# BEVERAGE CONSUMPTION AND BODY COMPOSITION AMONG COLLEGE-

## AGED WOMEN

A Thesis Presented

by

## MATTHEW SLOAN

Submitted to the Graduate School of the University of Massachusetts Amherst in partial fulfillment of the requirements for the degree of

## MASTERS OF SCIENCE

May 2011

Biostatistics and Epidemiology

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Approved as to style and content by:

Elizabeth Bertone-Johnson, Chair

Lisa Chasan-Taber, Member

Ed Stanek, Member

Alayne Ronnenberg, Member

Ed Stanek, Department Head, Department of Public Health

#### ACKNOWLEDGMENTS

I would like to thank my advisor, Elizabeth Bertone-Johnson, for her guidance and support. Thanks also to Lisa Chasan-Taber for her many meticulous reviews and comments on my proposal, thesis, and presentation. Thanks to Alayne Ronnenberg for taking me on as an undergraduate, and first permitting my involvement with the UMass Vitamin D Status Study. Thanks to Ed Stanek for teaching me the principles of biostatistics and data analysis that were essential for my thesis. Special thanks to Sofija Zagarins, Biki Takashima, and Patricia Chocano-Bedoya for their work collecting and making the data for this project accessible.

Thank you to all of the participants in the study who volunteered their time and permitted the exams required to gather our data. Thank you to my labmates Constance Barysauskas and Lori Peterson whose friendship and camaraderie helped keep me on track to finish this project and provided important feedback during lab meetings.

Thank you to all my friends and family who supported me through this endeavor.

#### ABSTRACT

## BEVERAGE CONSUMPTION AND BODY COMPOSITION AMONG COLLEGE-AGED WOMEN MAY 2011

## MATTHEW SLOAN, B.S., UNIVERSITY OF MASSACHUSETTS AMHERST, M.S., UNIVERSITY OF MASSCHUSETTS AMHERST

Directed by: Professor Elizabeth Bertone-Johnson

In the U.S., over 67 million adults are obese and 300,000 annual deaths are related to obesity. Among college-aged women, over 60% report daily consumption of caloric beverages. Prior studies indicate positive associations between these beverages and obesity, but conflicting results for diet drinks. Studies were limited, however, by obesity measures that failed to accurately assess abdominal adiposity or percent body fat, and few studies included college-aged women.

We examined this relationship among participants aged 18-30 in the University of Massachusetts Vitamin D Status Study (n=237). We assessed average diet in the past two months using a modified version of the Harvard Food Frequency Questionnaire and calculated percent body fat by dual-energy X-ray absorptiomtery. Confounding factors were assessed using a lifestyle questionnaire. Multiple logistic regression was used to adjust for important risk factors.

We found no association between intake of sugar-sweetened beverages or juice and obesity after controlling for confounding factors. However, high consumption of diet drinks (i.e., >2 servings per week) was associated with an increased risk of overweight (BMI>25) (OR=2.88, 95% CI 1.34, 6.21), high waist circumference (>80 cm) (OR=3.14,

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95% CI 1.56, 6.35) and high percent body fat (>33%) (OR=2.86, 95% CI 1.42, 5.77) as compared to light consumption (i.e, <1 serving per month). These associations were not attenuated by controlling for total caloric intake. Findings should be evaluated in additional longitudinal studies to determine whether diet drinks contribute to adiposity or if the association is due to higher diet drink consumption by overweight women.

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#### CHAPTER I

#### INTRODUCTION

Obesity rates have reached epidemic proportions. Over 67 million American adults are obese (Body mass index [BMI; kg/m<sup>2</sup>]  $\ge$  30) and 300,000 annual deaths are related to obesity. In 2008, the prevalence rate of obesity among adult women was 34%, an increase from 15.7% in 1962 (1-4). There are now more than one billion overweight (BMI  $\ge$ 25 kg/m<sup>2</sup>) individuals globally (5). Obesity rates are increasing across all age groups in the United States. Among college-aged students, the prevalence rate of overweight and obesity is 35% (6). Obesity is related to cardiovascular disease (CVD), insulin resistance and type 2 diabetes, hypertension, stroke, cancer, and all-cause mortality (7-12).

Many established risk factors for obesity have been identified. Childhood obesity has been associated with a number of risk factors including parental obesity, excessive television watching, early weight gain, birth weight, and inadequate sleep (13). In adolescence, risk factors for obesity include lack of exercise, total energy consumed per day, and fiber consumed per day (14). In women followed from adolescence into adulthood, the percent of energy intake as carbohydrate was significantly associated with skinfold measures of obesity (15). The consumption of fructose, a component of table sugar and high-fructose corn syrup has recently been positively associated with body weight and hazardous effects on cardiometabolic health in children, adolescents and adults (16).

Ecologic data have suggested a link between beverage consumption and obesity in a number of populations, since beverage consumption has paralleled the rise in obesity (17-19). Since 1977, the absolute national intake of sugar-sweetened beverages and fruit juice has significantly increased, as has the percentage of total energy intake from sugarsweetened beverages and fruit juice. This observation has led to further investigation of beverage consumption as an emerging risk factor for obesity. More than half of the increase in intake of caloric sweeteners since 1977 came from increases in beverage consumption (17). Among American women, approximately 20% report consuming more than one caloric beverage per week (20). In comparison, more than 60% of female college students report daily intake of sugar-sweetened beverages (21).

Sugar-sweetened beverage consumption is believed to lead to obesity through four mechanisms: 1) conversion of fructose to fat in the liver; 2) genetic effects; 3) decreased satiety from liquid calories; and 4) increased caloric intake. The association between fruit juice and obesity is believed to result from the latter three hypothesized mechanisms. The mechanism between diet drinks and obesity is unclear, but may involve modified taste preference or, alternatively, a link with healthy behavior and decreased caloric intake.

Prior epidemiologic studies assessing the association of sugar-sweetened beverage, fruit juice, diet drink intake, and risk of obesity suggest either positive associations or null association for SSB and fruit juice consumption (16, 20, 22-41), and no association or an inverse association for diet drink consumption (26, 28, 39). The strength of these associations varied depending on the statistical analysis methods used and the comparison groups chosen, but increased consumption of sugar-sweetened

beverages and fruit juice remained significantly associated with increased BMI and incidence of obesity in one large cohort of adults after adjusting for total energy intake (26). However, in a prospective study of children, this association did not persist after energy adjustment, nor was it present for any of the assessed beverages (39).

The association between beverages and obesity in populations of exclusively college-aged women has not yet been addressed in the literature. Additionally, the majority of studies in other populations used self-reported height and weight to calculate BMI, which was then used as the measure of body composition. Even in studies that used trained examiners to measure height and weight, BMI has limited ability to predict negative health outcomes. Waist circumference, a measure of abdominal adiposity, has been shown to predict mortality independently after adjusting for BMI (42); this indicates that BMI alone is an insufficient body composition measure to fully capture adiposity and should be paired with additional measures, such as waist circumference or percent body fat, in order to account for the unique contribution of these outcomes to health risks.

Results from previous studies have also varied by age, with some studies finding no association between SSB consumption and obesity (39) or an inverse association between juice consumption and obesity in children (35), but a positive association between sugar-sweetened beverages and fruit juice and risk of obesity in adults (24, 26). It is important to evaluate this association among college-aged women because identification of factors related to obesity in this population will allow for targeted intervention. This intervention could provide an opportunity to modify long-term risk factors prior to the onset of obesity. Further, children with obese parents are at greater risk of becoming obese (13, 14). College-aged women are entering their child-bearing

years and strategies to minimize obesity in this age group may decrease the likelihood of having obese children, because behaviors leading to obesity are likely passed from parents to children. Also, early college years are a period of substantial potential weight gain, making this an opportune time for intervention.

Therefore, we evaluated the cross-sectional association between beverage consumption and obesity in a population of college-aged women. Dietary information on sugar-sweetened beverage, fruit juice, and diet drink consumption was ascertained via a validated food frequency questionnaire. Body composition was assessed by three measures: BMI, waist circumference, and percent body fat. These multiple measures of obesity allowed us to accurately identify the contribution of beverage intake to obesity risk.

#### CHAPTER II

#### PHYSIOLOGY OF BEVERAGE CONSUMPTION AND OBESITY

Ecologic studies have suggested a positive association between beverage consumption and obesity (43, 44), although the mechanism is unclear. Proposed mechanisms linking beverage consumption to obesity include conversion of fructose to fat in the liver (45), an interaction between the dietary components of beverages and the genes that predispose toward obesity (46), low satiety from liquid calories compared to solid calories (47, 48), and increased caloric intake (20).

The first potential mechanism involves differences in the metabolism of fructose and glucose in the liver. Fructose is found naturally in fruits and honey. The largest component of fructose in the diet, however, comes from added sugar and high-fructose corn syrup. More than one-third of these sweeteners are consumed in the form of sugarsweetened beverages and nearly one-tenth are consumed as fruit juice (17). Glucose requires the presence of insulin for uptake into cells, while fructose is metabolized independently from insulin (49). In the liver, glucose is metabolized into glycogen or ATP as needed. Fructose can be isomerized to glucose or converted to fat. Fructose ingestion in humans has been shown to cause greater lipogenesis than glucose and does not stimulate insulin nor leptin secretion, which are involved in energy homeostasis (45). This could result in greater fat accumulation from consuming diets high in fructose than diets high in glucose.

The second potential mechanism is based on the observation that as much as a 40% of obesity may be explained by genetic factors. (46) It is possible that sugarsweetened beverage and juice consumption may interact with genes predisposing obesity

by affecting regulatory hormones involved in energy intake and expenditure. No studies have yet addressed this hypothesized interaction.

The third potential mechanism suggests that food form relates directly to energy consumption, with solid foods associated with greater satiety than liquids of similar energy density. It has been shown that eating solid foods of equal caloric density to sugar-sweetened beverages or juice leads individuals to consume fewer calories. (48) Further, high glycemic index foods, such as sugar-sweetened beverages and fruit juice, are digested rapidly in the small intestine, and nutrient receptors in the gastrointestinal tract are stimulated for shorter periods of time than high glycemic index foods. This leads to decreased satiety of high glycemic index foods relative to low glycemic index foods (47).

The fourth potential mechanism for the association between sugar-sweetened beverage and juice consumption and risk of obesity is related to overall increased energy intake and positive energy balance. Cross-sectional analyses in one population show sugar-sweetened beverages (+215.9 kcal, p<0.001) and fruit juice (+270.0 kcal, p<0.001) to be significantly positively associated with energy intake (20). In contrast, diet drink intake (-11.9 kcal, p=0.45) was not associated with increased energy intake (20). Increased energy intake, if not compensated for with increased energy expenditure, leads to positive energy balance and subsequent weight gain. In a review of 21 articles, either significantly positive or null associations between sugar-sweetened beverages and total energy intake were found in all studies (27). This mechanism stands out among the proposed hypotheses as the best explanation so far, because adjustment for total energy has been found to eliminate significant associations between sugar-sweetened beverages

and juice and risk of obesity (20). This indicates that the added caloric contribution of these beverages might be the main link between beverage consumption and obesity.

Diet beverage intake may plausibly be associated with higher or lower risk of obesity. Diet beverage consumption and obesity may be related through an effect on the development of preference for sweet foods. These preferences are the primary contributor to food selection in childhood, but only partially contribute, along with nutritional decisions, to food selection in adults (50). Preference for sweet tastes could link diet drink consumption with intake of other calorie-dense foods and subsequent positive energy balance and, therefore, be positively associated with obesity risk. In contrast, diet drink consumption may be associated with increased calcium intake and health-conscious diet decisions, such as weight-loss food choices or displacement of caloric beverages, in some populations (20, 25, 51). This could result in an inverse association between diet drink consumption and weight gain.

In summary, the mechanistic explanations for the association between sugarsweetened beverage and juice consumption and body composition are not fully established, but include the increased lipogenic ability of fructose-containing foods (like sugar-sweetened beverages and juice), the interaction of beverages with genes predisposing for obesity, decreased satiety from caloric beverages, and increased caloric intake. Mechanisms explaining a positive association between diet drinks and obesity is associated with taste preference for sweet, calorie-dense foods. Alternatively, the mechanism explaining an inverse association between diet drink and obesity relies on the high correlation of diet drinks with weight-loss food choices and displacement of caloric beverages.

#### CHAPTER III

#### EPIDEMIOLOGY OF BEVERAGE CONSUMPTION AND OBESITY

We identified three cross-sectional studies of the association between beverage consumption and obesity (24, 40, 52), ten longitudinal studies (20, 22, 23, 25, 26, 35, 38, 39, 53, 54), and three randomized trials (28, 34, 55). Among the cross-sectional studies, one found a significantly positive association between sugar-sweetened beverage consumption and risk of obesity (40) and one found a positive association between fruit juice consumption and overweight status (24). Several of these studies are described in detail below.

Of the prospective cohorts, seven found significant positive associations between sugar-sweetened beverage consumption and obesity (20, 22, 25, 26, 38, 53, 54), while two found no association (35, 39), and one study found a positive association between juice consumption and obesity (23). Three prospective studies evaluated diet drink consumption and obesity and all failed to find a significant association (20, 26, 39).

Among the randomized trials, all three found positive associations between sugarsweetened beverage consumption and body weight (28, 34, 55). One of these trials involved substituting diet drinks for sugar-sweetened beverages, and found no significant association between diet drink and body weight (55). No trials on the association between fruit juice and obesity have been conducted.

The first prospective study to address the association between sugar-sweetened beverage consumption and body composition was conducted by Ludwig and colleagues in five Boston schools between 1995 and 1997 (25). Beverage consumption was ascertained from 548 students by a validated youth food frequency questionnaire

addressing intake of specified foods over the past 30 days. Body composition was measured by calculating BMI from height and weight at baseline and after a 19-month follow-up period. Obesity was defined as greater than the 85<sup>th</sup> percentile of a composite score of age-specific BMI and triceps-skinfold thickness. The analysis estimated the effect of change in sugar-sweetened beverage consumption on change in BMI and dichotomous obesity incidence.

Ludwig *et al.* found that for each additional daily serving of sugar-sweetened beverages above baseline, BMI increased  $0.24 \text{ kg/m}^2$  (95% CI 0.10-0.39, p=0.03) after multivariable adjustment for baseline anthropometrics, demographics, dietary variables, physical activity, television watching, and total energy intake. Similarly, for each additional daily serving of SSB above baseline, the incidence of obesity increased 60 percent (95% CI 1.14-2.24, p=0.02).

The main shortcoming of this study was the use of BMI as the only outcome measure. While BMI is convenient for assessing body composition in large populations and has been shown to predict metabolic syndrome (56), BMI alone is insufficient for measuring all important aspects of adiposity. Waist circumference and other measures have been shown to independently predict all-cause mortality risk after adjusting for BMI (42). Thus, it is best to measure body composition with BMI in conjunction with other measures to account for disease risk attributable to central adiposity (57).

One of the largest prospective cohort studies testing the association between SSB consumption and obesity in children and adolescents was performed by Berkey and colleagues (20). The study analyzed data from 16,771 children in the Growing Up Today Study. The subjects were from 50 states and were offspring of participants in the Nurses'

Health Study II. Over three years, beverage intake was assessed by a validated 132-item food frequency questionnaire. Body composition was measured each year by calculating BMI from self-reported height and weight. The analysis estimated the effect of one year change in beverage consumption on BMI change during the same year.

The results of this longitudinal analysis suggested a weak linear association for girls between sugar-sweetened beverage consumption and BMI change (Beta=0.03, p=0.08), which was attenuated after adjusting for total calorie intake (p=0.16). Overall, this did not support an association for the unique effect of sugar-sweetened beverage consumption on body composition independent of calorie intake. The primary shortcoming of this study was the use of BMI as the only body composition measure. As with the previous study, this measure may not sufficiently predict adverse health outcomes.

The largest prospective cohort study assessing sugar-sweetened beverage consumption and weight change in adult women was conducted by Schulze and colleagues (26). The study evaluated 51,603 women in the Nurses' Health Study II. Beverage consumption was ascertained by a mailed 133-item semiquantitative food frequency questionnaire three times over a nine year period. Body composition was calculated using self-reported height and weight at each dietary assessment. The analysis assessed mean weight change and mean BMI change for groups by specified changes in beverage consumption. Results were adjusted for age, alcohol, smoking, physical activity, BMI, baseline energy intake, and other confounders identified at baseline.

Compared to women whose intake of sugar-sweetened beverages remained the same or decreased, women who changed from low to high intake of sugar-sweetened

beverages gained significantly more weight (4.69 kg versus 1.34 kg, p<0.001) and BMI (1.72 kg/m<sup>2</sup> versus 0.49 kg/m<sup>2</sup>, p<0.001). All groups of women experienced weight gain, but the group with the lowest change in weight included women who decreased from high sugar-sweetened beverage intake to low sugar-sweetened beverage intake. Compared to women who decreased their juice consumption, women who increased their fruit juice consumption from one drink or less per week to one drink or more per day gained significantly more weight (4.03 kg versus 2.32 kg, p<.001). The weight change between the high-to-low and low-to-high groups was enhanced after adjusting for baseline energy intake. This indicates that the additional calories gained beyond baseline from changes in beverage consumption might be responsible for the weight gain differences between these groups.

Results for diet drink consumption were in the opposite direction as those for sugar-sweetened beverage and fruit juice consumption. Compared with women who decreased their diet drink consumption, women who increased consumption of diet drinks experienced significantly less weight gain (1.59 kg versus 4.25 kg, p<.001).

This study had the benefit of a large cohort to increase power to observe modest differences among groups. Additionally, the authors were able to address associations between fruit juice and diet drinks and risk of weight gain. However, the use of mean change in total weight and BMI is an important shortcoming of the study, because these intermediate endpoints do not fully predict disease risk (42). In comparison to the previous study by Berkey *et al.*, Schulze and colleagues adjusted for baseline energy intake, rather than total energy intake. The adjustment for baseline energy intake allows estimation of effects between individuals with different intake at baseline, whereas

adjustment for total energy intake allows for estimation of effects between individuals with different overall intake. The positive association persisted after this baseline energy adjustment, suggesting that excess caloric intake may be responsible for the observed results. However, no analysis was done adjusting for total energy intake, which limits the ability to determine whether excess caloric intake was solely responsible for this change.

In summary, the majority of studies suggest a significant positive association between consumption of sugar-sweetened beverages and obesity, though one study found that this association disappeared after adjustment for total energy intake. Similar results have been observed for the association of fruit juice. Some studies found that diet drink consumption had no association with obesity, but others found an inverse association.

#### CHAPTER IV

#### SUMMARY OF LITERATURE

Obesity in the U.S. and across the world is on the rise and is linked with a large number of chronic diseases. Increases in consumption of sugar-sweetened beverages, fruit juice, and diet drinks have paralleled the rise in obesity, leading to interest in whether consumption of these beverages is a contributor to the obesity epidemic. The potential mechanisms relating sugar-sweetened beverages and fruit juice to obesity involve fructose metabolism, genetic predisposition, reduced satiety, or increased caloric intake. Cross-sectional and prospective data appear to support a link between sugar-sweetened beverage and fruit juice consumption and risk of obesity, while diet drink consumption appears to have an inverse association with obesity, likely due to its link with healthy dietary choices or displacement of caloric beverages.

Three large prospective studies on sugar-sweetened beverage and fruit juice consumption and risk of obesity demonstrate significant positive associations, while one of these studies found a significant inverse association between diet drink consumption and risk of obesity. Two of these studies focused on young population and one focused on adult women. This suggests that sugar-sweetened beverage and fruit juice may play a role in the etiology of obesity across different age groups and genders. The association between diet drink and obesity remains unclear.

The greatest limitation to the current literature is the lack of comprehensive measures of obesity. Most studies rely on BMI as the primary indicator of body composition, and the majority of these studies obtained this information by self-report. It is important to address multiple measures of obesity, such as waist circumference and

percent body fat, which have been shown to be independently predictive of negative health outcomes after adjusting for BMI. It is essential to consider multiple measures to clarify the association between beverage consumption and the aspects of body composition most closely linked with disease, such as central adiposity.

Therefore, we proposed to evaluate the association between beverage consumption and body composition using three measures of body composition: BMI, waist circumference, and percent body fat among college-aged women. The population of college-aged women, a group with particularly high exposure to regular beverage consumption, has not been thoroughly addressed in the literature.

#### CHAPTER V

#### HYPOTHESES AND SPECIFIC AIMS

Specific Aim #1: Using a cross-sectional study design, we proposed to examine the association between consumption of various beverage types and body composition in college-aged women. The following hypotheses will be addressed:

Hypothesis 1a: Among college-aged women, those with higher levels of sugar-sweetened beverage consumption will have greater body mass index (BMI), waist circumference, and percent body fat than those with lower levels of sugar-sweetened beverage consumption; we further hypothesized that this will be a dose-response association.

Hypothesis 1b: Among college-aged women, those with higher levels of fruit juice consumption will have greater body mass index (BMI), waist circumference, and percent body fat than those with lower levels of fruit juice consumption; we further hypothesized that this will be a dose-response association.

Hypothesis 1c: Among college-aged women, those with higher levels of diet drink consumption will have lower body mass index (BMI), waist circumference, and percent body fat than those with lower levels of diet drink consumption; we further hypothesized that this will be a dose-response association.

#### CHAPTER VI

#### **METHODS**

Study Design

With a cross-sectional design, we assessed the association between beverage consumption and body composition using data from the University of Massachusetts Vitamin D Status Study (58). Participants were 237 healthy, premenopausal women aged 18-30 living in Amherst, MA, USA area and were enrolled from March 2006 to December 2010.

Women were ineligible if they: 1) were pregnant or not menstruating at the time of visit; 2) reported a history of high blood pressure or elevated cholesterol, kidney or liver disease, bone disease such as osteomalacia, digestive disorders, rheumatologic disease, multiple sclerosis, thyroid disease, hyperparathyroidism, cancer, type 1 or type 2 diabetes, polycystic ovaries, or experiencing untreated depression; or 3) reported taking corticosteroids, anabolic steroids, anticonvulsants, cimetidine, or propanolol (58).

All measurements were collected in a single clinic visit scheduled for the late luteal phase of each participant's menstrual cycle. Dual energy X-Ray absorptiometry (DXA) scans were completed on the morning of the study visit for all but ten participants from the beginning of the study.

#### Exposure Assessment

The exposure of interest for this study was beverage consumption. We assessed each subject's frequency of intake of 131 food items and supplements over the previous two months using a modified version of the Harvard Food Frequency Questionnaire

(FFQ) (59). Women were asked to report the number of servings per day they consumed of three different groups of beverages: sugar-sweetened beverages, fruit juice, and diet drinks. Sugar-sweetened beverages included: Coke, Pepsi, or other colas with sugar; caffeine-free Coke, Pepsi, or other caffeine-free colas with sugar; other carbonated beverages with sugar; Hawaiian Punch; lemonade; and other non-carbonated fruit drinks. One serving of sugar-sweetened beverages was equivalent to one glass, bottle, or can. Fruit juice included apple juice or cider, orange juice, grapefruit juice and other fruit juices. One serving of fruit juice was equivalent to 1 small glass. Diet drinks included low-calorie cola and low-calorie caffeine-free cola. One serving of diet drink was equivalent to one glass, bottle, or can.

Beverage intake was analyzed as a categorical variable divided into three categories. Analyses compared each category of intake (" $\geq 1$  serving per month to < 2 servings per week"; " $\geq 2$  servings per week to < 1 serving per day"; and " $\geq 1$  serving per day") to the referent group, which is the lowest category of intake ("Never to < 1 serving per month").

#### Validation of Exposure

The Harvard FFQ has been extensively validated for use in U.S. women (59). Mean nutrient intakes estimated by four one-week diet records completed over one year were compared to those estimated from FFQ's administered one year apart. Diet records are intended to be completed each time a food item is consumed over a one-week period and are believed to be the most valid method of dietary reporting, because they minimize recall bias, or participants' ability to forget what they have eaten. The range of intraclass correlations for the four diet records ranged from 0.41 for vitamin A to 0.79 for vitamin B6, and for the two FFQs the intraclass correlations ranged from 0.49 for vitamin A to 0.71 for sucrose. This indicates a similarity between these methods in terms of reproducibility. Participants in the lowest quintile of total energy intake as computed from the diet records were in the lowest one or two quintiles of total energy intake computed from the FFQ 74% of the time. Participants in the highest quintile of total energy intake from the diet records were in the highest one of two quintiles from the FFQ 74% of the time. Participants in the highest quintile of total energy intake from the diet records were in the highest one of two quintiles from the FFQ 77% of the time (59). This indicates that the FFQ is relatively valid over one year in comparison to four one-week diet records.

Intraclass correlations for beverages measured on the two FFQs completed one year apart ranged from 0.24 for fruit punch to 0.93 for beer (60). Correlations between diet records and the FFQs ranged from 0.46 for high-energy drinks to 0.83 for coffee (61). This indicates that reproducibility and validity is high for beverages over a one year period.

#### Outcome Assessment

Obesity can be defined as an accumulation of excess adipose tissue. Due to its ease of use and cost-effectiveness, BMI has been used as a primary measurement of adiposity in the clinical setting. Other methods are available for more specific body composition assessment. Waist circumference provides an easy measure of central adiposity, and has been shown to correlate more strongly with all-cause mortality than BMI (42). DXA is able to calculate an individual's percent body fat and differentiate between fat mass and fat-free mass. This measure is highly predictive of metabolic

syndrome, and captures information not ascertainable by measuring BMI or waist circumference (62).

In the current study, body composition was calculated via these three measures: BMI, waist circumference, and percent body fat. The two examiners directly measured waist circumference at the clinic visit. Weight and height were directly measured by the examiners at the clinic visit and used to calculate BMI (kg/m<sup>2</sup>). Scales were balanced routinely to measure weight, and height was measured using a stadiometer. We calculated percent body fat directly by DXA (total fat mass/total body mass) using the total body scan mode on a narrow angle fan GE Lunar Prodigy scanner (GE Lunar Corp., Madison, WI). We performed daily calibrations of the DXA using the standard calibration phantom provided by the manufacturer. We analyzed all scans using the manufacturer's enCORE 2002 software package, version 6.80.002. The *in vivo* precision of this machine ranges from 1.0% to 2.2% for BMC (63), and from 1.1% to 2.7% for lean mass and 2.6% to 3.9% for fat mass (63-65). Two examiners (SZ and BT) performed and analyzed all scans (66).

All measures of body composition were analyzed as continuous variables. BMI was categorized into underweight (<18.5 kg/m<sup>2</sup>), normal weight (18.5-24.9 kg/m<sup>2</sup>), and overweight ( $\geq$ 25 kg/m<sup>2</sup>) (67). Waist circumference was categorized as normal weight (<80 cm) and overweight ( $\geq$ 80 cm) according to World Health Organization guidelines for women (68). Percent body fat will be categorized as low (<21% body fat), normal weight (21-33% body fat), and high (>33% body fat) (69).

#### Validation of Outcome

BMI and waist circumference have been shown to be associated with risk of mortality in women (70, 71). In the Nurses' Health Study, women in the highest category of BMI had 2.2 times the risk of all-cause mortality relative to the women in the lowest category (95% CI 1.4, 3.4; p<0.001)(70). After adjusting for BMI, waist circumference was independently associated with all-cause mortality (71). Among women in a large Danish cohort, a 10% increase in waist circumference was associated with a 30% increase in risk of all-cause mortality (95% CI 1.17-1.44) (71). Thus far, percent body fat has shown an inconsistent association with morbidity and mortality due to a lack of prospective studies (69). DXA has been shown to be effective in accurately quantifying adipose tissue mass and location (72). The DXA scan has been used to quantify body shape as barrel-shaped versus non-barrel-shaped and, in a Swedish cohort, barrel-shaped individuals (73). This indicates that BMI, waist circumference, and percent body fat are all capable of independently predicting all-cause mortality.

According to the Third National Health and Nutrition Examination Survey, height and weight measured directly by an investigator is considered the gold standard for classification of overweight status (74). Self-reported height and weight is less accurately reported with increasing age. Among a group of children and adolescents, direct waist circumference measurement was highly correlated with central adiposity measured by DXA (75). This indicates that direct measurement of waist circumference is a valid obesity assessment tool.

#### Covariate Assessment

Factors were evaluated as possible confounders if they had been shown to be associated with body composition and beverage consumption in prior studies. Additional dietary confounders were assessed by the study FFQ. These included total energy, fiber, alcohol, milk, glycemic index, caffeine, and multivitamin use (20, 26, 47, 76, 77). We collected information on age, lifestyle, and demographic factors by self-reported questionnaire, including current smoking status, and physical activity. To measure physical activity, we asked participants to report the time they spent each week engaged in specific activities including walking, jogging, running, bicycling, aerobics/dancing, tennis/racket sports, swimming, yoga/Pilates, and weight training. These questions were based on those used in the Nurses' Health Study II and have been previously validated in that population (78). We then calculated total MET-hours per week of activity using the method of Ainsworth et al. (79).

#### **Statistical Analysis**

Specific Aim #1: Using a cross-sectional study design, we propose to examine the association between consumption of various beverage types and body composition in college-aged women. We calculated the mean (SD) of continuous demographic characteristics and the number (%) of categorical demographic characteristics of the study population (Table 1). We calculated the number and percent of participants within each category of beverage intake (Table 2) as well as mean (SD) beverage intake for each of the three beverage types (Table 3). We calculated the mean, median, interquartile range (IQR) of BMI, waist circumference, and percent body fat along with number and

percent of participants who are underweight, normal weight, and overweight as defined by BMI, waist circumference, and percent body fat Table 4).

#### **Bivariate Analysis**

Confounding by continuous covariates was assessed by comparing means (SD) across categories of sugar-sweetened beverage (Table 5), diet drink (Table 6), and fruit juice (Table 7) using an ANOVA procedure to compare groups. The same assessment was repeated across categories of BMI (Table 8), waist circumference (Table 9), and percent body fat (Table 10). Categorical covariates were assessed as confounders by comparing number and percent within all exposure (Tables 5-7) and outcome (Tables 8-10) categories, using a chi-square test to compare groups. For cross-tabulations with small cell frequencies (n<5), Fisher's Exact Test was used to compare groups.

We compared the mean BMI (Table 11), waist circumference (Table 12), and percent body fat (Table 13) of women in different categories of beverage intake. We compared these distributions using chi-square tests and Fisher's Exact Tests for crosstabulations with small cell frequencies. Unadjusted odds ratios and 95% confidence intervals were calculated to show the crude association between categorical beverage consumption and overweight across BMI (Tables 14), waist circumference (Table 15), and percent body fat (Table 16).

#### Multivariable Analysis

Multiple logistic regression was used to model the relation between beverage intake and overweight as assessed by BMI (Table 17), waist circumference (Table 18),

and percent body fat (Table 19). Covariates whose addition to the regression model resulted in a 15% or greater change in the coefficient for beverage intake was considered confounding factors and included in the final model. Prior studies have shown total energy consumption to be strongly associated with body composition, but this has been considered a possible mechanistic explanation for the proposed association. To address this covariate as a possible confounder and mechanistic explanation, two models were used to assess the association between beverage consumption and body composition: one will include total energy and one will not.

#### CHAPTER VII

#### HUMAN SUBJECT PROTECTION

The University of Massachusetts Vitamin D Status Study was approved by the University of Massachusetts Human Subjects Review Committee. All participants are required to sign an informed consent document explaining the purpose of the study and the procedures to obtain data including the fasting blood sample, urine sample, anthropometric measurement, and lifestyle and diet questionnaires. The document contains information on the analyses that will be conducted on the biologic samples. The possible risks and discomfort associated with all procedures are explicitly disclosed.

Participants are under no obligation to participate and may withdraw from the study at any time. All information is kept confidential and will not be sold or shared with anyone outside of the study. Any published data will have identifying information removed. Investigators are able to link participants' names with their personal data for the sole purpose of providing them with their test results.

The benefits of participation include the results from the blood sugar test, DXA scan including body fat distribution and bone density, diet analysis including nutrient content, and blood and urine nutrient analysis. Participants may benefit from the knowledge that they are improving scientific understanding of diet and its impact on health.

## CHAPTER VIII

## PERMISSION TO ACCESS DATA

All investigators have completed human subject certification prior to accessing

any data from this study. Access was granted by the two principal investigators.

#### CHAPTER IX

#### RESULTS

The average age of the population was 21.6 years (SD=3.1) (Table 1).

Participants consumed 2198 kcal (SD=825) of energy per day on average, expended roughly 177 METs (SD=69) per week, and drank one daily serving (SD=1.3) of coffee or tea. Most participants were white (86%) and currently enrolled in college (79%). Study participants were distributed evenly across juice (p=0.24) consumption categories, but were more likely to be moderate consumers ( $\geq$ 1 serving per month to < 2 servings per week) of sugar-sweetened beverages and light consumers (0 servings per month to <1 serving per month) of diet drinks (Table 2). Participants tended to consume more juice than other beverages (Table 3).

The majority of participants in our study were classified as normal weight based on BMI (73%) and waist circumference (65%) (Table 4). According to percent body fat, participants were evenly distributed across normal weight (46%) and overweight categories (42%). Few participants were underweight (BMI=3%, percent body fat=12%), we combined these groups with the normal weight women for analyses.

Compared to light consumers, heavy consumers of sugar-sweetened beverages (>2 servings per week) tended to be younger, consumed more calories, consumed less fiber, and had a higher glycemic index (Table 5). Heavy consumers of diet drinks tended to be younger, and consumed more alcohol compared to light consumers (Table 6). Heavy consumers of juice tended to consume more calories, drank more alcohol, and had a higher glycemic index compared to light consumers (Table 7).

We did not find that participant characteristics such as age, activity, or total energy intake varied across categories of BMI (Table 8). Participants in the overweight group, classified by waist circumference ( $\geq$ 80 cm), drank more alcohol compared with the normal weight group (Table 9). Overweight women, classified by percent body fat ( $\geq$ 33%), reported fewer METs per week of physical activity, consumed less fiber, and drank fewer daily servings of coffee and tea compared to normal weight women (Table 10).

In bivariate analyses, sugar-sweetened beverage and juice consumption were not associated with body composition (Tables 11-13). Diet drink consumption, however, was positively associated with overweight status in analyses using all three body composition assessments.

In univariable and age-adjusted logistic regression analyses there was no association between sugar-sweetened beverage and juice consumption and overweight status assessed by BMI (Table 14). Heavy diet drink consumers were 2.5 (95% CI 1.20, 5.07) times more likely to be overweight based on BMI compared to light diet drink consumers, and this association persisted after age adjustment.

In univariable and age-adjusted logistic regression analyses there was no association between sugar-sweetened beverage and juice consumption and overweight status assessed by waist circumference (Table 15). Moderate diet drink consumers were 2.2 (95% CI 1.12, 4.18) times more likely to be overweight based on waist circumference compared to light diet drink consumers, and this association persisted after age adjustment. Heavy diet drink consumers were 2.7 (95% CI 1.40, 5.25) times more likely

to be overweight based on waist circumference compared to light diet drink consumers, and this association, likewise, persisted after age adjustment.

In univariable and age-adjusted logistic regression analyses there was no association between sugar-sweetened beverage and juice consumption with overweight status as assessed by percent body fat (Table 16). Moderate diet drink consumers were 2.2 (95% CI 1.17, 4.12) times more likely to be overweight based on percent body fat compared to light diet drink consumers, and this association persisted after age adjustment. Heavy diet drink consumers were 2.1 (95% CI 1.12, 4.03) times more likely to be overweight based on percent body fat compared to light diet drink consumers, and this association, similarly, persisted after age adjustment.

In multivariable models there was no association between sugar-sweetened beverage and juice consumption and overweight status as assessed by BMI (Table 17). The association between diet drink consumption and overweight status was strengthened after adjustment for confounders in the two models (with and without energy adjustment) compared to the age-adjusted model. Heavy diet drink consumers were 2.9 (95% CI 1.34, 6.21; *p*-trend=.02) times more likely to be overweight based on percent body fat compared to light diet drink consumers.

In multivariable models there was no association between sugar-sweetened beverage and juice consumption and overweight status assessed by waist circumference (Table 18). The association between diet drink consumption and overweight status was strengthened after adjustment for confounders in the two models compared to the ageadjusted model. Moderate diet drink consumers were 2.3 (95% CI 1.14, 4.62) times more likely to be overweight based on waist circumference compared to light diet drink

consumers. Heavy diet drink consumers were 3.1 (95% CI 1.56, 6.35; *p*-trend=.01) times more likely to be overweight based on waist circumference compared to light diet drink consumers.

In multivariable models there was no association between sugar-sweetened beverage and juice consumption and overweight status assessed by percent body fat (Table 19). The association between diet drink consumption and overweight status was strengthened after adjustment for confounders in the two models compared to the ageadjusted model. Moderate diet drink consumers were 2.9 (95% CI 1.43, 5.76) times more likely to be overweight based on waist circumference compared to light diet drink consumers. Heavy diet drink consumers were 2.9 (95% CI 1.42, 5.77; *p*-trend=.07) times more likely to be overweight based on waist circumference compared to light diet drink consumers.

#### CHAPTER X

## DISCUSSION

In this cross-sectional study among college-aged women, we found a two- to three-fold increase in odds of being overweight among heavy consumers of diet drinks compared to light consumers after adjusting for age, daily coffee and tea intake, physical activity, and total energy. This association exhibited a dose-response relationship, as odds of overweight increased linearly with increasing consumption of diet drinks. This linear trend was observed when we classified overweight based on BMI and waist circumference, and was nearly significant when overweight was based on percent body fat. No appreciable increase in odds of overweight was observed in relation to increasing consumption of sugar-sweetened beverages or juice.

Our findings are not consistent with the majority of literature on beverage consumption and body composition. Unlike prior studies (20, 22, 25, 26, 38, 53, 54), we found no association between sugar-sweetened beverage consumption or juice consumption and overweight. Most prospective studies, such as those conducted by Ludwig *et al.* (25) and Schulze and colleagues (26), have observed increased risk of obesity with increased sugar-sweetened beverage and juice consumption. Our study is consistent with the findings of Berkey and colleagues (20), who observed no association between sugar-sweetened beverage consumption and BMI in a prospective cohort.

Further, the literature on diet drink consumption and body consumption is conflicting, but has generally observed an inverse (26) or null (20, 39) association in prospective studies. The study by Schulze and colleagues (26) was one of the largest to assess diet drink consumption and obesity prospectively and observed an inverse

association between diet drink consumption and body composition. Women in this study who increased their diet drink consumption gained significantly less weight (+1.59 kg) over four years of observation compared to women who decreased their diet drink consumption (+4.25 kg, p<0.001). Our findings were not consistent with this outcome, since we found a 2.88 times increased odds (95% CI 1.34, 6.21) of being overweight, as assessed by BMI, for heavy diet drink consumers compared to light consumers.

#### Non-differential Misclassification of Exposure

The most significant limitation to our study is non-differential misclassification of beverage exposure. Our exposure assessment tool was the food frequency questionnaire. Although validated, the tool relies on memory, comprehension of serving sizes, and ability to estimate intake over a two-month period. This could have led to inaccuracies in measuring beverage intake. The error, however, should not have differed in a systematic way by body composition. Therefore, the misclassification would have equally distorted the true exposure in overweight cases and non-cases, which would have caused an attenuation of our study results. To minimize the impact of this misclassification our analyses grouped exposure status categorically. Classifying participants into categories creates a relative measure that is more accurate than the absolute values and will limit misclassification error.

## Non-differential Misclassification of Outcome

Non-differential misclassification of our outcome, body composition, was unlikely in this study. We used trained investigators to directly measure height, weight, and waist circumference. Participants' percent body fat was measured by DXA scan. Percent body fat by DXA is highly correlated with percent body fat by computed tomography, the gold standard, for measuring visceral adipose tissue volume (r=0.72) and deep subcutaneous adipose tissue volume (r=0.75) (80). If present, it could be that measurement error existed for all participants, irrespective of their beverage consumption status. This would lead to an attenuation of our observed results. However, this problem is unlikely to have impacted our results our DXA instruments were calibrated regularly.

## Selection Bias

Selection bias could have been present if those who were light beverage consumers and had low body fat were more motivated to participate in our study than those with other patterns of beverage consumption and body composition. In this case, both exposure and outcome would influence participation and this would have resulted in a moderate increase in our estimate of risk beyond the true association. Or, if those who were high beverage consumers and had low body fat were more motivated to participate in our study than those with other patterns of beverage consumption and body composition, then our estimate would be moderately underestimated. Because beverage consumption was not the primary exposure under investigation in this study, we assume that beverage consumption would not be strongly associated with participation, and this bias should not be present.

## Information Bias

Information bias could occur if overweight participants recalled or misrepresented their beverage intake in a systematically different way than normal weight participants. If overweight participants tended to underreport sugar-sweetened beverage intake, this could have lead to the null findings even if an association were present. If these participants tended to overreport their diet drink intake, this could have led to the strong positive association we observed by overestimating the true association. We expect this was not likely, because participants' dietary data was submitted anonymously to limit distortion due to any preconceived social stigma related to beverage consumption and body composition. Further, we tried to minimize the residual impact on our results from systematic underreporting or overreporting by sorting beverage intake into broad categories to capture any variation.

## Confounding

In our model we controlled for dietary and lifestyle factors, but we did not have information on participants' level of daily stress. Stress could have acted as a confounding factor as it has been shown to be positively associated with consumption of carbohydrate-rich snacks (81) such as sugar-sweetened beverages or juice and chronic stress is negatively associated with overweight status through mechanisms related to corticosteroid mobilization of fat stores (82). This would have lead to an underestimate of the relative risk. If the distribution of this and other unmeasured confounding factors is unrelated to beverage consumption and body composition, this confounding should not dramatically impact our results. In addition, residual confounding is possible after multivariable adjustment if measured confounders were inaccurately quantified.

# Generalizability

As volunteers in a health related study at a state university, participants were predominantly white and enrolled in higher education, which may indicated higher socioeconomic status, and greater access to health care resources than other American women. However, it is unlikely that the biological mechanisms proposed for the association between beverage intake and body composition would act differently in the general population. Our results may not be generalizable to the very young or old, as these groups may differ significantly in their beverage metabolism.

## Temporality

One major concern for the interpretation of a cross-sectional study is assessing temporality between the exposure and the outcome. Since we measured beverage intake and body composition at the same time, it is impossible to determine whether beverage intake occurred prior to the observed weight status and acted as the causal agent for that weight status. It is conceivable that participants may have reached their observed weight and subsequently changed their pattern of beverage consumption or may be trying to achieve their ideal weight by switching to drinking diet drinks instead of full-calorie drinks. This limitation precludes us from drawing conclusions about causality.

In this case, the significant increase in odds of obesity among heavy consumers of diet drinks compared to light consumers is believed to be due to overweight women choosing diet drinks in order to lose weight. If this reverse causality was occurring, it would resolve the inconsistency between our findings and the prior literature. In

addition, the dose-response relationship between diet drink consumption and odds of obesity was observed when overweight status was based on BMI and waist circumference, but not when it was based on percent body fat. This supports the theorized explanation, because women may have been motivated by visible, aesthetic factors like BMI and waist circumference. Women likely did not know their percent body fat, since it is not visually observable and not strongly correlated with BMI in overweight women in our population, and thus, percent body fat would have been less likely to motivate consuming diet drinks as a weight loss strategy.

# Survival Bias

A final concern with cross-sectional studies is the potential for survival bias. This could have occurred if individuals with high beverage intake and overweight status were more likely to die prior to our study, making them unavailable for participation in our study. This would have biased the results of our study to the null relative to the true association. However, overweight status is unlikely to result in death in the age group we were investigating, so survival bias should not have impacted our results.

## Conclusion

Our findings indicate that diet drink consumption is strongly associated with increased odds of obesity. In contrast to prior studies (20, 22, 25, 26, 38, 53, 54), sugar-sweetened beverages and juice were not associated with increased odds of obesity. We believe that these results may be due to reverse causality which we were unable to assess due to the cross-sectional design of our study. The main strength of our study was the

use of multiple measure of obesity, and we recommend using these multiple measures prospectively to look at the association in the future.

Vitamin D Status Study, 2006-2010	
Continuous Variable	Mean (SD)
Age (y)	21.6 (3.0)
Sugar-sweetened Beverage (serving/d)	0.2 (0.5)
Diet Drink (serving/d)	0.3 (1.3)
Juice (serving/d)	0.6 (1.0)
Milk (serving/d)	1.1 (2.0)
Coffee/Tea (serving/d)	1.0 (1.3)
Body Mass Index $(kg/m^2)$	23.0 (3.2)
Waist Circumference (cm)	78.1 (8.6)
Percent Body Fat (%)	· · · · ·
Activity (METs/wk)	32 (7.8) 176 (69)
Total Energy (kcal)	2198 (825)
Fiber (g/d)	30.6 (17.9)
Alcohol (g/d)	6.5 (8.3)
Glycemic Index	50.7 (4.9)
Categorical Variable	N (%)
Age	
18-22	191 (81)
23-26	22 (9)
27-30	24 (10)
Race	202 (86)
White Other	203 (86)
Education	34 (14)
High School	4 (2)
Some College	187 (79)
College	9 (4)
Some Grad	24 (10)
Grad	13 (6)
Sugar-sweetened Beverage	- (-)
Light Consumer (<1 serving/mo)	89 (38)
Moderate Consumer (>1/mo, <2/wk)	97 (41)
Heavy Consumer (>2/wk)	51 (22)
Diet Drink	
Light Consumer (<1 serving/mo)	116 (49)
Moderate Consumer (>1/mo, <2/wk)	62 (26)
Heavy Consumer (>2/wk)	59 (25)
Juice	
Light Consumer (<1 serving/mo)	58 (24)
Moderate Consumer (>1/mo, <2/wk)	68 (29)
Heavy Consumer (>2/wk, <1/d)	64 (27)
Very Heavy Consumer (>1/d)	47 (20)
Milk	45 (10)
Light Consumer	45 (19)
Moderate Consumer	55 (23) 71 (20)
Heavy Consumer	71 (30)
Very Heavy Consumer (Continued on next page)	66 (28)
(continued on next page)	

 Table 1. Descriptive characteristics of participants in University of Massachusetts

 Vitamin D Status Study, 2006-2010

Body Mass Index (kg/m <sup>2</sup> )	
Underweight (<18.5)	8 (3)
Normal weight (18.5-24.9)	174 (73)
Overweight $(\geq 25)$	55 (23)
Waist Circumference	
Normal weight (< 80 cm)	154 (65)
Overweight ( $\geq 80 \text{ cm}$ )	83 (35)
Percent Body Fat	
Underweight (<21)	29 (12)
Normal weight (21-33)	108 (46)
Overweight (>33)	100 (42)
Multivitamin Use	
Yes	130 (55)
No	106 (45)
Current Smoker	
Yes	226 (95)
No	11 (5)

Table 2. Distribution of intake across beverage categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

			Heavy	
	Light	Moderate	Consumer,	
	Consumer,	Consumer,	> 2 per week	Very Heavy
	Never to $< 1$	$\geq 1$ per month to	to	Consumer,
	per month	2 per week,	1 per day,	$\geq 1$ per day
	N (%)	N (%)	N (%)	[Juice Only]
Sugar-sweetened beverages	89 (38)	97 (41)	51 (22)	
Diet Drinks	116 (49)	62 (26)	59 (25)	
Fruit Juice	58 (24)	68 (29)	64 (27)	47 (20)

Table 3. Mean and median consumption across beverage categories among collegeaged women in the UMass Vitamin D Status Study, 2006-2010

	Mean (SD), servings/day	Median, Servings/day
Sugar-sweetened beverages	0.22 (0.5)	0.07
Diet Drinks	0.32 (1.3)	0.07
Fruit Juice	0.58 (1.0)	0.21

Table 4. Population distribution of body composition outcomes among college-aged women in the UMass Vitamin D Status Study, 2006-2010

	Mean (SD)	Median	Interquartile Range (IQR)	Underweight, N (%)	Normal weight, N (%)	Overweight, $N(\%)$
BMI (kg/m <sup>2</sup> )*	23.0 (3.2)	22.7	20.6 - 24.9	8 (3)	174 (73)	55 (23)
WC (cm)**	78.1 (8.6)	77.5	71.1 - 83.8	N/A	154 (65)	83 (35)
BF%***	32.0 (7.8)	31.8	26.3 - 37.8	29 (12)	108 (46)	100 (42)

\* Underweight = BMI <18.5 kg/m<sup>2</sup>; Normal weight = BMI 18.5-24.9 kg/m<sup>2</sup>; Overweight = BMI  $\ge 25$  kg/m<sup>2</sup>

\*\* Normal weight = non-pregnant WC < 80 cm; Overweight = non-pregnant WC  $\ge$  80 cm

\*\*\* Among white women: Underweight = < 21%; Normal weight = 21-32.9%; Overweight =  $\ge 33\%$ 

Table 5. Distribution of covariates according to sugar-sweetened beverage consumption category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

		Moderate	Heavy	
	Light Consumer,	Consumer,	Consumer,	
	Never to $< 1$ per	$\geq 1$ per month to	> 2 per week to	
	month	2 per week	1 per day	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	Mean (SD)	
Age (y)	22.2 (3.0)	21.4 (3.1)	20.7 (2.8)	0.02
Activity (MET/wk)	180.5 (63.9)	172.5 (66.5)	177.5 (83.3)	0.74
Total Energy (kcal)	2014 (830)	2161 (731)	2589 (870)	0.0003
Alcohol (g/d)	5.5 (6.2)	7.0 (7.4)	7.4 (12.2)	0.33
Milk (serving/d)	1.0 (2.1)	1.2 (2.2)	1.0 (1.5)	0.74
Fiber (g/d)	34.8 (22.6)	28.6 (14.9)	27.1 (11.7)	0.02
Coffee/Tea (serv/d)	1.1 (1.5)	1.0 (1.1)	0.9 (1.3)	0.65
Glycemic Index	48.7 (6.2)	51.3 (3.3)	52.9 (3.4)	< 0.0001
	N (%)	N (%)	N (%)	
Age (y)				
18-22	64 (72)	80 (82)	47 (92)	0.008
23-26	15 (17)	7 (7)	0 (0)	
27-30	10 (11)	10 (10)	4 (8)	
Milk Intake				
Light	25 (28)	16 (16)	4 (8)	0.08
Moderate	20 (22)	21 (22)	14 (27)	
Heavy	21 (24)	30 (31)	20 (39)	
Very Heavy	23 (26)	30 (31)	13 (25)	
Current Smoker				
Yes	1 (1)	8 (8)	2 (4)	0.06
No	88 (99)	89 (92)	49 (96)	
Multivitamin Use				
Yes	39 (44)	47 (48)	20 (39)	0.56
$\frac{No}{1}$ n values for continuous t	49 (56)	50 (52)	31 (61)	

<sup>1</sup> p-values for continuous measures from ANOVA test, p-values for categorical measures from Chi-square test or Fisher's exact test

		Moderate		
	Light Consumer,	Consumer,	Heavy Consumer,	
	Never to $< 1$ per	$\geq 1$ per month to	> 2 per week to	
	month	2 per week	1 per day	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	Mean (SD)	
Age (y)	22.1 (3.4)	21.2 (2.7)	21.0 (2.7)	0.04
Activity (MET/wk)	167.5 (68.8)	189.1 (71.8)	181.5 (66.6)	0.12
Total Energy (kcal)	2204 (839)	2203 (806)	2180 (832)	0.98
Alcohol (g/d)	5.3 (6.8)	9.5 (11.6)	5.8 (5.7)	0.004
Milk (serving/d)	1.2 (2.3)	1.0 (1.7)	1.0 (1.9)	0.83
Fiber (g/d)	31.2 (17.5)	28.9 (16.7)	31.1 (20.0)	0.70
Coffee/Tea (serv/d)	0.9 (1.2)	1.1 (1.2)	1.3 (1.5)	0.18
Glycemic Index	50.4 (5.8)	51.1 (3.4)	50.6 (4.3)	0.69
	N (%)	N (%)	N (%)	
Age (y)				
18-22	88 (76)	52 (84)	51 (86)	0.43
23-26	12 (10)	6 (10)	4 (7)	
27-30	16 (14)	4 (6)	4 (7)	
Milk Intake				
Light	28 (24)	10 (16)	7 (12)	0.24
Moderate	22 (19)	13 (21)	20 (34)	
Heavy	34 (29)	21 (34)	16 (27)	
Very Heavy	32 (28)	18 (29)	16 (27)	
Current Smoker				
Yes	6 (5)	2 (3)	3 (5)	0.85
No	110 (95)	60 (97)	56 (95)	
Multivitamin Use				
Yes	49 (43)	32 (52)	25 (42)	0.47
No	66 (57)	30 (48)	34 (58)	

Table 6. Distribution of covariates according to diet drink consumption category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

 $rac{1000007}{1}$  p-values for continuous measures from ANOVA test, p-values for categorical measures from Chi-square test or Fisher's exact test

		Moderate	Heavy		
	Light	Consumer,	Consumer,		
	Consumer,	$\geq 1 \text{ per}$	> 2 per week	Very Heavy	
	Never to $< 1$	month to	to	Consumer,	• 1
	per month	2 per week	1 per day	$\geq 1$ per day	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	
Age (y)	21.9 (2.8)	21.6 (2.9)	21.6 (3.2)	21.1 (3.4)	0.71
Activity (MET/wk)	184.5 (64.2)	163.9 (65.1)	180.0 (74.0)	180.2 (74.4)	0.36
Total Energy (kcal)	2256 (968)	1927 (738)	2167 (679)	2560 (812)	0.0007
Alcohol (g/d)	9.0 (11.4)	5.7 (6.6)	6.6 (8.2)	4.6 (5.0)	0.04
Milk (serving/d)	1.0 (2.0)	1.4 (2.7)	0.8 (1.6)	1.0 (1.5)	0.50
Fiber (g/d)	34.5 (21.3)	28.7 (17.4)	28.5 (17.2)	31.5 (14.3)	0.22
Coffee/Tea (serving/d)	1.2 (1.6)	1.0 (1.2)	1.2 (1.4)	0.6 (0.6)	0.09
Glycemic Index	48.1 (7.1)	50.9 (3.7)	51.4 (3.4)	52.5 (3.5)	< 0.0001
	N (%)	N (%)	N (%)	N (%)	
Age (y)					
18-22	44 (76)	55 (81)	51 (80)	41 (87)	0.45
23-26	9 (16)	6 (9)	6 (9)	1 (2)	
27-30	5 (9)	7 (10)	7 (11)	5 (11)	
Milk					
Light	18 (31)	9 (13)	11 (17)	7 (15)	0.23
Moderate	14 (24)	17 (25)	14 (22)	10 (21)	
Heavy	10 (17)	22 (32)	24 (38)	15 (32)	
Very Heavy	16 (28)	20 (29)	15 (23)	15 (32)	
Current Smoker					
Yes	4 (7)	3 (4)	3 (5)	1 (2)	0.74
No	54 (93)	65 (96)	61 (95)	46 (98)	
Multivitamin Use		. /	. /		
Yes	29 (51)	30 (44)	30 (47)	17 (36)	0.50
No	28 (49)	38 (56)	34 (53)	30 (64)	

Table 7. Distribution of covariates according to fruit juice consumption category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup> p-values for continuous measures from ANOVA test, p-values for categorical measures from Chi-square test or Fisher's exact test

	Underweight (< 18.5 kg/m <sup>2</sup> )	Normal weight (18.5-24.9 kg/m <sup>2</sup> )	Overweight $(\geq 25)$	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	Mean (SD)	
Age (y)	21.2 (3.1)	21.5 (3.0)	21.7 (3.2)	0.91
Activity (MET/wk)	147.7 (41.1)	174.5 (66.1)	187.5 (81.2)	0.24
Total Energy (kcal)	1970 (885)	2201 (820)	2222 (843)	0.72
Alcohol (g/d)	5.6 (5.0)	6.4 (7.5)	7.1 (10.9)	0.80
Milk (serving/d)	0.7 (0.9)	1.0 (2.1)	1.2 (2.1)	0.70
Fiber (g/d)	28.5 (14.6)	31.0 (19.1)	29.6 (14.4)	0.82
Coffee/Tea (serving/d)	1.0 (1.1)	1.1 (1.4)	0.9 (1.1)	0.62
Glycemic Index	50.7 (3.7)	50.6 (3.9)	50.7 (7.4)	0.99
	N (%)	N (%)	N (%)	
Age (y)				
18-22	7 (88)	142 (82)	42 (76)	0.75
23-26	0 (0)	15 (9)	7 (13)	
27-30	1 (13)	17 (10)	6 (11)	
Milk				
Light	2 (25)	34 (20)	9 (16)	0.96
Moderate	1 (13)	39 (22)	15 (27)	
Heavy	3 (38)	53 (30)	15 (27)	
Very Heavy	2 (25)	48 (28)	16 (29)	
Current Smoker				
Yes	0 (0)	11 (6)	0 (0)	0.13
No	8 (100)	163 (94)	55 (100)	
Multivitamin Use				
Yes	4 (50)	79 (46)	23 (42)	0.81
No	4 (50)	94 (54)	32 (58)	

Table 8. Distribution of covariates according to BMI category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-values for continuous measures from ANOVA test, *p*-values for categorical measures from Chi-square test or Fisher's exact test

	Normal weight (< 80 cm)	Overweight $(\geq 80 \text{ cm})$	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	
Age (y)	21.5 (3.0)	21.6 (3.2)	0.80
Activity (MET/wk)	172.7 (64.2)	183.5 (77.9)	0.26
Total Energy (kcal)	2147 (790)	2293 (883)	0.19
Alcohol (g/d)	5.8 (6.9)	8.0 (10.3)	0.05
Milk (serving/d)	1.1 (2.2)	1.0 (1.7)	0.67
Fiber (g/d)	31.0 (18.4)	29.9 (17.0)	0.66
Coffee/Tea (serving/d)	1.1 (1.2)	1.0 (1.4)	0.71
Glycemic Index	50.5 (4.0)	51.0 (6.3)	0.40
	N (%)	N (%)	
Age (y)			
18-22	126 (82)	65 (78)	0.75
23-26	14 (9)	8 (10)	
27-30	14 (9)	10 (12)	
Milk Intake			
Light	33 (21)	12 (14)	0.57
Moderate	35 (23)	20 (24)	
Heavy	43 (28)	28 (34)	
Very Heavy	43 (28)	23 (28)	
Current Smoker			
Yes	10 (7)	1 (1)	0.10
No	144 (94)	82 (99)	
Multivitamin Use			
Yes	74 (48)	32 (39)	0.15
No	79 (52)	51 (62)	

Table 9. Distribution of covariates according to waist circumference category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

 $r_{p}$ -values for continuous measures from ANOVA test, *p*-values for categorical measures from Chi-square test or Fisher's exact test

	Underweight (< 21%)	Normal weight (21-33%)	Overweight (> 33%)	<i>p</i> -value <sup>1</sup>
	Mean (SD)	Mean (SD)	Mean (SD)	
Age (y)	22.4 (3.6)	21.4 (2.9)	21.5 (3.1)	0.26
Activity (MET/wk)	168.1 (70.7)	190.5 (68.9)	164.3 (67.5)	0.02
Total Energy (kcal)	2011 (872)	2300 (833)	2143 (796)	0.17
Alcohol (g/d)	5.2 (5.5)	5.6 (7.0)	7.9 (10.0)	0.09
Milk (serving/d)	0.4 (0.5)	1.3 (2.3)	1.0 (2.0)	0.07
Fiber (g/d)	29.8 (16.8)	34.0 (21.5)	27.2 (12.7)	0.02
Coffee/Tea (serving/d)	1.3 (1.6)	1.2 (1.5)	0.8 (0.9)	0.05
Glycemic Index	51.3 (4.3)	50.3 (4.0)	50.9 (5.9)	0.53
	N (%)	N (%)	N (%)	
Age (y)				
18-22	19 (66)	91 (84)	81 (81)	0.25
23-26	5 (17)	8 (7)	9 (9)	
27-30	5 (17)	9 (8)	10 (10)	
Milk Intake				
Light	9 (31)	23 (21)	13 (13)	0.04
Moderate	6 (21)	20 (19)	29 (29)	
Heavy	10 (34)	27 (25)	34 (34)	
Very Heavy	4 (14)	38 (35)	24 (24)	
Current Smoker				
Yes	1 (4)	6 (6)	4 (4)	0.91
No	28 (97)	102 (94)	96 (96)	
Multivitamin Use				
Yes	16 (55)	49 (45)	41 (41)	0.42
No	13 (45)	59 (55)	58 (59)	

Table 10. Distribution of covariates according to percent body fat category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

1015 (45)59 (55)58 (59) $^{1}p$ -values for continuous measures from ANOVA test, *p*-values for categorical measures from<br/>Chi-square test or Fisher's exact test

	Normal weight (< 25 kg/m <sup>2</sup> )	Overweight $(\geq 25 \text{ kg/m}^2)$	<i>p</i> -value <sup>1</sup>
	N (%)	N (%)	
Sugar-sweetened beverages			
Light	68 (37)	21 (38)	0.87
Moderate	76 (42)	21 (38)	
Heavy	38 (21)	13 (24)	
Diet Drinks			
Light	96 (53)	20 (36)	0.05
Moderate	47 (26)	15 (27)	
Heavy	39 (21)	20 (36)	
Fruit Juice			
Light	47 (26)	11 (20)	0.21
Moderate	46 (25)	22 (40)	
Heavy	51 (28)	13 (24)	
Very Heavy	38 (21)	9 (16)	

Table 11. Distribution of beverage intake according to BMI category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-values from Chi-square test or Fisher's exact test

	Normal weight (< 80 cm)	Overweight $(\geq 80 \text{ cm})$	<i>p</i> -value <sup>1</sup>	
	N (%)	N (%)		
Sugar-sweetened beverages				
Light	59 (38)	30 (36)	0.58	
Moderate	65 (42)	32 (39)		
Heavy	30 (19)	21 (25)		
Diet Drinks				
Light	87 (56)	29 (35)	0.005	
Moderate	36 (23)	26 (31)		
Heavy	31 (20)	28 (34)		
Fruit Juice				
Light	40 (26)	18 (22)	0.64	
Moderate	41 (27)	27 (33)		
Heavy	44 (29)	20 (24)		
Very Heavy	29 (19)	18 (22)		

Table 12. Distribution of beverage intake according to waist circumference category among college-aged women in the UMass Vitamin D Status Study, 2006-2010

p-values from Chi-square test or Fisher's exact test

	Normal weight (< 33%)	Overweight $(\geq 33\%)$	<i>p</i> -value <sup>1</sup>
	N (%)	N (%)	
Sugar-sweetened beverages			
Light	52 (38)	37 (37)	0.09
Moderate	62 (45)	35 (35)	
Heavy	23 (17)	28 (28)	
Diet Drinks			
Light	78 (57)	38 (38)	0.02
Moderate	30 (22)	32 (32)	
Heavy	29 (21)	30 (30)	
Fruit Juice			
Light	32 (23)	26 (26)	0.97
Moderate	40 (29)	28 (28)	
Heavy	37 (27)	27 (27)	
Very Heavy	28 (20)	19 (19)	

Table 13. Distribution of beverage intake according to percent body fat category
among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-values from Chi-square test or Fisher's exact test

		verweight $\geq 25 \text{ kg/m}^2$ )				
	Cases	Non-cases	– Crude OR	95% CI	Age- adjusted OR	95% CI
	N(%)	N (%)				
Sugar-sweetened beverages						
Light	21 (38)	68 (37)	1.00	Referent	1.00	Referent
Moderate	21 (38)	76 (42)	0.90	0.45, 1.78	0.91	0.45, 1.81
Heavy	13 (24)	38 (21)	1.11	0.50, 2.46	1.14	0.51, 2.56
Diet Drinks						
Light	20 (36)	96 (53)	1.00	Referent	1.00	Referent
Moderate	15 (27)	47 (26)	1.53	0.72, 3.26	1.59	0.74, 3.40
Heavy	20 (36)	39 (21)	2.46	1.20, 5.07	2.57	1.23, 5.37
Fruit Juice						
Light	11 (20)	47 (26)	1.00	Referent	1.00	Referent
Moderate	22 (40)	46 (25)	2.04	0.89, 4.69	2.05	0.89, 4.71
Heavy	13 (24)	51 (28)	1.09	0.45, 2.67	1.09	0.45, 2.68
Very Heavy	9 (16)	38 (21)	1.01	0.38, 2.69	1.02	0.38, 2.73

Table 14. Crude odds ratio and 95% CI of overweight as assessed by BMI across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

		erweight $2 \ge 80 \text{ cm}$				
	Cases	Non-cases	Crude OR	95% CI	Age- adjusted OR	95% CI
	N (%)	N (%)	011	<i><i>yo n o i</i></i>		
Sugar-sweetened beverages						
Light	30 (36)	59 (38)	1.00	Referent	1.00	Referent
Moderate	32 (29)	65 (42)	0.97	0.53, 1.78	0.98	0.53, 1.81
Heavy	21 (35)	30 (19)	1.38	0.68, 2.80	1.41	0.69, 2.91
Diet Drinks						
Light	29 (35)	87 (56)	1.00	Referent	1.00	Referent
Moderate	26 (31)	36 (23)	2.17	1.12, 4.18	2.25	1.16, 4.37
Heavy	28 (34)	31 (20)	2.71	1.40, 5.25	2.84	1.45, 5.56
Fruit Juice						
Light	18 (22)	40 (26)	1.00	Referent	1.00	Referent
Moderate	27 (33)	41 (27)	1.46	0.70, 3.06	1.47	0.70, 3.08
Heavy	20 (24)	44 (29)	1.01	0.47, 2.18	1.01	0.47, 2.19
Very Heavy	18 (22)	29 (19)	1.38	0.61, 3.10	1.39	0.62, 3.14

Table 15. Crude odds ratio and 95% CI of overweight as assessed by waist circumference across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

Table 16. Crude odds ratio and 95% CI of overweight as assessed by percent body fat across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

	Overweight $(\%BF \ge 33)$				Age-	
			Crude		adjusted	
	Cases	Non-cases	OR	95% CI	OR	95% CI
	N(%)	N(%)				
Sugar-sweetened beverages						
Light	37 (37)	52 (38)	1.00	Referent	1.00	Referent
Moderate	35 (35)	62 (45)	0.79	0.44, 1.43	0.79	0.44, 1.44
Heavy	28 (28)	23 (17)	1.71	0.86, 3.43	1.71	0.85, 3.47
Diet Drinks						
Light	38 (38)	78 (57)	1.00	Referent	1.00	Referent
Moderate	32 (32)	30 (22)	2.19	1.17, 4.12	2.21	1.17, 4.19
Heavy	30 (30)	29 (21)	2.12	1.12, 4.03	2.15	1.13, 4.12
Fruit Juice						
Light	26 (26)	32 (23)	1.00	Referent	1.00	Referent
Moderate	28 (28)	40 (29)	0.86	0.42, 1.75	0.86	0.42, 1.74
Heavy	27 (27)	37 (27)	0.90	0.44, 1.84	0.90	0.44, 1.84
Very Heavy	19 (19)	28 (20)	0.84	0.38, 1.82	0.83	0.38, 1.81

	Light	Moderate	Heavy		
	Consumer,	Consumer,	Consumer,		
	Never to $< 1$	$\geq 1$ per month to	> 2 per week to 1	Very Heavy	
	per month	2 per week,	per day,	Consumer,	1
	N (%)	N (%)	N (%)	$\geq 1$ per day	<i>p</i> -trend <sup>1</sup>
Sugar-sweetened beverages					
Model 1*	1.00	0.88 (0.43, 1.81)	1.17 (0.51, 2.68)	_	0.73
Model 2**	1.00	0.88 (0.43, 1.81)	1.16 (0.49, 2.74)	_	
Diet Drinks					
Model 1	1.00	1.45 (0.64, 3.28)	2.86 (1.33, 6.14)	_	0.02
Model 2	1.00	1.47 (0.65, 3.31)	2.88 (1.34, 6.21)	_	
Fruit Juice					
Model 1	1.00	2.37 (0.99, 5.72)	1.16 (0.46, 2.94)	0.90 (0.32, 2.55)	0.39
Model 2	1.00	2.47 (1.02, 6.00)	1.18 (0.46, 2.98)	0.85 (0.30, 2.44)	

Table 17. Multivariable-adjusted odds ratio and 95% CI of overweight as assessed by BMI across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-trend calculated by modeling BMI outcome by the median servings per day of each beverage type and testing for linearity

\* Model 1 includes age (continuous), daily coffee and tea intake (serving/d), and physical activity (continuous)

\*\* Model 2 includes variables in Model 1 and total energy intake

	Light	Moderate	Heavy		
	Consumer,	Consumer,	Consumer,		
	Never to $< 1$ per month N(%)	$\geq$ 1 per month to 2 per week, N(%)	> 2 per week to 1 per day, N (%)	Very Heavy Consumer, ≥ 1 per day	<i>p</i> -trend <sup>1</sup>
Sugar-sweetened beverages					
Model 1*	1.00	0.88 (0.47, 1.64)	1.40 (0.67, 2.91)	_	0.33
Model 2**	1.00	0.84 (0.45, 1.59)	1.23 (0.58, 2.64)	_	
Diet Drinks					
Model 1	1.00	2.21 (1.10, 4.41)	2.98 (1.49, 5.95)	_	0.01
Model 2	1.00	2.30 (1.14,4.62)	3.14 (1.56, 6.35)	_	
Fruit Juice					
Model 1	1.00	1.59 (0.74, 3.41)	1.03 (0.47, 2.25)	1.34 (0.58, 3.12)	0.75
Model 2	1.00	1.73 (0.79, 3.77)	1.06 (0.48, 2.33)	1.23 (0.30, 2.89)	

Table 18. Multivariable-adjusted odds ratio and 95% CI of overweight as assessed by waist circumference across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-trend calculated by modeling BMI outcome by the median servings per day of each beverage type and testing for linearity

\* Model 1 includes age (continuous), daily coffee and tea intake (serving/d), and physical activity (continuous)

\*\* Model 2 includes variables in Model 1 and total energy intake

	Light	Moderate	Heavy		
	Consumer,	Consumer,	Consumer,		
	Never to $< 1$ per month	$\geq$ 1 per month to 2 per week,	> 2 per week to 1 per day,	Very Heavy Consumer,	
	N (%)	N (%)	N (%)	$\geq 1$ per day	<i>p</i> -trend <sup>1</sup>
Sugar-sweetened beverages					
Model 1*	1.00	0.74 (0.40, 1.38)	1.75 (0.84, 3.67)	_	0.07
Model 2**	1.00	0.74 (0.40, 1.39)	1.78 (0.82, 3.84)	_	
Diet Drinks					
Model 1	1.00	2.83 (1.41, 5.69)	2.82 (1.40, 5.69)	_	0.07
Model 2	1.00	2.87 (1.43, 5.76)	2.86 (1.42, 5.77)	_	
Fruit Juice					
Model 1	1.00	0.87 (0.41, 1.83)	0.92 (0.44, 1.96)	0.73 (0.32, 1.67)	0.74
Model 2	1.00	0.88 (0.42, 1.88)	0.93 (0.44, 1.97)	0.71 (0.31, 1.63)	

Table 19. Multivariable-adjusted odds ratio and 95% CI of overweight as assessed by percent body fat across beverage intake categories among college-aged women in the UMass Vitamin D Status Study, 2006-2010

<sup>1</sup>*p*-trend calculated by modeling BMI outcome by the median servings per day of each beverage type and testing for linearity

\* Model 1 includes age (continuous), daily coffee and tea intake (serving/d), and physical activity (continuous)

\*\* Model 2 includes variables in Model 1 and total energy intake

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