for a peer-reviewed journal and will be presenting these findings at the Annual Meeting of the Association of Chemoreception Sciences in April 2011. If we are successful in obtaining more funding, we hope to develop a program on the effects of parental smoking on child's health and strengthen our collaborations with Dr. Lerman and colleagues at the University of Pennsylvania.

13. New Investigator Training and Development. Did students participate in project supported internships or graduate or post-graduate training for at least one semester or one summer?

Yes_____x___ No_____

If yes, how many students? Please specify in the tables below:

	Undergraduate/HS	Masters	Pre-doc	Post-doc
Male	0	0	0	0
Female	3	1	0	2
Unknown	0	0	0	0
Total	**3**	**1**	**0**	**2**

	Undergraduate/HS	Masters	Pre-doc	Post-doc
Hispanic	0	1	0	0
Non-Hispanic	3	0	0	2
Unknown	0	0	0	0
Total	**3**	**1**	**0**	**2**

	Undergraduate/HS	Masters	Pre-doc	Post-doc
White	2	1	0	2
Black	1	0	0	0
Asian	0	0	0	0
Other	0	0	0	0
Unknown	0	0	0	0
Total	**3**	**1**	**0**	**2**

14. **Recruitment of Out-of–State Researchers**. Did you bring researchers into Pennsylvania to carry out this research project?

    Yes_____ No_____x_____

    If yes, please list the name and degree of each researcher and his/her previous affiliation:

15. **Impact on Research Capacity and Quality**. Did the health research project enhance the quality and/or capacity of research at your institution?

    Yes__x_____ No_____

    If yes, describe how improvements in infrastructure, the addition of new investigators, and other resources have led to more and better research.

    This project allowed us to train new investigators in the field. One of the post-doctoral fellows, Dr. Stacie Miller, is planning on submitting a National Research Service Award (NRSA) to NIH to study the effects of taste and olfactory learning in children whose parents smoke.

16. Collaboration, business and community involvement.

16(A) Did the health research funds lead to collaboration with research partners outside of your institution (e.g., entire university, entire hospital system)?

Yes_____ No____x_____

If yes, please describe the collaborations:

16(B) Did the research project result in commercial development of any research products?

Yes_____ No____x_____

If yes, please describe commercial development activities that resulted from the research project:

16(C) Did the research lead to new involvement with the community?

Yes____x_____ No_____

If yes, please describe involvement with community groups that resulted from the research project:

The opportunity to participate in this research project was available to all women with children regardless of race or income. As a result, some of our subjects were recruited through the WIC (Women, Infant, and Children) programs. Participation in this research program raises awareness about nutrition and taste among women from all strata of education and income and therefore is of benefit to the participants.

17. Progress in Achieving Research Goals, Objectives and Aims.

List the project goals, objectives and specific aims (as contained in the grant application's strategic plan). Summarize the progress made in achieving these goals, objectives and aims for the entire grant award period. Indicate whether or not each goal/objective/aim was achieved; if something was not achieved, note the reasons why. Describe the methods used. If changes were made to the research goals/objectives/aims, methods, design or timeline since the original grant application was submitted, please describe the changes. Provide detailed results of the project. Include evidence of the data that was generated and analyzed, and provide tables, graphs, and figures of the data. List published abstracts, poster presentations and scientific meeting presentations at the end of the summary of progress; peer-reviewed publications should be listed under item 20.

This response should be a DETAILED report of the methods and findings. It is not sufficient to state that the work was completed. Insufficient information may result in an unfavorable performance review, which may jeopardize future funding. If research findings are pending publication you must still include enough detail for the expert peer reviewers to evaluate the progress during the course of the project.

Health research grants funded under the Tobacco Settlement Act will be evaluated via a performance review by an expert panel of researchers and clinicians who will assess project work using this Final Progress Report, all project Annual Reports and the project's strategic plan. After the final performance review of each project is complete, approximately 12-16 months after the end of the grant, this Final Progress Report, as well as the Final Performance Review Report containing the comments of the expert review panel, and the grantee's written response to the Final Performance Review Report, will be posted on the CURE Web site.

There is no limit to the length of your response. Responses must be single-spaced below, no smaller than 12-point type. If you cut and paste text from a publication, be sure symbols print properly, e.g., the Greek symbol for alpha (α) and beta (ß) should not print as boxes (□) and include the appropriate citation(s). DO NOT DELETE THESE INSTRUCTIONS.

Project Goals and Specific Aims

The overall goal of this research project was to characterize the taste preferences of children and their mothers for sweet and salty taste and to test the hypothesis that women who smoke and their children prefer higher concentrations of salt and sugar and consume more foods of low quality, e.g., high sugar, high salt, junk food, which are related to blood pressure. We also examined whether high salt preferences are related to high sweet preferences, e.g., a liking for extremes of tastes often found in highly processed food, or whether there is the opposite relationship, e.g., that some children prefer high salt foods whereas others have a sweet tooth. These individual differences in taste preferences were evaluated for their genetic roots through association studies.

To this end, the research project had two specific aims. First, we used a sophisticated psychophysical technique, which was developed at the Monell Center, to assess taste preferences in children and their mothers. This allowed us to determine whether children of smokers prefer a higher level of salt, which may be related to dietary salt intake and blood pressure. Although never to our knowledge studied in children of smokers, there is evidence that the level of salt preferred by adults is dependent on level of salt consumed in the diet (1-3). Further, there is a positive and significant association between salt intake and systolic blood pressure in children (4). The data also allowed us to explore for the first time whether there is a relationship between salt and sweet liking in adults and children, specifically whether greater salt taste liking drives consumption of salt and sweetened foods and beverages. We expected that children whose parents smoke would prefer more intense sweetness and eat more sweets, as assessed by dietary records, than children of nonsmokers. This expectation is based on our previous findings that children with a family history of addiction prefer stronger sweet solutions than those without (5, 6) and 2) adults with a family history of alcoholism are more likely to smoke tobacco (7, 8).

The second aim related to the influence of genetic variation on sweet and salty perception and liking in children and their mothers. To this end, genes known to affect sweet sensitivity in humans were genotyped (*TAS1R3* (9)) as well as alleles related to the consumption of sweet food (10, 11) . While there are no known genes and genetic variants that affect salt intake, recent advances at Monell towards the discovery of the salt receptor and key components in sensory transduction have been discovered and allelic variation within the salt receptor gene was typed (*SCNN1D*). There may be a

direct effect of taste on smoking behavior because people who are genetically blind to some types of bitter taste might find tobacco smoke particle less bitter and thus more willing to experiment with smoking and become habitual smokers (12-14). This hypothesis has recently gained support because taste receptors, similar to those found in the tongue, are also found in the airways (15-17). The best known example of genetic blindness to bitter comes from the study of *TAS2R38*, one of the 25 human bitter receptors. This gene has two common alleles, a taster and non-taster form, and the distribution of these genotypes among smoker and non-smoker groups were examined. The hypothesis tested is that people who are genetically blind to certain types of bitter molecules are more likely to be smokers.

Experimental Design/Overview.

Each mother-child dyad was phenotyped for a) preferences and sensitivity for salts and sugar using age-appropriate psychophysical testings (e.g., two-alternative, forced-choice tracking procedure), b) anthoprometry and biometrics, c) mood and dietary intake. Cheekswab/saliva samples were obtained from which DNA was isolated, purified and quantitated for alleles of the human sweet receptor (*TAS1R3*) and other alleles previously identified as important in sweet taste perception (9, 18). For salt, alleles of the putative receptor were genotyped (19). Smoking history, and food habits/cravings were asessed by standardized questionnaires, as described herein.

As shown in **Table 1**, each mother and child was tested on two days separated by one week, approximately 1 hour before their next scheduled meal. Testing lasted 1.5-2 hours each day and occurred in a newly renovated testing facility designed for psychophysical testing of children and their parents. At the beginning of each test day, the mother and child were asked to breath into Vitalograph (Lenexa, KS) to obtain a measure of carbon monoxide levels, we used psychophysical test to determine salt and sucrose preferences. On each testing day, blood pressure readings were obtained to determine whether salt liking was related to blood pressure in children and adults. Dietary recall data were used to determine whether higher intake of salt related to higher salt liking. While their children were being tested and throughout the test sesion, mothers were asked questions related to their and their children's food habits and mood states. Mothers and children were assessed for various aspects of taste perception and related phenotypes. The techniques used herein have a long history of reliable use and validity in our laboratory. The measures involve asking children and their mothers to taste foods or liquids and, using forced-choice tracking procedures, to idenfity the ones they prefer. These tasks were developed at Monell to meet the communicative, cognitive (attention and memory) and behavioral capacities of pediatric populations.

I. Psychophysical Testing Procedures. These testing procedures capture a wide range of perceptual and preferences data regarding salt and sweet psychophysics.

<u>IA. Salt and Sucrose Preferences</u>. The most preferred level of sucrose in water (0.09, 0.18, 0.35, 0.70, 1.05M or 3, 6, 12, 24 and 36 % wt/vol, respectively) and salt in vegetable broth (0.16, 0.24, 0.38, 0.61, 1.05M or 0.92, 1.39, 2.20, 3.59, 6.14 % wt/vol, respectively) were assessed by a 2-series, forced-choice tracking psychophysical technique developed at the Monell Center (20). In brief, subjects were presented with pairs of solutions and they were allowed to taste each solution for 5 seconds without swallowing, and then point to which of the pair they liked better. Each subsequent pair was then determined by the participant's preceding preference choice. Participants rinsed and expectorated with water once after tasting each sample and twice between each pair of solutions, and a timer was used to ensure that a one-minute interval separated each pair presentation. The procedure

continues until the participant chose either a given concentration of sucrose (or salt) when it was paired with both a higher and lower concentration or when they chose the highest or lowest concentration two consecutive times.

In addition to using sucrose and salty solutions for evaluating hedonic responses to these tastes, we also evaluated preferences using foods, the latter of which is a better choice for taste hedonics (21). We used the method of Bertino, Beauchamp and colleagues (3) to determine the concentrations of added salt most preferred in crackers (0.80, 2.90 and 6.12 % wt/wt respectively) and concentration of sugar most preferred in jellies (30, 40, 50, 60 and 70 %wt/wt respectively).

II. Anthromopetry and Biometrics. These methods were employed to measure blood pressure and several aspects of obesity: body weight, percent body fat and body shape, e.g., apple versus pear body shapes.

IIA. BMI and Weight Category. Weight and height measurements were obtained for each subject. Body mass index (BMI; the weight in kilograms divided by the square of the height in meters) was then computed. For women, BMI was categorized as follows: 18.5 kg/m^2 to 24.9 kg/m^2 (normal weight), 25.0 kg/m^2 to 29.9 kg/m^2 (overweight), and 30.0 kg/m^2 or more (obese). For children, their BMI for age was computed and then classified in one of four categories (i.e., underweight, healthy weight, overweight or obese) using the Center for Disease Control and Prevention pediatric growth charts (22).

IIB. Body Composition. Percentage body fat was estimated by bioelectrical impedance analysis (BIA)(23) using the Quantum X instrument (RJL Systems, MI). While procedures were the same for children and adults, Body Composition 2.1 software with a New Pediatric equation validated for children 3.9 to 19.3 years of age was used so that BIA could be computed for children.

IIC. Waist-to-Hip Ratio. Abdominal circumference (waist) was measured by having the subject stand comfortably with his or her weight evenly distributed on both feet, and the feet about 25-30 cm apart. The measurement was taken midway between the inferior margin of the last rib and the crest of the ileum, in a horizontal plane. Hip circumference was measured on top of the clothes. The subject stood with arms at his or her sides and feet together. The measurer sits at the side of the subject and placed the tape measure around the maximum extension of the buttocks in a horizontal plane. The waist and hip measurement were recorded to the nearest 0.1 cm.

IID. Blood Pressure. Blood pressure (BP) was measured using an automated cuff (DINAMAP®, GE Medical Systems, FL) in children and adults. Appropriate child-sized cuffs, based on the participant's arm size, were used for the children. The average of four blood pressure measurements was used for analyses. For mothers, systolic blood pressure (SBP) and diastolic blood pressure (DBP) was categorized using the CDC standards (www.cdc.gov/bloodpressure/what_you_can_do.htm) as follows: normal SBP<120 mmHg and DBP< 80mmHg, at risk SBP=120-139 mmHg and DBP=80-89 mmHg and high SPB>140 mmHg and DBP>90 mmHg (high). For children, blood pressure percentile (BP%) was calculated using BP tables developed by the National High-Blood Pressure Education Program Working Group on High-Blood Pressure in Children and Adolescents. Then, children were classified as normotensive: systolic or diastolic BP% <90th percentile, prehypertensive: BP% =90–94th percentile, or hypertensive BP% ≥95th percentile.

III. Mood, Food Preferences and Dietary Intake. Mothers were asked to complete standardized questionnaires relative to their and their child's food preferences, eating habits and mood states.

Children were also asked several questions directly to determine the reliability between maternal and child reports. The following provides a brief summary of each measure. The questionnaires described in section A-F were given to mothers whereas those in section F were asked to children.

IIIA. Dietary Intake (ASA). We used the ASA24, an automated, web-based, self-administered 24 hour dietary recall instrument developed by the National Cancer Institute and is publically available for scientific researchers to use online (http://riskfactor.cancer.gov/tools/instruments/asa24/). The application uses state-of-the-art computer technology, including a tutorial, graphic enhancements, and animated characters with audio-language cues to guide participants. Respondents selected foods consumed from a list based on the US Department of Agriculture's (USDA) survey database. Respondents found foods by browsing through food groups or typing and searching. Detailed questions about reported food, adapted from USDA's Automated Multiple Pass Method (AMPM) used in national dietary surveillance, collect information about preparation methods, additions to food, and portion size. Up to 8 sequentially-sized digital pictures aided respondents in reporting portions. Respondents were given multiple opportunities to edit their food lists. Outcome measures included calories, fat, protein, carbohydrates and sodium content in the subjects' diet. The final measure was the average of the 2 days of diet reports for each subject.

IIIB. Sweet Taste Questionnaire (STQ) is a12-item questionnaire which assesses sensitivity to the mood-altering effects of sweet foods and impaired control over eating sweet foods among adults (24).

IIIC. Food Craving Inventory (FCI) is a 28-item validated questionnaire designed to measure the frequency of overall food cravings as well as cravings for specific types of foods (25). Cravings for specific types of foods (i.e., an intense desire for a specific food that is difficult to resist) were measured by 4 independent subscales, each consisting of 4 to 8 items within the food category: high fats (i.e., fried chicken, gravy, sausage, hot dogs, fried fish, corn bread, bacon, steaks); sweets (i.e., cakes, cinnamon rolls, ice cream, cookies, chocolate, donuts, candy, brownies); carbohydrates/ starches (i.e., sandwich bread, rice, biscuits, pasta, pancakes/waffles, rolls, cereal, baked potato) and fast-food fats (i.e., pizza, French fries, hamburger, chips). Participants rated how often they experienced a craving for each of the foods over the past month using a 5-point Likert scale (1=never, 5=always/almost every day).

IIID. Fat Preference Questionnaire is a self-administered instrument to assess preferences for dietary fat (for adults)(26).

IIIE. Child Temperament and Food Neophobia Questionnaire: a 25-item scale completed by mothers that measures her child's temperament dimensions of emotionality, shyness, activity, sociability and negative reactivity to food (27). Neophobia is the fear of trying new foods and is common in children.

IIIF. Children Food Liking Score. In order to relate sweet and salt preferences, as assessed in the laboratory, with preferences in daily life, children were presented with several pairs of food and asked to indicate which they preferred; one of each pair tastes sweet (i.e., ice cream, cookies, pancakes, candy, dessert) whereas the other tastes salty and/or savory (i.e., chips, pretzels, bacon, Doritos, salty snacks) (28). They were asked whether they like to add sugar to their cereal or salt to their foods. Children were asked to name their favorite candy and to indicate whether they prefer chocolate milk or juice. From these data, sweet-liking and salt-liking scores were calculated.

IV. Mood and Personality Traits. Subjects completed a variety of questionnaires related to mood state so that we could further explore the relationship between genetics, smoking, taste preferences, and

mood states. The measures of depression and mood are included because of the known associations between affective state, smoking and heightened sweet preferences.

IVA. Profile of Mood States Questionnaire (POMS) is a 65-item questionnaire which measures six independent, transient mood states: tension (range 0 - 36), depression (range 0 - 60), anger (range 0 - 48), vigor (range 0 - 32), fatigue (range 0 - 28), and confusion (range 0 – 28); higher scores reflect higher intensity of the mood states (29).

IVB. Becks Depression Inventory (BDI-II) 21-item questionnaire which measures characteristic attitudes and symptoms of depression (30).

IVC. Restrained Eating Behavior is a 21-item questionnaire that measures conscious restriction of food intake in order to control body weight or to promote weight loss (31).

IVD. Pictorial Depression Scale (PDS) is a 23-item scale (32). In brief, each child was presented with pictures of two side-by-side identical figures with neutral facial expressions (i.e., the figures corresponded to the sex and race of the child). The experimenter reads aloud the pairs of statements to the child. Each statement of the pair was assigned to one of the figures and one of the statements reflected depressive symptomology. The child was then asked to point to the figure that was most like him or her. Responses reflecting depressive affect were given a score of two and the sum of these scores constitutes the total score for the measure; those that scored 10 or more were consider to be exhibiting depressive symptomology.

IVE. Nicotine Dependence (smokers only) and Family History of Addictions and Obesity. To evaluate nicotine self-administration behavior in smokers, subjects were given the Fagerstrom test of Nicotine dependence (33), the Michigan Nicotine Reinforcement questionnaire (34) (35). Smoking history was obtained by asking subjects the following questions: Do you smoke cigarettes? How old were you when you smoked your first cigarette? How old were you when you first started regular daily cigarette smoking? How many cigarettes are you currently smoking per day? When smoking the heaviest, how many cigarettes did you smoke per day? We also asked mothers to complete standardized questionnaires regarding their weight histories of themselves and their child, i.e., Weight and Lifestyle Inventory (36), family history of alcoholism (37) and family history of obesity (38) because there are studies showing a relationship between obesity, addiction and taste preferences (5, 39-41).

V. Genotyping for Salt and Sweet Taste, Addiction, and Hypertension. Genomic DNA was extracted following the directions of the manufacturer (Epicenter, Madison, WI). Alleles of the *TAS2R38* gene (accession no. NM_176817) were genotyped using real time PCR single nucleotide polymorphism (SNP) genotyping assays (*rs713598*, *rs1726866*, and *rs10246939*) with the Prism 7000, manufactured by Applied Biosystems (Foster City, CA). To that end, the resulting data were checked for appropriate segregation to detect genotyping or family history errors. Genes that may play a role in salty or sweet taste perception or nutrient selection were also genotyped, such as the putative salt (*SCNN1D*) and sweet receptor (*TAS1R3*). In addition, we also tested alleleic variants in two genes with a role in addition, one of the dopamine receptors (*DRD2*) and one of the opiate receptors (*OPRM1*). Variant sites were genotyped using the subject's DNA, and the correlation between nucleotide variants within these genes with the phenotypic responses of the subjects was examined.

VI. Statistical Analyses

Data were analyzed by ANOVA or general linear models using smoking status as fixed factor in all analyses (mother current smoker/mother never smoker). The groups were compared for

demographics, dietary habits and mood and sweet and salt preference. In some analyses, children were analyzed separately from mothers whereas in other analyses we combined the groups to determine whether there were age-related differences in sweet and salt preferences and how that related to dietary habits. Other analyses were undertaken to understand global effects of genotype on taste preferences and dietary intake, blood pressure, and if these differences are detected, genotype was incoporated into a general linear model to understand its contribution to group differences and to test for interactions with smoking status. Correlations between children and their mothers for sweet/salt taste perception and preferrence were assessed for heritablity.

Results

The data are presented below in the following order: a) description of subject population; b) smoking habits of mothers by self-report and subject characteristics (e.g., health characteristics) by maternal smoking status; c) direct measures of sweet and salty taste preference; bitter taste and other genes by smoking status.

A. Description of Study Population. We projected that fifty mother-child pairs would be tested on two days separated by one week during the project period (January 1st 2010 to Dec 31st 2010). Mothers were recruited from newspaper advertisements and initial interviews to screen subjects for the inclusion criteria were conducted over the telephone. Mothers were excluded from participation if they were diabetic, pregnant, lactating or taking any prescription medication, with the exception of birth control pills. Children were excluded if they were less than five or more than ten years of age. Using these criteria, this goal for subject recruitment and testing was met and exceeded. As seen in **Figure 1**, the total of subjects tested was 83 mothers and 109 children. The number of children tested was larger than the number of mothers because some mothers had more than one eligible child who particiapted in the experiment. Specifically, there were 20 sibling pairs and 3 sibling triads. Of those tested, we had a sample of 70 mothers and 98 children included in final analyses presented herein (see **Figure 1** for explanation; hereafter referred to as the final sample).

A1. Description of Final Sample**. Table 2** summarizes the demographic characteristics of this racially diverse sample of mothers and **Table 3** summarizes the demographic characterics of their children. On average, the mothers were 35.8 years old, had an average of 14 years of school (equivalent to two years of college). Some mothers reported an annual family income of less than $15,000 US dollars per year (20.3%) but the sample was also economically diverse, with 7.2% reporting an income of over $75,000 per year.

B. Smoking Habits of Mothers. Of the 70 mothers in the final sample, 20 were current smokers and 50 women were never smokers. A ‘current smoker’ was defined as someone who is currently smoking at least 2 cigarettes a day and has been smoking for more than one year whereas a ‘never smoker’ was defined as someone who never smoked in her lifetime.

As shown in **Table 4**, there was a range of nicotine dependence in the mothers who were current smokers, with some women scoring 13 on the Fagerstrom scale (indicating extreme nicotine dependence) whereas a few women reported relatively low measures of nicotine dependence, with the average rating being 8. The average rating corresponds to a high level of dependence, which is consistent with the inclusion criterion. Most women reported smoking from an early age; the average

age of the first cigarette was 16.2 years. As expected, the age of the first cigarette was about two years before smoking was established as an habitual behavior: the average age for starting smoking cigarettes routinely was 18.8 years.

Commensurate with the range of scores of nicotine dependence, there was a range in the number of cigarettes smoked per day, with a maximum number being 40. In general, people vary in how heavily they smoke over time. For instance, in periods or stress, people might smoke more or in response to health concerns (or practical limitations on smoking such as working in a non-smoking environment), they will smoke less. Therefore subjects were asked to report the number of cigarettes smoked during their period of heaviest smoking and these data suggest that on average, during this period, people smoked about 16.6 cigarettes per day. As expected from the current average age of the women (33 years old) and the average age which smoking began (16 to 17 years old), most women had been smoking for an average of 15 years, or most of their adult life (although about 55% of the women reported a smoking hiatus during pregnancy). When each mother's smoking history was tabulated, the average pack/years was 187.

C. Characteristics by maternal smoking status: demographcis, health and mood. In the first analysis, we stratified mothers and children by maternal smoking status and compared for age, education, income and obesity. The results of these measures for mothers are shown in **Table 5** and for children in **Table 6**.

Demographics. There were no differences in age or race/ethnicity by smoking status ($p>0.13$, data not shown). However there were statistical differences between current smokers and never-smokers in education and income. Current smokers completed fewer years of school and had a lower household income compared with women who were never-smokers. In addition, current smokers had higher body weights than never smokers ($p=0.02$). The average age of the children did not differ by maternal smoking status and both groups were (on average) 8 years old. There were differences in the distribution of boys and girls in the groups: there were more boys than girls from smoking mothers ($p=0.01$). However because of the lack of epidemiological evidence for a birth sex-bias and smoking, these differences are probably due to chance. Children did not differ in years of education, which is to be expected because each group was the same approximate age. The ethnicity/race of the children was similar to that of their mothers and did not differ based on maternal smoking status. The largest difference between children based on maternal smoking was for BMI category: 44% of children of current smokers, but only 22% of the children of never smokers, were overweight or obese.

Health. For health, we used additional methods: waist circumference, waist-to-hip ratio (WHR), percent body fat, basal metabolic rate and blood pressure (see above description; Section II). Smokers had significantly higher WHR than never smokers, even after controlling for BMI ($p=0.01$) and percentage of body fat ($p=0.01$). Further, current smokers had higher average SBP and DBP pressure ($p=0.02$ and $p=0.01$ respectively). Also current smoker mothers were more likely to be classified as having high blood pressure ($p=0.04$).

As shown in **Table 6,** the two groups of children did not differ in WHR, fat percentage of body weight, blood pressure and basal metabolic rate (all p's>0.3). However, children of smokers had larger waist circumferences than children of never smokers ($p=0.03$).

Cravings, Eating Habits and Mood States. Food craving patterns were significantly different between current smokers and never smokers, see **Table 7**. Current smokers craved sweets and fast food more frequently than never smokers. There was a tendency among current smokers to crave carbohydrates and high fat foods more frequently than never smokers. Also, current smokers craved foods in general more often than never smokers. There were no differences between the groups in the incidence of depression, tension, anger, vigor, fatigue or confusion.

As shown in **Table 8**, children of current smokers reported to like more salty foods than children of never smokers. Children of smokers were less shy, more social and more active and they tended to be less depressed than children on never smokers (**see Table 8**). However, there were no differences between the groups for maternal-child interaction as it relates to feeding (e.g., amount of restriction of the child's food intake; perceived pickiness of their children).

Dietary Habits. There were significant differences between current smoker and never smoker mothers in their dietary habits (see **Table 9**). Current smokers had a higher total energy intake but consumed the same number of food items in a day than did the never smoker mothers. Women who smoked also had higher daily intakes of sodium, potassium, phosphorous, iron, and cholesterol. A similar pattern was observed for the children (**Table 10**) but children of smokers ate diets higher in protein than nonsmoker.

Psychophysical Measures of Salt and Sucrose Preferences. When we evaluated the sweet or salt preferences based on the smoking status of the mother, we found no differences (**Table 11**). However, as described above, there are profound differences between women who smoke and women who never smoked in a variety of measures, including dietary habits, food cravings and health outcomes such as blood pressure. Therefore, we conducted a variety of analyses to probe the interaction between smoking, diet and taste preferences. We found that among smokers, the number of cigarettes the mother smoked at her heaviest smoking period, the greater the sodium content in their most preferred soup ($r(19df)=0.50$; $p=0.03$).

To test whether there was a relationship between salt preferences, salt intake and blood pressure, Pearson correlation coefficients were determined for children and mothers separately. For mothers, the intensity of salt most preferred in soup was positively correlated with systolic blood pressure ($r(68df)= 0.26$, $p = 0.03$). There was no relationship between salt preferences measured in the laboratory and sodium intake ($r(68df)= -0.13$, $p > .05$).

For children, there was a positive relationship between salt preferences and sodium intake ($r(87df)=0.26$, $p =0.01$). Furthermore, systolic blood pressure was positively related to sodium intake ($r(91df) = 0.28$, $p =0.01$) but not salt preferences ($r(85df)= 0.08$, $p >0 .05$). Although diastolic blood pressure was highly correlated to systolic blood pressure in both children ($r(92df)= 0.56$, $p <0 .01$) and adults ($r(68df) = 0.70$, $p < 0.01$), it was not related to either salt preferences or sodium intake in children or adults.

To determine for the first time whether sweet taste preferences are related to salt preferences, we conducted several correlational analyses. The level of sweet in water most preferred was significantly related to the level of salt most preferred in soup in adults ($r(69df)=0.36$; $=0.003$) and children (soup: $r(87df)=0.20$; $p=0.057$; crackers: ($r(87df)=0.28$; $p=0.007$).

We found consistency in the level of sweetness and saltiness preferred, independent of the food stimuli. Regardless of age, the level of sweetness preferred in water was related to the level of sweetness preferred in jellies (p=0.03). Likewise, the level of saltiness preferred in crackers was related to the level of saltiness preferred in soup (p=0.003).

Mothers vs Children: Age- and race- related effects. The most pronounced effects were the developmental difference in taste preferences (see **Figure 2**). Children preferred a more concentrated salt solution in the broth and a more concentrated sucrose solution and sweeter jelly than their mothers (**Table 12**). The preferences for salt were especially pronounced among African-American children when compared with Caucasian children (F(1,59)=6.09; p=0.02); African-American children preferred higher levels of salt in soup (2.73 vs. 1.96 %wt/v).

Genetics. Cigarettes can have a taste and it is possible that people with differing sensitivity and preference for a variety of tastes may be more or less likely to smoke. To test this hypothesis directly, we obtained samples of genomic DNA from saliva and genotyped the samples for alleles of the *TAS2R38* gene (**Table 13**). We also tested the hypothesis that alleles in sweet and salty perceptual genes (*TAS1R3* and *SCNN1D*) as well as the addiction genes (*DRD2* and *OPRM1*) might affect the preference for sweet and salty tastes and taste-related behavior in children and mothers.

Women were grouped by their smoking status to determine whether smokers were more likely to be insensitive to some bitter compounds. The results indicate no difference in the frequency of the nontaster haplotypes between never-smokers and current smokers (**Table 13**). These data demonstrate no difference between current and never-smokers in the frequency of the alleles associated with increased bitter, sweet or salty perception. We also tested genes associated with craving and addiction: the dopamine receptor gene *DRD2* and the opioid receptor gene, *OPRM1* but found no differences between smoking and never smoking groups in allele frequency (**Table 13**). To summarize the gene results, there were no differences in allele frequencies between smoking groups for genes relevant to taste behavior (*TAS2R38*, *TAS1R3* or *SCNN1D*), or addiction (*DRD2* and *OPRM1*).

Three of the genes analyzed in the current study are related to taste behavior and have been linked to perception of sweet (*TAS2R38* and *TAS1R3*) and salty (*SCNN1D*) compounds. We wanted to see if these associations were also present in the current study in children or their mothers.

All three genes showed a correlation between genotype and sucrose preference in water (**Table 14**). Associations were seen between *TAS2R38*, *TAS1R3* and *SCNN1D* and sucrose preference in mothers, but in children the only effect was seen in relation to *TAS2R38*.

When examining salt preference in crackers, all three genes showed an association, however the effects were different in mothers and their children (**Table 15**). Mothers who were homozygous for the most common form of the *SCNN1D* genotype were much more likely to prefer lower salt crackers than heterozygous mothers or those homozygous for the rare genotype. In children, no association was seen with *SCNN1D* genotypes, however there were associations between salt preference in crackers and both *TAS1R3* and *TAS2R38*. Alleles of *TAS1R3* also showed a significant association with salt in soup in children (data not shown).

In summary, there are associations between sweet and salt preference and genes relevant for taste behavior (*TAS2R38*, *TAS1R3* or *SCNN1D*) in both mothers and children. We saw more significant differences in allele frequencies between mothers who prefered high vs. low sucrose concentrations than we did for children. Conversely, we saw more differences for children who prefered high vs. low salt concentrations than we did for mothers.

Figure 1. Flow diagram of subjects tested in the *Relationship between Parental Smoking, Dietary Habits, Salt and Sweet Preferences and Blood Pressure among Children Study* at the Monell Center: 1/1/2010-12/31/2010

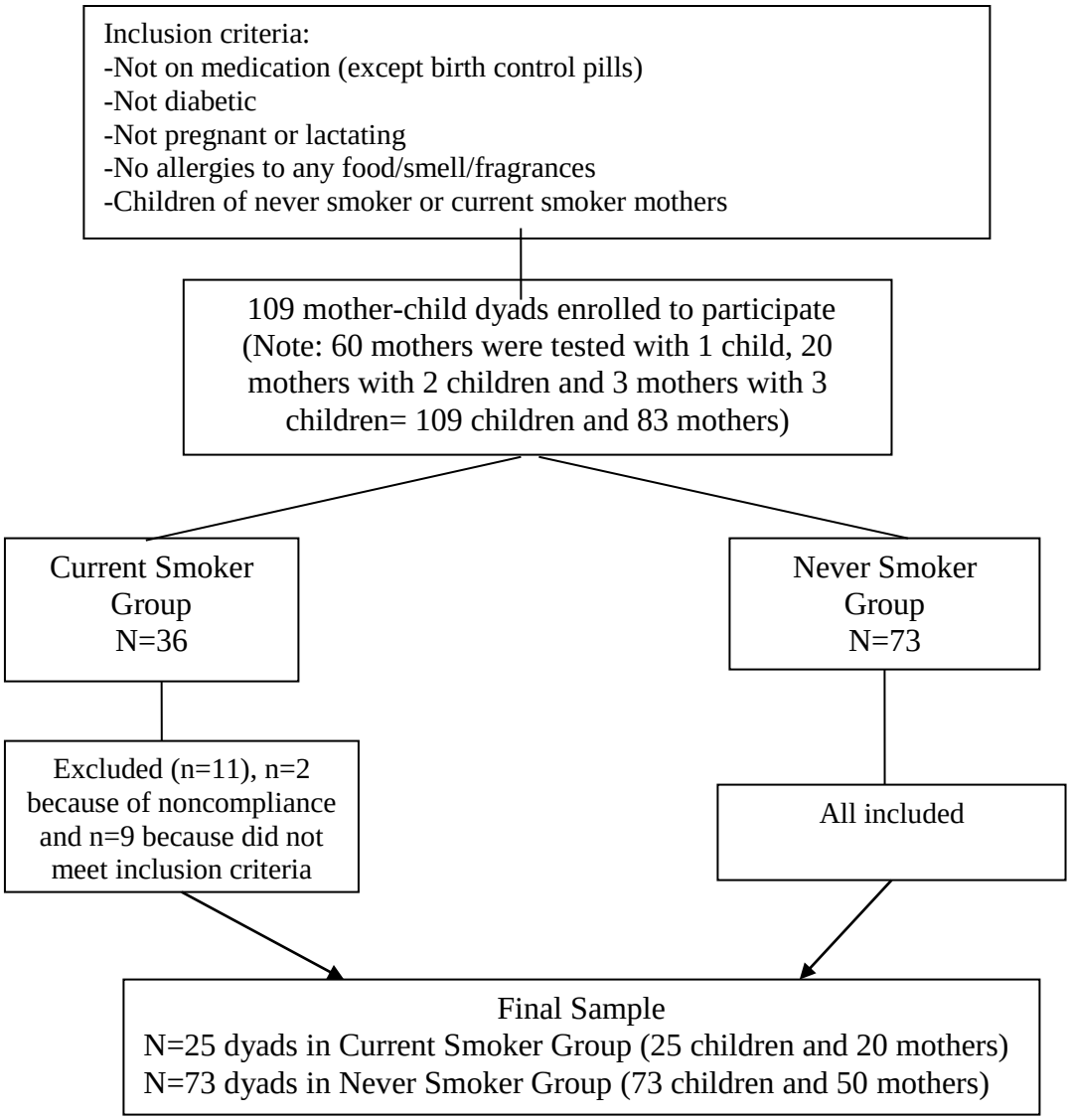

Figure 2. Age-related differences in sweet (red bars; left axis) and salt (black bars; right axis) preferences; * indicates significant differences at p<0.05 level.

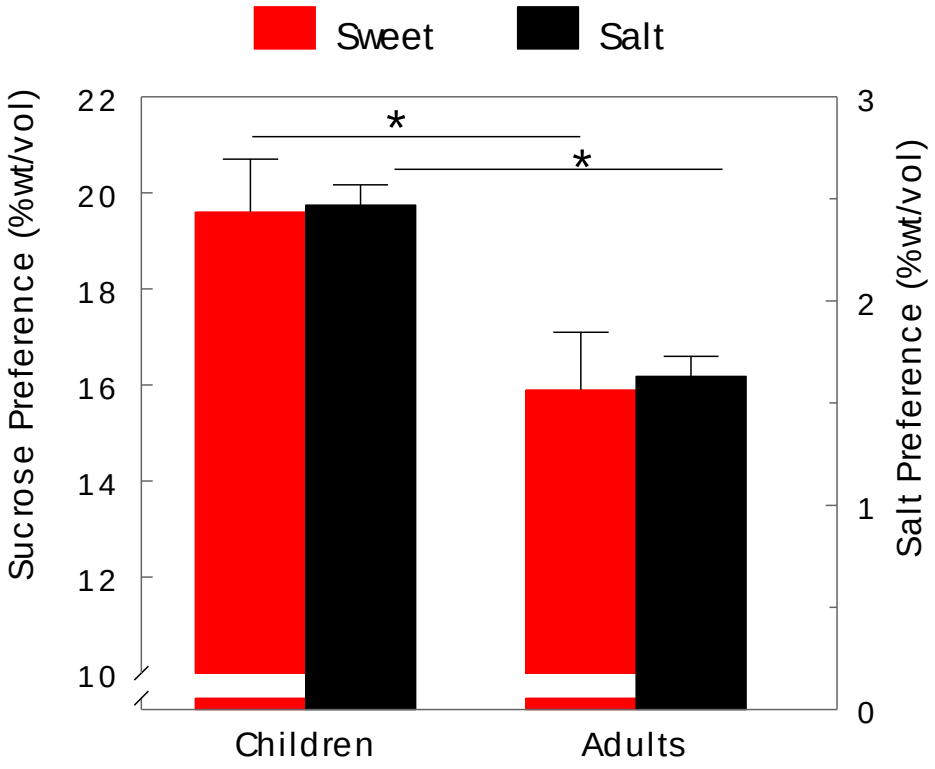

Table 1. Schedule of Events for Days A and B	
Day A	**Day B**
Carbon Monoxide Levels	Carbon Monoxide Levels
Cheek Swab/Saliva (DNA) Collection	Blood Pressure Readings
Blood Pressure Readings	Anthropometry
Anthropometry	Salt and Sweet Liking in Foods
Salt and Sucrose in Solutions Preference Test	24-hour dietary recall
Questionnaires	Questionnaires
24-hour dietary recall	24-hour urine collection

Table 2: Demographic characteristics of mothers	
Number of mothers	N=70
Age , mean (SD)	35.8 (8.2)
Race % African American % Caucasian % Hispanic % Asian % Other/More than one race)	 54.3%,n=38 30.0%,n=21 5.7%,n=4 1.4%,n=1 8.6%,n=6
Yrs of school, Mean (SD)	14.0(2.1)
Family yearly income %<$15,000 %$15,000-$35,000 %$35,000-$75,000 %>$75,000	 20.3%,n=14 39.1%,n,=27 33.3%,n=23 7.2%,n=5
Smoking status: Never-smokers Current-smoker	 71.4%,n=50 28.6%,n=20
BMI, Mean (SD)	28.4(6.8)
Body Weight category: Underweight Normal-weight Overweight Obese	 2.9 %,n=2 34.3%,n=24 25.7%, n=18 37.1%, n=26

SD=standard deviation. Yrs=Years. Family yearly income is reported in US dollars ($). Smoking status was determined through questionnaire and confirmed with empirical measures (breath carbon monoxide concentrations). School refers to formal education, high school, community or four-year college. BMI=body mass index, a measure of obesity. BMI=kg/m^2 where kg is weight in kilograms and m is height measured in meters. For women, BMI was categorized by standards set by the Center for Disease Control (CDC): 18.5 kg/m^2 to 24.9 kg/m^2 (normal weight), 25.0 kg/m^2 to 29.9 kg/m^2 (overweight), and 30.0 kg/m^2 or more (obese).

Table 3: Demographic characteristics of children	
Number of children	N=98
Age, mean (SD)	7.6(1.9)
Race % African American % Caucasian % Hispanic % Asian % Other/More than One race)	 45.9%, n=48 25.5%, n=25 10.2%, n=2 2.0%, n=2 16.3%, n=16
Sex % Female % Male	 61.2 %, n=61 37.8%, n=37
Yrs of school, Mean (SD)	2.6(1.9)
By smoking status of the mother % Never-smokers % Current-smokers	 74.5%, n=73 25.5%, n=25
Smoking Status of Child's Biological Father Never-smoker Current smoker Former Smoker Unknown	 49%, n=48 36.7%, n=36 10.2%, n=10 4.1%, n=4
Body Weight category % Underweight % Healthy-weight % Overweight % Obese	 6.1 % , n=6 66.3%, n=65 18.4%, n=18 9.2%, n=9

SD=standard deviation. Yrs=Years. Smoking status was determined through questionnaire of the child's mother which was confirmed with empirical measures (breath carbon monoxide concentrations). BMI was computed for each child and then the child was classified into one of four weight categories (i.e., underweight, healthy weight, overweight or obese) using the Center for Disease Control and Prevention pediatric growth charts.

Table 4: Nicotine dependence and smoking habits of mothers who are current smokers		
Measures	Mean	Range
Fagerstrom (measure of nicotine dependence) Score: 0-6: low to moderate nicotine dependence 7 and up : high nicotine dependence	8 (2.4)	5-13
Age of first cigarette (yrs)	16.2 (4.9)	10-33
Age started regular smoking (yrs)	18.8 (5.2)	11-33
# Cigarettes/day	10.8 (9.4)	2-40
#Cigarettes/day (when smoking the heaviest)	16.6 (9.1)	6-40
Years smoking	14.7 (6.2)	3.7-24.4
Dose (pack/years) Max Dose(pack/years)	187 (114.8) 227.5 (120.5)	31.7-456.6 37.3-456.6
Did you smoke during pregnancy with child who was tested?	45% reported smoking during pregnancy	

Yrs=years; Mean values are followed by standard deviations, in parenthesis.

Table 5: Subject Characteristics of mothers when grouped by smoking status			
Measures	Never Smoker Group	Current Smoker Group	p-value
N=68	N=50	N=20	
Age (Mean)(SD)	36.7(8.3)	33.4(7.7)	0.13
Years of school	14.6(2.0)	12.8(1.8)	**0.00**
Yearly income <$15,000 $15,000-35,000 $35,000-75,000 >$75,000	 16.33%,n=8 32.65%,n=16 40.82%,n=20 10.2%,n=5	 30%,n=6 55%,n=11 15%,n=3 0%,n=0	**0.05**
BMI (kg/m$^{2)}$	27.2(6.2)	31.4(7.4)	**0.02**
Waist-to-hip ratio, Mean (SD)	0.78(0.06)	0.84(0.07)	**0.00**
Waist circumference (cm)	84.3(12.6)	95.7(14)	**0.00**
% body fat	35.3(8.8)	40.1(7.0)	**0.03**
BMR (calories)	1465(175)	1604(200)	**0.01**
Systolic Pressure, mmHg (average of 4 measurements)	122.5(13.0)	133.2(24.2)	**0.02**
Diastolic Pressure, mmHg (average of 4 measurements)	73.8(9.3)	80.4(11.5)	**0.01**
Systolic Pressure Category Normal <120 mmHg At risk 120-139 mmHg High >140 mmHg Diastolic Pressure Category Normal< 80 mmHg At risk 80-89 mmHg High >90 mmHg	 44%, n=22 48%, n=24 8%, n=4 78%, n=39 16%, n=8 6%, n=3	 30%, n=6 35%, n=7 35%, n=7 55%, n=11 25%, n=5 20%, n=4	**0.02** 0.10
Blood Pressure Category **Normal**, SP<120 mm and DBP Hg< 80 mmHg **At Risk**, SBP=120-139 mmHg and DBP=80-89 and mmHg **High**, >140 mmHg and >90 mmHg	 42%, n=21 48%, n=24 10%, n=5	 30%, n=6 35%, n=7 35%, n=7	**0.04**

SD=standard deviation. For quantitative traits, statistics were conducted using an ANOVA with smoking status as the fixed factor and the demographic value as the dependent variable, e.g., years of school. Post-hoc tests were conducted to assign group differences. For categorical data, non-parametric tests like Chi-Square were conducted. Criterion p-value is $p \leq 0.05$. BMR=basal metabolic rate as measured in calories.
SBP=Systolic Blood Pressure, DBP=Diastolic Blood Pressure

Table 6: Characteristic of children grouped by smoking status of the mother			
	Smoking status of the mother		
	Never Smoker	Current Smoker	p-value
N=98	73	25	
Age (Mean±SD)	7.6 (1.9)	7.7 (1.9)	0.70
Sex (%) Female Male	 69.9%, n=51 30.1%, n=22	 40%, n=10 60%, n=15	**0.01**
Years of school	2.6 (2.0)	2.5 (1.7)	0.85
Race (%) African American Caucasian Other(more than one race) Hispanic Asian	 41.1%, n=30 31.5%, n=23 11%, n=8 13.7%, n=10 2.7%, n=2	 60%, n=15 8%, n=2 32%, n=8 0%, n=0 0%, n=0	**0.01**
BMI category % Underweight % Healthy Weight % Overweight % Obese	 8.2%, n=6 69.9%, n=51 12.3%, n=9 9.6%, n=7	 0.0%, n=0 56.0%, n=14 36.0%, n=9 8%, n=2	**0.04**
Waist-to-hip ratio	0.83 (0.04)	0.83 (0.05)	0.72
Waist circumference (cm)	57.7 (7.7)	62.0 (10.5)	**0.03**
% fat in body	24.5 (8.9)	25.1 (10.1)	0.79
BMR	1096 (128)	1133 (192)	0.29
Systolic Pressure (average of 4 measurements), mmHg	110.4 (8.8)	112.7 (10.2)	0.29
Diastolic Pressure (average of 4 measurements), mm Hg	65.4 (4.3)	64.9 (4.5)	0.64
Systolic Pressure Category By Percentile <90% 90-94% >95%	 55.6%, n=40 19.4%, n=14 25%, n=18	 36%, n=9 20%, n=5 44%, n=11	0.16
Diastolic Pressure Category By Percentile <90 % 90-94%	 87.5%, n=63 12.5%, n=9	 100%, n=25 0%, n=0	0.06
Blood Pressure Category Normal: SBP and DBP <90% Prehypertensive: SBP and DBP=90-94% Hypertensive: SBP and DBP>95%	 50%, n=36 25%, n=18 25%, n=18	 36%, n=9 20%, n=5 44%, n=11	0.20

SD=standard deviation. For quantitative traits, statistics were conducted using an ANOVA with smoking status as the fixed factor and the demographic value as the dependent variable, e.g., years of school. Post-hoc tests were conducted to assign group differences. For categorical data, non-parametric tests like Chi-Square were conducted. Criterion p-value is $p \leq 0.05$. BMR=basal metabolic rate as measured in calories.

Table 7: Fat and sweet liking, Food Craving, Dietary Restriction and Mood States of mothers grouped by smoking status			
	Never Smoker	Smokers	p
N=70	50	20	
Fat preference questionnaire -% of high fat foods reported to taste better. -%of high fat foods reported eaten more often.	65.4 (16.13) 55.3 (20.7)	68.9(19.31) 61.1(24.7)	0.43 0.32
Restrained Eating Behavior	9.6 (5.6)	8.5 (8.5)	0.46
Sweet Questionnaire Q1: Sensitivity to mood altering effects of sweet food Q2: Impaired control over eating sweets	18.9 (8.2) 9.1 (7.8)	20.4 (9.5) 8.1 (7.0)	0.52 0.61
Food Craving Inventory High-fats Carbohydrates Sweets Fast-Food-fats Average Total Cravings	 2.36 (0.92) 2.42 (0.74) 2.38 (0.73) 2.65 (0.84) 2.45 (0.61)	 2.81 (1.1) 2.83 (0.99) 2.88 (1.07) 3.23 (0.92) 2.94 (0.87)	 0.08 0.06 **0.03** **0.01** **0.01**
POMS–covaried by age Tension Depression Anger Vigor Fatigue Confusion Total POMS	 7.40 (5.84) 4.98 (6.84) 4.90 (6.19) 15.98 (5.75) 5.66 (3.96) 5.26 (3.64) 12.22 (24.77)	 8.05 (4.89) 6.05 (4.98) 7.26 (6.62) 16.47 (8.83) 6.84 (4.92) 5.94 (4.05) 17.68 (23.13)	 0.82 0.70 0.28 0.65 0.37 0.60 0.58
Becks Depression Inventory % mild to moderate depression (Score of 10 or more)	16%, n=8	30%, n=6	0.19

SD=standard deviation. POMS; Profile of mood states. Measures which are statistically different between the groups are underlined for emphasis.

Table 8: Child's feeding behavior, temperament and sweet and salty food liking scores; children were group according to smoking status of the mothers			
	Smoking Status of the Mothers		
	Never Smokers	Current-Smokers	p
Child Feeding Questionnaire (1-5)			
	4.37 (0.66)	4.57 (0.62)	0.16
Overall Resp. Feeding the	4.06 (0.94)	4.18 (0.85)	0.65
Child	2.87 (1.15)	2.88 (0.90)	1
Monitoring	3.22 (0.80)	3.33 (0.19)	0.65
Pressure to Eat	3.54 (0.69)	3.80 (0.76)	0.12
Restriction	2.96 (0.51)	3.08 (0.57)	0.32
Perceived Parent's Weight	1.89 (1.22)	2.16 (1.53)	0.38
Perceived Child's Weight	2.45 (1.16)	2.06 (1.30)	0.30
Weight Concerns for Child	1.56 (0.76)	1.48 (0.60)	0.71
Pickiness			
Time to Eat Healthy			
Child Temperament and Eating Behavior (higher numbers represent more of the trait)			
Shyness	2.70 (0.79)	2.24 (0.82)	**0.02**
Emotion	2.76 (1.0)	3.19 (1.0)	0.07
Sociability	3.33 (0.64)	3.82 (0.64)	**0.00**
Negative Reaction to Food	2.85 (0.90)	3.08 (1.16)	0.34
Activity	3.66 (0.77)	4.07 (0.61)	**0.02**
Food Neophobia	2.80 (0.90)	2.67 (1.08)	0.56
Pictorial Depression Scale			
% depressed, score >9	40.58%, N=28	20%, N=5	0.06
Children's Sweet Liking Score (score 0-6)	3.66 (1.47)	3.38 (1.31)	0.40
Children's Salt Liking Score (score (0-6)	2.13 (1.52)	2.83 (1.52)	**0.05**

SD=standard deviation. Measures which are statistically different between the groups are underlined for emphasis.

Table 9: Percentage of energy from macronutrients and daily sodium and micronutrient intake for mothers grouped by smoking status			
	Never Smoker N=50	Smokers N=20	p
Total Energy, Kilocalories (SD)	1708(542)	2349(912)	**0.00**
Number of food Items, mean (SD)	14.4 (4.2)	14.7(3.8)	0.92
Protein,% (SD)	15.0 (3.6)	16.0 (3.5)	0.33
Total Fat, % (SD)	35.6 (7.3)	36.4 (5.6)	0.65
Saturated Fat, % (SD)	11.8 (3.9)	12.3 (2.1)	0.56
Monounsaturated Fat, % (SD)	13.0 (2.7)	13.4 (2.7)	0.61
Polyunsaturated Fat, % (SD)	7.4 (2.4)	7.0 (1.4)	0.59
Carbohydrates, % (SD)	50.4 (10.0)	47.7 (7.3)	0.27
Simple Sugars, % (SD)	24.8 (9.1)	25.8 (8.7)	0.68
Cholesterol(mg/day)	250.8(163.0)	389.3(197.6)	**0.00**
Fiber (g/day)	11.2(5.4)	11.1(4.3)	0.95
Calcium (mg/day)	693.6(361.1)	761.1(356.5)	0.48
Caffeine (mg/day)	105.9(140.8)	100.9(104.5)	0.89
Iron (mg/day)	11.1(4.3)	14.4(5.7)	**0.01**
Magnesium(mg/day)	205.2(78.8)	234.9(84.7)	0.17
Phosphorous (mg/day)	1007.4(395.4)	1275.1(513.0)	**0.02**
Potassium(mg/day)	2021.2(818.6)	2434.7(911.2)	0.07
Sodium (mg/day)	2991.6(1186.2)	3805.7(1964.3)	**0.04**

SD=standard deviation. For quantitative traits, statistics were conducted using an ANOVA with smoking status as the fixed factor and % of kilocalories from macronutrient as the dependent variable, e.g., % of calories from protein. Measures which are statistically different between the groups are by smoking status are underlined for emphasis.

Table 10 : Percentage of energy from macronutrients and micronutrients for children grouped by smoking status			
	Never Smoker	Smokers	p
	72	25	
Total Energy, Kilocalories (SD)	1766(555)	2109(560)	**0.01**
Number of food Items, mean (SD)	14.5(3.2)	14.3(3.2)	0.76
Protein,% (SD)	13.3 (3.0)	15.4 (3.5)	**0.01**
Total Fat, % (SD)	35.0 (5.6)	34.3 (5.7)	0.62
Saturated Fat, % (SD)	11.8 (2.9)	12.1 (2.0)	0.62
Monounsaturated Fat, % (SD)	13.4 (2.9)	13.2 (2.9)	0.78
Polyunsaturated Fat, % (SD)	7.1 (2.4)	6.2 (1.9)	0.10
Carbohydrates, % (SD)	52.8 (7.0)	51.0 (8.1)	0.28
Simple Sugar, % (SD)	26.5 (7.1)	26.7 (9.7)	0.93
Cholesterol(mg/day)	194.3(111.7)	276.6(133.9)	**0.00**
Fiber (g/day)	10.5(3.6)	11.3(4.6)	0.41
Calcium (mg/day)	762.0(359.3)	873.5(337.6)	0.18
Caffeine (mg/day)	15.6(24.4)	13.3(14.3)	0.65
Iron (mg/day)	11.5(4.4)	14.1(5.3)	**0.02**
Magnesium(mg/day)	184.0(52.8)	220.3(87.1)	**0.02**
Phosphorous (mg/day)	1011.2(343.1)	1218.7(411.9)	**0.02**
Potassium(mg/day)	1905.4(649.7)	2203.8(694.1)	**0.05**
Sodium (mg/day)	2794.5(968.7)	3430.3(1102.0)	**0.01**

Mean(SD) For quantitative traits, statistics were conducted using an ANOVA with smoking status as the fixed factor and % of kilocalories from macronutrient as the dependent variable, e.g., % of kilocalories from protein. Criterion p-value is $p \leq 0.05$.

Table 11: Laboratory measures of sweet and salty preference of mothers and children grouped by smoking status (GeoMean)(SD)(SE)			
	Smoking Status of the mother		
	Never Smoker	Current Smokers	p
Psychophysical Test Measures:	**Mothers**		
Sucrose in water preference (%wt/vol)	16.1% (11.1)(1.5)	15.5 (10.7)(2.5)	0.85
Sugar in Grape Jelly preference (% sugar wt/wt)	48.4 (11.5)(1.63)	46.5 (11.4)(2.5)	0.53
Salt in Vegetable Broth (%wtNaCl/vol)	1.60 (0.73)(0.11)	1.70 (0.78)(0.17)	0.64
Salt in Cracker (% wt/wt NaCl)	3.5 (1.83) (0.26)	3.2 (2.13)(0.48)	0.65
	Children		
Sucrose in water preference (%wt/vol)	18.85 (9.9)(1.2)	21.6 (9.6)(2.0)	0.24
Sugar in Grape Jelly preference (%wt/wt)	53.7 (12.8)(1.6)	52.2 (12.4)(2.6)	0.63
Salt in Vegetable Broth (%wt/vol)	2.42 (1.23)(0.16)	2.59 (1.25)(0.26)	0.58
Salt in Cracker (% wt/wt NaCl)	3.38 (1.84)(0.22)	3.81 (1.89)(0.38)	0.32

GeoMean=geometric mean; SD=standard deviation; SE=standard error of the mean

Table 12: Laboratory measures of sweet and salty preference children versus mothers (GeoMean)(SD)(SE)			
	Children	Mothers	p-value
Taste Test	N=98	N=70	
Sucrose in water preference (% wt/v)	19.6 (9.8)(1.1)	15.9 (10.9)(1.2)	**0.03**
Sugar in Grape Jelly preference (% sugar wt/wt)	53.30 (12.65)(1.27)	47.86 (11.41)(1.45)	**0.01**
Salt in Vegetable Broth (%wtNaCl/vol)	2.47 (1.23)(0.11)	1.63 (0.74)(0.13)	**0.00**

GeoMean=geometric mean; SD=standard deviation; SE=standard error of the mean

Table 13: Allele frequency of mothers by their smoking status				
Gene/Genotype	**Never Smoker (NS)**	**%NS**	**Current Smoker (CS)**	**%CS**
***TAS2R38* (Bitter Receptor)**				
Nontaster	12	24	6	32
Taster	37	76	13	68
***SCNN1D* (Salt Receptor)**				
A/A	12	26	5	38
A/B	12	26	5	38
B/B	22	48	3	23
***TAS1R3* -1266 (Sweet Receptor)**				
A/A	4	27	3	30
A/B	8	53	4	40
B/B	3	20	3	30
***TAS1R3* -1572 (Sweet Receptor)**				
A/A	5	33	4	40
A/B	9	60	4	40
B/B	1	7	2	20
***DRD2* (Dopamine Receptor)**				
A/A	2	13	3	30
A/B	10	67	5	50
B/B	3	20	2	20
***OPRM1* (Opiate Receptor)**				
A/A	14	93	8	80
A/B	1	7	2	20
B/B	0	0	0	0

Table 14: Sweet preferences of mothers and children by genotype				
Gene/Genotype	**Low Sucrose (3-6%)**	**%LS**	**High Sucrose (12-36%)**	**%HS**
***TAS2R38* (Bitter Receptor)**				
Mothers, p=0.04				
Nontaster	12	39	6	16
Taster	19	61	31	84
***SCNN1D* (Salt Receptor)**				
Mothers, p=0.04				
A/A	4	14	13	43
A/B	10	34	7	23
B/B	15	52	10	33
***TAS1R3* -1572 (Sweet Receptor)**				
Mothers, p=0.03				
A/A	9	43	0	0
A/B	7	51	6	75
B/B	1	6	2	25
	Low Sucrose (3-12%)	**%LS**	**High Sucrose (24-36%)**	**%HS**
***TAS2R38* (Bitter Receptor)**				
Children, p=0.02				
Nontaster	12	29	4	9
Taster	29	71	40	91

Table 15: Salt in cracker preferences of mothers and children by genotype				
Gene/Genotype	**Low/Med (0.8, 2.9%)**	**% LM**	**High (6.12%)**	**% H**
***SCNN1D* (Salt Receptor)**				
Mothers, p=0.02				
A/A	16	36	1	7
A/B	9	20	8	53
B/B	19	43	6	40
***TAS2R38* (Bitter Receptor)**				
Children, p=0.01				
Nontaster (AVI/AVI)	17	27	1	4
Taster (other genotypes)	47	73	25	96
***TAS1R3* -1266 (Sweet Receptor)**				
Children, p=0.03				
A/A	9	36	2	25
A/B	14	56	2	25
B/B	2	8	4	50
***TAS1R3* -1572 (Sweet Receptor)**				
Children, p=0.04				
A/A	11	44	3	38
A/B	13	52	2	25
B/B	1	4	3	38

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18. Extent of Clinical Activities Initiated and Completed. Items 18(A) and 18(B) should be completed for all research projects. If the project was restricted to secondary analysis of clinical data or data analysis of clinical research, then responses to 18(A) and 18(B) should be "No."

18(A) Did you initiate a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
__X___No

18(B) Did you complete a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
___X__No

If "Yes" to either 18(A) or 18(B), items 18(C) – (F) must also be completed. (Do NOT complete 18(C-F) if 18(A) and 18(B) are both "No.")

18(C) How many hospital and health care professionals were involved in the research project?

_____Number of hospital and health care professionals involved in the research project

18(D) How many subjects were included in the study compared to targeted goals?

_____Number of subjects originally targeted to be included in the study
_____Number of subjects enrolled in the study

18(E) How many subjects were enrolled in the study by gender, ethnicity and race?

Gender:
_____Males
_____Females
_____Unknown

Ethnicity:
_____Latinos or Hispanics
_____Not Latinos or Hispanics
_____Unknown

Race:
_____American Indian or Alaska Native
_____Asian
_____Blacks or African American
_____Native Hawaiian or Other Pacific Islander
_____White
_____Other, specify:
_____Unknown

18(F) Where was the research study conducted? (List the county where the research study was conducted. If the treatment, prevention and diagnostic tests were offered in more than one county, list all of the counties where the research study was conducted.)

19. Human Embryonic Stem Cell Research. Item 19(A) should be completed for all research projects. If the research project involved human embryonic stem cells, items 19(B) and 19(C) must also be completed.

19(A) Did this project involve, in any capacity, human embryonic stem cells?

_____Yes
___X__ No

19(B) Were these stem cell lines NIH-approved lines that were derived outside of Pennsylvania?

_____Yes

_____ No

19(C) Please describe how this project involved human embryonic stem cells:

20. Articles Submitted to Peer-Reviewed Publications.

20(A) Identify all publications that resulted from the research performed during the funding period and that have been submitted to peer-reviewed publications. Do not list journal abstracts or presentations at professional meetings; abstract and meeting presentations should be listed at the end of item 17. **Include only those publications that acknowledge the Pennsylvania Department of Health as a funding source** (as required in the grant agreement). List the title of the journal article, the authors, the name of the peer-reviewed publication, the month and year when it was submitted, and the status of publication (submitted for publication, accepted for publication or published.). Submit an electronic copy of each publication, listed in the table, in a PDF version 5.0.5 format, 1,200 dpi. Filenames for each publication should include the number of the research project, the last name of the PI, the number of the publication and an abbreviated research project title. For example, if you submit two publications for PI Smith for the "Cognition and MRI in Older Adults" research project (Project 1), and two publications for PI Zhang for the "Lung Cancer" research project (Project 3), the filenames should be:

Project 1 – Smith – Publication 1 – Cognition and MRI
Project 1 – Smith – Publication 2 – Cognition and MRI
Project 3 – Zhang – Publication 1 – Lung Cancer
Project 3 – Zhang – Publication 2 – Lung Cancer

If the publication is not available electronically, provide 5 paper copies of the publication.

Note: The grant agreement requires that recipients acknowledge the Pennsylvania Department of Health funding in all publications. Please ensure that all publications listed acknowledge the Department of Health funding. If a publication does not acknowledge the funding from the Commonwealth, do not list the publication.

Title of Journal Article:	Authors:	Name of Peer-reviewed Publication:	Month and Year Submitted:	Publication Status (check appropriate box below):
1.None				☐Submitted ☐Accepted ☐Published

20(B) Based on this project, are you planning to submit articles to peer-reviewed publications in the future?

Yes____X_____ No_____

If yes, please describe your plans:

The following abstract was accepted for submission at the 2011 Annual Meeting for the Association of Chemoreception Sciences; a manuscript describing these findings is in preparation.

How the sensory world of children differs from adults: sweets, salt and fat

Stacie S. Miller, PhD, Susanna Finkbeiner, Aleida Silva-Garcia, Danielle R. Reed, PhD and Julie A. Mennella, PhD
Monell Chemical Senses Center, Philadelphia, PA 19104

Although modernization and industrialization of the food supply has produced many benefits, unanticipated consequences from eating diets rich in sugars, salt and fats have become increasingly commonplace. Thus, health organizations worldwide recommend that both adults and children limit their intake of salt and simple sugars. This recommendation may be particularly difficult for pediatric populations since findings from basic research strongly suggest that children prefer more intense sweet or salty tastes than adults. However, there is a paucity of information on the relationship between sweet and salty taste preferences during childhood and adulthood and whether these preferences relate, if at all, to the liking of creaminess in foods. To this end, we phenotyped 162, 5- to 10-year-old children and their mothers for the intensity of sucrose most preferred in water; of those 162 dyads, 84 were also phenotyped for preferences for creaminess and sweetness in pudding, and 99 were phenotyped for preferences for salt in a soup matrix and crackers and sweetness in jelly. Children preferred significantly higher levels of sweet and salt, but lower levels of creaminess, than did their mothers, thus suggesting that the age-related differences in taste preferences were not due solely to children preferring more intense stimuli. For both children and adults, the most preferred level of sucrose in water was significantly correlated with the most preferred level of salt in soup. We also found that the intensity of sweetness preferred in water was related to preferences for sweetness in pudding and jelly, and the intensity of saltiness preferred in soup was related to preference for saltiness in crackers, thus suggesting real-world significance of our measurements.

The project described was funded, in part, by a grant from the Pennsylvania Department of Health. The Pennsylvania Department of Health specifically disclaims responsibility for any analyses, interpretations, or conclusions.

21. Changes in Outcome, Impact and Effectiveness Attributable to the Research Project. Describe the outcome, impact, and effectiveness of the research project by summarizing its impact on the incidence of disease, death from disease, stage of disease at time of diagnosis, or other relevant measures of outcome, impact or effectiveness of the research project. If there were no changes, insert "None"; do not use "Not applicable." Responses must be single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

The outcome of this research is to understand the relationship between smoking, taste preferences and health in both the mother who smokes and her child. The information gleaned will be helpful for caregivers such as nutritionists, physicians, nurses and psychologists who treat nicotine addiction

and craving. The main findings of the present study are that women who smoke crave fast foods and sweet foods more than nonsmokers and their diets are higher in sodium. Dietary sodium intake was related to various health measures (e.g., blood pressure). Similar relationships were found between dietary salt intake and blood pressure in children whose preferences for salt and sweet were elevated when compared with adults.

22. Major Discoveries, New Drugs, and New Approaches for Prevention Diagnosis and Treatment. Describe major discoveries, new drugs, and new approaches for prevention, diagnosis and treatment that are attributable to the completed research project. If there were no major discoveries, drugs or approaches, insert "None"; do not use "Not applicable." Responses must be single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

The major discoveries of this project were threefold. First, children have more elevated sweet and salt preferences than adults and there is a relationship between the preferences for these basic tastes in both children and adults. Second, the diets of smokers and their children are rich in sodium and dietary intake of sodium is related to blood pressure in both children and adults. Third, variation in taste receptor genes has a measureable effect on taste perception and preferences. These findings raise an interesting further question, which is whether smoking changes preferences for high-salt foods or whether people with food preferences are more likely to start smoking. There is a hypothesis that smoking is one of a constellation of behaviors on an addiction spectrum and that people who have the genetic and biological predisposition to smoke may also have food preferences for less wholesome food.

23. Inventions, Patents and Commercial Development Opportunities.

23(A) Were any inventions, which may be patentable or otherwise protectable under Title 35 of the United States Code, conceived or first actually reduced to practice in the performance of work under this health research grant? Yes_____ No___x_____

If "Yes" to 23(A), complete items a – g below for each invention. (Do NOT complete items a - g if 23(A) is "No.")

a. Title of Invention:

b. Name of Inventor(s):

c. Technical Description of Invention (describe nature, purpose, operation and physical, chemical, biological or electrical characteristics of the invention):

d. Was a patent filed for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No____

If yes, indicate date patent was filed:

e. Was a patent issued for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No____
If yes, indicate number of patent, title and date issued:
Patent number:
Title of patent:
Date issued:

f. Were any licenses granted for the patent obtained as a result of work performed under this health research grant? Yes_____ No____

If yes, how many licenses were granted?_____

g. Were any commercial development activities taken to develop the invention into a commercial product or service for manufacture or sale? Yes___ No____

If yes, describe the commercial development activities:

23(B) Based on the results of this project, are you planning to file for any licenses or patents, or undertake any commercial development opportunities in the future?

Yes_____ No ___X

If yes, please describe your plans:

24. Key Investigator Qualifications. Briefly describe the education, research interests and experience and professional commitments of the Principal Investigator and all other key investigators. In place of narrative you may insert the NIH biosketch form here; however, please limit each biosketch to 1-2 pages. *For Nonformula grants only – include information for only those key investigators whose biosketches were not included in the original grant application.*

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors in the order listed on Form Page 2.
Follow this format for each person. **DO NOT EXCEED FOUR PAGES.**

NAME	POSITION TITLE
Julie A. Mennella	Member
eRA COMMONS USER NAME (credential, e.g., agency login)	
JMennella	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	MM/YY	FIELD OF STUDY
Loyola University, Chicago, IL	BS	06/77	Biology
DePaul University, Chicago, IL	MS	02/81	Biology
The University of Chicago, Chicago, IL	PhD	08/88	Biopsychology
Monell Chemical Senses Center, Philadelphia, PA	Postdoc	09/88-06/90	Chemical Senses

A. Personal Statement

When I was a post-doctoral fellow conducting research on the transfer of dietary volatiles to human milk, I made a serendipitous finding that human milk was flavored after a lactating woman drank one glass of beer. This led to a systematic study of the transfer of alcohol to milk and its effects on infant nursing and mother-infant interaction, the findings of which were the lead article in the October 3rd, 1991 issue of the *New England Journal of Medicine*. I have expanded this research area to include 1) the study of the effects of tobacco on lactation; 2) the study of the effect of family history of addictions on taste preferences in adults and children; and 3) the effects of environmental tobacco smoke (ETS) exposure on health outcome as well as the emotional attachment to the odor of ETS during childhood. This research topic then became a passion for it combined my interests in lactation and child development, endocrinology, women's health and history of science, for this was an area for which there was more folklore than scientific knowledge and no established experimental paradigms. I have also made a strong commitment over the years to ensure that the information gleaned from this basic research program is communicated to need-to-know groups s (see Outreach, below).

B. Positions and Honors

Positions and Employment:

1990-1995 Assistant Member, Monell Chemical Senses Center, Philadelphia, PA
1995-1999 Associate Member, Monell Chemical Senses Center, Philadelphia, PA
1999-present Member, Monell Chemical Senses Center, Philadelphia, PA

Selected Honors and Service:

1991-1993 First recipient of the Morley R. Kare Fellowship, Monell Chemical Senses Center
1991-2008 Founding Director of the Monell Science Apprenticeship Program for High School Students and Undergraduates
1996 First recipient of the Avanelle Kirksey Lectureship Award for research excellence in maternal nutrition and infant behavior, Purdue University, West Lafayette, Indiana
1997 National Institute on Alcohol Abuse and Alcoholism, Extramural Scientific Advisory Board on Fetal Alcohol Syndrome

1997	Association for Chemoreception Sciences (AChemS) Moskowitz-Jacobs Award for research excellence in psychophysics of taste and smell
1999	Elizabeth W. Bingham Award for science mentoring, Association for Women in Science
1999-present	Member, Minority and Clinical Travel Fellowship Awards Committee, AChemS
2001	L. J. Filer, Jr. Lecturer, The University of Iowa School of Medicine, Department of Pediatrics
2002-present	Consultant for US Agency for International Development (USAID); Infant feeding project, Lilongwe, Malawi
2003	Invited Speaker, WHO Global Health Forum, Geneva, Switzerland
2003	Member of the NIAAA Dietary Guidelines (Moderate Drinking) Working Group
2004-present	Member, Scientific Advisory Committee, Shape up America!
2005-present	Chair, Taste and Flavor Working Group, Best Pharmaceuticals for Children Act, Pediatric Formulation Initiative Working Group, National Institute of Child Health and Human Development.
2006-present	Member, Editorial Board, Developmental Psychobiology
2007-2009	Member, Writing Group, American Heart Association "Helping Families Implement Nutrition Recommendations"
2007-present	Consultant, NIH Toolbox of Neurological and Behavioral Function: Taste and Olfaction Groups
2009	Invited Seminar Speaker: Adequate Pediatric Formulations in the 21st Century (Hot Topic Seminar); American Association of Pharmaceutical Sciences, Los Angeles, CA
2010	Panel Member, NIH Consensus Development Conference: Lactose Intolerance and Health
2010	Bring the Family Address, Association for Psychological Sciences 22nd annual convention, Boston, MA

Professional Memberships: Society for the Study of Ingestive Behavior; International Society for Developmental Psychobiology; International Society for Research in Human Milk and Lactation; Association for Psychological Sciences; American Psychological Association; Association of Chemoreception Sciences; Research Society on Alcoholism, Fetal Alcohol Spectrum Disorder Study Group.

Outreach to Need-to-Know Groups: Speaker and lecturer for continuing education courses for lactation consultants, dieticians, and physicians on various topics including the formation of food preferences, physiology of lactation, and effects of moderate drinking on lactation.

C. Selected Peer-Reviewed Publications (Selected from more than 90 peer-reviewed publications)

1. Mennella JA, Beauchamp GK. The transfer of alcohol to human milk: Effects on flavor and the infant's behavior. *New England Journal of Medicine*, 325: 981-985, 1991
2. Mennella JA, Beauchamp GK. Smoking and the flavor of breast milk- Letter. *New England Journal of Medicine* 339: 1559-60, 1998.
3. Mennella JA, Garcia PL. The child's hedonic response to the smell of alcohol: Effects of parental drinking habits. *Alcoholism: Clinical and Experimental Research* 24: 1167-1171, 2000.
4. Mennella JA, Pepino MY, Reed DR. Genetic and environmental determinants of bitter perception and sweet preferences in children and adults. *Pediatrics* 115:e216-22, 2005. PMCID: 1397914

5. Pepino MY, Mennella JA. Sucrose-induced analgesia is related to sweet preferences in children but not adults. *Pain* 119: 210-217, 2005.
6. Forestell CA, Mennella JA. Children's hedonic judgments of cigarette odor: Effects of parental smoking and maternal mood. *Psychology of Addictive Behaviors* 19: 423-432, 2005.
7. Mennella JA, Yourshaw LM, Morgan, LK. Breastfeeding and smoking: Short-term effects on infant feeding and sleep. *Pediatrics* 120:497-502, 2007. PMCID: 2277470
8. Pepino MY, Mennella JA. Effects of cigarette smoking and family history of alcoholism on sweet taste perception and food cravings in women. *Alcohol: Clinical and Experimental Research* 31:1891-9, 2007. PMCID: 2268904
9. Mennella JA, Forestell CA. Children's hedonic responses to the odors of alcoholic beverages: A window to emotions. *Alcohol* 42:249-60, 2008. PMCID: 2483837
10. Gidding SS, Lichtenstein A, Faith MS, Karpyn A, **Mennella JA**, Popkin B, Rowe J, Van Horn L, Whitsel L. Implementing American Heart Association pediatric and adult nutrition guidelines: a scientific statement from the American Heart Association Nutrition Committee of the Council on Nutrition, Physical Activity and Metabolism, Council on Cardiovascular Disease in the Young, Council on Arteriosclerosis, Thrombosis and Vascular Biology, Council on Cardiovascular Nursing, Council on Epidemiology and Prevention, and Council for High Blood Pressure Research. *Circulation* 119: 1161-1175; 2009.
11. **Mennella JA**, Pepino, MY, Duke F, Reed DR. Age modifies the genotype-phenotype relationship for the bitter receptor *TAS2R38*. *BMC Genetics* 11:60, 2010. PMID: 20594349. PubMed - indexed for MEDLINE.

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors in the order listed on Form Page 2.
Follow this format for each person. **DO NOT EXCEED FOUR PAGES.**

NAME	POSITION TITLE
Danielle Renee Reed	Member
eRA COMMONS USER NAME (credential, e.g., agency login) DANIELLEREED	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	MM/YY	FIELD OF STUDY
University of Washington	B.S.	05/84	Psychology
Yale University	Ph.D.	05/90	Psychology
Monell Chemical Senses Center		05/90	Metabolism
University of Pennsylvania	Postdoc	05/95	Genetics

A. Personal statement.

I am particularly well suited to be a collaborator for this project because Dr. Mennella and I have an established system for managing a large collection of DNA samples from children and parents tested for taste and other phenotypes. Dr. Mennella is an investigator for several of my genetically oriented projects, and I likewise serve in this capacity on her behaviorally oriented projects. We have published papers in *Pediatrics, BMC Genetics, and Chemical Senses* (listed below) that focus on genotype and how it affects taste perception in children and adults. My laboratory is well staffed and well equipped to undertake the genetics work outlined in this application. We currently have several instruments for genotyping, well-trained personnel to manage the flow of DNA samples, and an established track record in this area, which includes genotyping for the NIH Toolbox tests designed for taste, as well as other NIH-funded investigators. I have published numerous papers on the genetics of flavor perception and consider myself expert in this area.

B. Positions and honors.

Yale Fellowship	1984-1987
Howard Heinz Endowment Fellowship	1987-1990
NIH-PHS Award # 5-T32-CA09430	1990-1992
NIH-NRSA Award #1-F32-DK08732	1992-1994
University of Pennsylvania, Research Assistant Professor	1995-2000
Monell Chemical Senses Center, Assistant Member	2000
Monell Chemical Senses Center, Associate Member	2000
NIH, Metabolism Study Section, Temporary Member	2004
NIH, IPOD Study Section, Temporary Member	2004-2005
NIH, CADO Study Section, Member	2004-2010
Queensland Institute Medical Research, Visiting Scientist	2005; 2010
Monell Chemical Senses Center, Member	2007-current
NIH, SCS Study Section Temporary Member	2008
IFF Award for Outstanding Research	2008
NIH, CIDO Study Section, Temporary Member	2009
NIH, Shared Equipment Study Section, Temporary Member	2009
Invited Speaker, Association for Chemoreception Scien	2009
Korean Taste Society, Invited Speaker	2010

D. Peer-reviewed publications or manuscripts in press most relevant to the current application (selected from 92).

Reed DR, Bartoshuk LM, Duffy V, Marino S, Price RA. Propylthiouracil tasting: determination of underlying threshold distributions using maximum likelihood. Chemical Senses, 1995, 20, 529-533. PMID: 8564427

Reed DR, Bartoshuk LM, Price RA. Localization of a gene for bitter-taste perception (PROP) to human chromosome 5p15, American Journal of Human Genetics, 1999, 64, 1478-1480. PMCID: PMC1377888

Guo SW, **Reed DR**. The genetics of phenylthiocarbamide perception. Annals of Human Biology, 28, 111-142, 2001. PMID: 11293722

Duffy VB, Davidson AC, Kidd JR, Kidd KK, Speed WC, Pakstis AJ, **Reed DR**, Snyder DJ, Bartoshuk LM. Bitter receptor gene (*TAS2R38*), 6-n-propylthiouracil (PROP) bitterness and alcohol intake. Alcoholism, Clinical and Experimental Research, 2004, 28, 1629-1637. PMCID: PMC1397913

Bufe B, Breslin PAS, Kuhn C, **Reed DR**, Tharp CD, Slack JP, Kim UK, Drayna D, Meyerhof W. The molecular basis of individual differences in phenylthiocarbamide and propylthiouracil bitterness perception. Current Biology, 2005, 15, 322-327. PMCID: PMC1400547

Mennella JA, Pepino Y, **Reed DR**. Genetic and environmental determinants of bitter perception and sweet preference. Pediatrics, 2005, 115, e216-e222. PMCID: PMC1397914

Hansen J, **Reed DR**, Wright M, Martin N, Breslin PAS. Heritability and genetic covariation of sensitivity to PROP, SOA, quinine HCl and caffeine. Chemical Senses, 2006, 31, 403-413. PMCID: PMC1475779

Reed DR, Tanaka T, McDaniel A. Diverse tastes: genetics of sweet and bitter perception. Physiology and Behavior, 88, 215-226, 2006. PMCID: PMC1698869

Wise P, Hansen JL, **Reed DR**, Breslin PAS. Twin study of the heritability of recognition thresholds for sour and salty taste. Chemical Senses, 2007, 32, 749-754. PMCID: PMC2085364

Reed DR. Birth of a new breed of supertaster. Chemical Senses, 2008, 33, 489-91. PMID: 1856244

Coldwell SE, Oswald TK, **Reed DR**. A marker for bone growth differs between adolescents with high vs. low sugar preference. Physiology and Behavior, 2009, 96, 574-580. PMCID: PMC2764307

Mennella, J.A., Duke, F., Pepino, M.Y., **Reed DR**. Age modifies the genotype-phenotype relationship for the bitter receptor TAS2R38 BMC Genetics, 2010, *11*, 60. PMCID: in progress.

Mennella,JA., Pepino, MY, Duke FF, **Reed DR**. Psychophysical dissection of genotype effects on human bitter perception, Chemical Senses, 2010, In progress. NIHMSID: 250589

Reed DR, Zhu G., Breslin PA, Duke FF, Henders AK, Campbell MJ, Montgomery GW, Medland SE, Martin NG, Wright MJ. The perception of quinine taste intensity is associated with common genetic variants in a bitter receptor cluster on chromosome 12. Hum Mol Genet., 2010, In progress. PMCID: 2951861

Pelchat ML, Bykowski C, Duke FF, **Reed DR**. Excretion and perception of a characteristic odor in urine after asparagus ingestion: a psychophysical and genetic study. Chemical Senses, 2010, 36, 9-17. PMCID: PMC3002398.

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors in the order listed on Form Page 2.
Follow this format for each person. **DO NOT EXCEED FOUR PAGES.**

NAME OF FELLOWSHIP APPLICANT Sarah V. Lipchock	POSITION TITLE Postdoctoral Fellow
eRA COMMONS USER NAME (credential, e.g., agency login) slipchock	

EDUCATION/TRAINING (Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	YEAR(s)	FIELD OF STUDY
McDaniel College, Westminster, MD	BA	2000-2004	Chemistry, minor in Math
Yale University, New Haven, CT	MS	2004-2005	Biophysical Chemistry
Yale University, New Haven, CT	PhD	2004-2010	Biophysical Chemistry
Monell Chemical Senses Center, Philadelphia, PA	Postdoc	2010-present	Psychophysics, Genetics

A. Personal Statement

I have a strong background in structure-function relationships of proteins and nucleic acids and have worked extensively on RNAs that regulate gene expression in cells. Genetics provides an ideal opportunity to show that a single molecular substitution can have a dramatic functional affect on both gene expression and gene product function. The taste system translates these small changes in function into measurable changes in human experience. The study of these genetic contributions to smoking status in mothers and their children builds on my prior experiences and provides significant new training, which can utilize my molecular background to ask important questions related to human health and nutrition.

B. Positions and Honors

ACTIVITY/OCCUPATION	BEGINNING DATE (mm/yy)	ENDING DATE (mm/yy)	FIELD	INSTITUTION/COMPANY	SUPERVISOR/ EMPLOYER
Lab Assistant/Tutor	08/02	05/04	Chemistry	McDaniel College	Marilyn Kroeger-Smith, PhD
Undergraduate Research Student	06/03	01/04	Computational Chemistry	McDaniel College	Richard Smith, PhD Marilyn Kroeger-Smith, PhD
Graduate Research Assistant	04/05	05/10	Biophysical Chemistry	Yale University	Scott Strobel, PhD
Graduate Teaching Assistant	01/06	12/07	Chemistry	Yale University	Kurt Zilm, PhD John Tulley, PhD Michael McBride, PhD
Instructor	08/09	12/09	First Year Seminar	University of Bridgeport	Jeffrey Johnson, PhD
Postdoctoral Fellow	07/10	present	Genetics and Psychophysics	Monell Chemical Senses Center	Danielle Reed, PhD, Julie Mennella, PhD

Academic and Professional Honors

2000	Valedictorian, Cambridge-South Dorchester High School, Cambridge, MD
2000-2004	Maryland Distinguished Scholar Finalist, Scholarship Recipient
2000-2004	McDaniel College Dean's Highest Honors
2003-2004	Gamma Sigma Epsilon, Chemistry Honor Society, Member
2003-2004	Kappa Mu Epsilon, Mathematics Honor Society, Member and Secretary
2003-2004	Omicron Delta Kappa, Leadership Honor Society, Member
2004	Phi Beta Kappa, Member
2004	College Scholar (Honors Program Graduate), McDaniel College
2004	Summa Cum Laude (3.99 GPA), Honors in Chemistry
2006	Chemistry Department Teaching Award, Yale University
2007	Yale University Molecular Biophysics and Biochemistry Poster Prize

C. Publications

Research Papers

Cochrane, JC, **Lipchock, SV**, Strobel, SA. "Structural investigation of the glmS ribozyme bound to its catalytic cofactor." *Chem Biol,* 2008. 14:97-105.

Lipchock, SV, Strobel, SA. "A relaxed active site following exon ligation by the group I intron." *Proc Natl Acad Sci*, 2008. 105: 5699-704.

Cochrane, JC, **Lipchock, SV**, Smith, KD, Strobel, SA. "Structural and chemical basis for glucosamine-6-phosphate binding and activation of the glmS ribozyme." *Biochemistry*, 2009. 48: 3239-46.

Smith, KD, **Lipchock, SV**, Ames, TD, Wang, J, Breaker, RR, Strobel, SA. "Structural basis of cyclic-di-GMP binding to the GEMM riboswitch." *Nat Struct Mol Biol*, 2009. 16: 1218-23.

Antonioli, AH, Cochrane, JC, **Lipchock, SV**, Strobel, SA. "Plasticity of the kink turn structural motif." *RNA*, 2010. 16: 762-8.

Smith, KD, **Lipchock, SV**, Livingston, A, Shanahan, CA, Strobel, SA. "Structural and biochemical determinants of ligand binding by a c-di-GMP riboswitch." *Biochemistry*, 2010. 49: 7351-9.

Book Chapters

Mennella, JA, Ventura, AK, **Lipchock, SV**. "The origins of food preferences." In *Sento dunque sono (I feel therefore I am)* Bellieni, CV (Ed.) 2011. Edizioni Cantagalli: Siena. *In Press.*

Abstracts

Lipchock, SV, Strobel, SA. "Structural studies of the group I intron splicing pathway." Oral presentation, 2006 Bristol Myers Squibb Symposium, Yale University.

Lipchock, SV, Strobel SA. "Visualization of the group I intron splicing pathway through crystal structures of splicing intermediates." Oral presentation, 2007 Biophysics Club, Yale University.

Lipchock, SV, Strobel, SA. "A relaxed active site after exon ligation by the group I intron." Poster presentation, 2007 RNA Society Meeting, Madison, WI.

Lipchock, SV, Strobel, SA. "A loosely bound spliced exon complex in the group I intron." Oral presentation, 2007 RNA Club, Yale University.

Lipchock, SV, Strobel, SA. “A relaxed active site following exon ligation by the group I intron.” Poster presentation, 2008 Nucleic Acids Gordon Conference, Newport, RI.

Lipchock, SV, Strobel, SA. “A relaxed active site following exon ligation by the group I intron.” Oral presentation, 2008 Yale Molecular Biophysics and Biochemistry Retreat, Woods Hole, MA.

Lipchock, SV, Strobel, SA. “A relaxed active site following exon ligation by the group I intron.” Oral presentation, 2008 Center for Structural Biology Seminar Series, Yale University.

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors in the order listed on Form Page 2.
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NAME	POSITION TITLE
Stacie S. Miller	Research Fellow- Postdoctoral
eRA COMMONS USER NAME (credential, e.g., agency login) SMILLER9	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	MM/YY	FIELD OF STUDY
Knox College, Galesburg, IL	B.A.	2003	Psychology
Binghamton University, Binghamton, NY	M.A.	2006	Psychology
Binghamton University, Binghamton, NY	Ph.D.	2010	Behavioral Neuroscience
Monell Chemical Senses Center, Philadelphia, PA		2010-present	Psychophysics, Neuophysiology

A. Personal Statement

My experience as a graduate student studying early ethanol exposure makes me particularly well suited for my role on this project. Early ethanol exposure changes ones response to ethanol odor as well as ethanol taste. Studying the effect of tobacco on later taste and food preferences was a natural extension of my previous training. My participation in this project gives me the opportunity to learn new techniques such as psychophysical taste testing.

B. Positions and Honors

ACTIVITY/OCCUPATION	BEGINNING DATE (mm/yy)	ENDING DATE (mm/yy)	FIELD	INSTITUTION/COMPANY	SUPERVISOR/ EMPLOYER
Research Project Assistant	07/03	08/03	Psychology	Binghamton University	Dr. Norman E. Spear
Graduate Teacher's Assistant	08/03	05/04	Psychology	Binghamton University	State of New York
Graduate Research Assistant	05/04	12/07	Psychology	Binghamton University	Dr. Norman E. Spear
NRSA Postdoctoral Fellow	12/07	12/09	Psychology	Binghamton University	NIAAA
Graduate Research Assistant	12/09	10/10	Psychology	Binghamton University	Dr. Norman E. Spear
Postdoctoral Fellow	11/10	present	Psychophysics, Neurophysiology	Monell Chemical Senses Center	Dr. Julie Mennella

Academic and Professional Honors

Ellen Browning Scripps Scholarship, 1999-2003
Richter Foundation award to fund honors research, 2002-2003
B.A. awarded with honors, 2003
NIH travel grant to attend the 2005 meeting of the International Society for Developmental Psychobiology

NIH travel grant to attend the 2006 meeting of the International Society for Developmental Psychobiology
NIH travel grant to attend the 2007 meeting of the International Society for Developmental Psychobiology
NIH travel grant to attend the 2008 meeting of the International Society for Developmental Psychobiology
RSA student merit award (travel award) to attend the 2009 meeting of the Research Society on Alcoholism

Memberships in professional societies:
Psi Chi, 2002 - 2003
Phi Theta Kappa, 2003
International Society for Developmental Psychobiology, 2005-2009
Society for Research in Child Development, 2006-2007
Research Society on Alcoholism, 2007-2010

C. Selected Peer-reviewed Publications

Pautassi, R. M., Sanders, S., **Miller, S**., Spear, N. E. & Molina, J. C. (2006). Early ethanol's anxiolytic effects assessed through an unconditional stimulus revaluation procedure. Alcoholism: Clinical and Experimental Research, 30, 448-459. PMID: 16499485

Miller, S. S., & Urcelay, G. P. (2007). The central amygdala joins the lateral amygdala in the fear memory party. The Journal of Neuroscience, 27, 2151-2152. [Journal Club article] PMID: 17329410

Miller, S. S., Hoffmann, H. L., & Mustanski, B. S. (2008). Fluctuating asymmetry and sexual orientation in men and women. *Archives of Sexual Behavior, 37,* 150-157. PMID: 18163207

Miller, S. S., & Spear, N. E. (2008). Olfactory learning in the rat neonate soon after birth. *Developmental Psychobiology, 50,* 554-565. PMID: 18683189

Arias, C., Mlewski, E. C., **Miller, S. S.**, Molina, J. C., & Spear, N. E. (2009). Novelty modulates the stimulating motor effects of ethanol in preweanling rats. *Pharmacology, Biochemistry and Behavior, 92*, 448-456. PMID: 19463258

Miller, S. S. (July 22, 2009). Alcohol can harm unborn or nursing babies. Guest Viewpoint published in the Viewpoints section of the *Press & Sun Bulletin*, Greater Binghamton's newspaper.

Miller, S. S., & Spear, N. E. (2009). Olfactory learning in the rat immediately after birth: unique salience of first odors. *Developmental Psychobiology, 51,* 488-504. PMID: 19582793

Miller, S. S., & Spear, N. E. (2010). Mere odor exposure learning in the rat neonate immediately after birth and 1 day later. *DevelopmentalPsychobiology, 52,* 343-351. PMID: 20411590

Arias, C., Solari, A. C., Mlewski, E. C., **Miller, S.**, Haymal, B., Spear, N. E., & Molina, J. C. (2010). Social isolation and stress related hormones modulate the stimulating effect of ethanol in preweanling rats. *Behavioural Brain Research, 211,* 64-70. PMID: 20226814

D. Research Support

Completed Research Support

1F31AA017339-01 Miller (PI)
12/05/07-12/31/09
Screening For Moderate Prenatal Ethanol Exposure Soon After Birth
National Research Service Award (NRSA) from the National Institute on Alcohol Abuse and Alcoholism