**TITLE PAGE**

**Associations of sugar sweetened beverage intake at ages 18 months and 5 years with adiposity outcomes at age 6 years: The Singapore GUSTO mother-offspring cohort**

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This study was registered at clinicaltrials.gov as NCT01174875 ([www.clinicaltrials.gov,NCT01174875](http://www.clinicaltrials.gov,NCT01174875))

**ABSTRACT**

Consumption of sugar sweetened beverages (SSBs) in infants and young children are less explored in Asian populations. The Growing in Singapore Towards healthy Outcomes (GUSTO) cohort study examined associations between SSB intakes at ages 18 months and 5 years with adiposity measures at age 6 years. We studied Singaporean infants/children with SSB intake assessed by food frequency questionnaires (FFQ) at ages 18 months (n=555) and 5 years (n=767). The median (interquartile range) for SSB intakes is 28(5.5-98) ml at age 18 months and 111 (57-198) ml at age 5 years.Associations between SSB intakes (100 ml/day increments and tertile categories) and adiposity measures (BMI standard deviation scores (s.d. unit), sum of skinfolds (SSFs)) and overweight/obesity status were examined using multivariable linear and Poisson regression models, respectively. After adjusting for confounders and additionally for energy intake, SSB intakes at age 18 months were not significantly associated with later adiposity measures and overweight/obesity outcomes. In contrast, at age 5 years, SSB intakes when modelled as 100ml/day increments were associated with higher BMI by 0.09 (95% CI: 0.02, 0.16) s.d. unit, higher SSF thickness by 0.68 (0.06, 1.44) mm, and increased risk for overweight/obesity by 1.2 times (1.07, 1.23) at age 6 years. Trends were consistent with SSB intakes modelled as categorical tertiles. In summary, SSB intake in young childhood is associated with higher risks of adiposity and risk for overweight/obesity. Public health policies working to reduce SSB consumption need to focus on prevention programs targeted at young children.

246 words

**INTRODUCTION**

The prevalence of overweight and obese preschoolers (24 to 72 months old) in Chinese Singaporeans range from 7.0% to 8.1% ([1](#_ENREF_1)). These numbers are noteworthy as Southeast Asians face a higher risk of obesity-related disorders like diabetes, hypertension and cardiovascular disease which can manifest in children of a younger age compared to ethnic Europeans ([2](#_ENREF_2)). Furthermore, childhood overweight and obesity tends to be stable (track) through to adulthood ([3](#_ENREF_3)). One well-documented aspect that may contribute to childhood overweight and obesity risk elsewhere, and now in Southeast Asia, is the increased consumption of soft drinks, juice drinks, and other sweetened drinks ([4](#_ENREF_4), [5](#_ENREF_5)).

Early childhood is also the period where food preferences and eating behaviors develop that might serve as the foundation for future eating habits ([6](#_ENREF_6)). Sugar sweetened beverage (SSB) consumption patterns in children that start as early as infancy ([7](#_ENREF_7), [8](#_ENREF_8)), and preschool ([9](#_ENREF_9)) might have lasting implications through childhood, adolescence and into adult years. The negative implications of the overconsumption of SSB on BMI in children aged 6-19 years are well-established ([4](#_ENREF_4), [5](#_ENREF_5)). However, the associations of SSB consumption with BMI in younger children may not be the same. Cross-sectional studies on SSB intake in preschoolers or younger children have mostly reported positive associations with BMI ([4](#_ENREF_4)). However, longitudinal studies relating to SSB intakes and BMI present equivocal results: One study reported positive associations between SSB consumption in infants 10-12 months of age and their BMI at 6 years ([10](#_ENREF_10)), while another study in 13 month olds observed higher BMI in girls, but not in boys at ages 2, 3, 4 and 6 years of age ([8](#_ENREF_8)). In children aged 2-5 years, one study reported a positive association between SSB consumption intakes at age 2 and higher BMI z-score between 2 to 4 years of age ([11](#_ENREF_11)), while the other study reported no association with BMI 6-12 months later ([12](#_ENREF_12)).

In the current literature, most studies to date have been conducted in children from Western populations ([4](#_ENREF_4)). There is a pertinent need to examine this in Asia, especially in Southeast Asia as lifestyles and diets of children here are rapidly changing and becoming increasingly urbanized ([13](#_ENREF_13)). However, to date, there are only a few studies in Asia itself, and largely confined to older children (6-14 years old). These limited cross-sectional studies in Asia also present conflicting findings. For example, consumption of SSBs in two studies from China reported positive associations with obesity in children aged between 3-7 years([14](#_ENREF_14)) and age 6-13 years ([15](#_ENREF_15)), but negatively associated with obesity in 9 to 14 years old Korean boys ([16](#_ENREF_16)), while a study in 7 to 12 years old Taiwanese children found no associations with BMI ([17](#_ENREF_17)).

To our knowledge, there are currently no studies examining SSB consumption patterns in Asian preschoolers or younger children and relating them to adiposity outcomes. To address this gap, we used data from a mother-offspring cohort in Singapore which is a microcosm of Asia, where three ethnicities (Chinese, Malay, Indians) corresponding to the three major population centers in Asia are represented ([2](#_ENREF_2)). We aim to describe the change in absolute amounts of SSB consumed in children aged 18 months and 5 years as part the descriptive analysis of the study, and to examine the associations of SSB intakes at both time points with adiposity measures (BMI Z-scores and skinfold thickness) and overweight/obesity status in children at age 6 years. SSB consumption is hypothesized to be higher in older children, while higher SSB consumption is hypothesized to be associated with higher adiposity measures and risk for overweight/obesity outcomes.

**METHOD**

**Study design**

The Growing Up in Singapore Towards healthy Outcomes (GUSTO) study has been

previously described in detail ([18](#_ENREF_18)). Briefly, GUSTO is a mother-offspring cohort where pregnant women of Chinese, Malay and Indian ethnicities were recruited in their first trimester between June 2009 and September 2010 at two major public maternity units, namely National University Hospital (NUH) and KK Women's and Children's Hospital (KKH). Of 3751 women screened, 2034 met eligibility criteria ([18](#_ENREF_18)) and 1247 were recruited into the study. The main exclusion criteria were non-homogeneous ethnic background (up to the four grandparents of the offspring), intention not to deliver in the study centers or not to remain in Singapore for the following 5 years. This study was carried out in accordance with the recommendations from the National Healthcare Group Domain Specific Review Board and the SingHealth Centralized Institutional Review Board with written informed consent from all participants. All participants gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the National Healthcare Group Domain Specific Review Board and the SingHealth Centralized Institutional Review Board (clinicaltrials.gov; NCT01174875).

At 18 months, there were 555 children with completed Food Frequency Questionnaires (FFQs) and BMI data; 407 children had completed skinfold thickness data. At age 5 years, there were 767 children with completed FFQ and BMI data; 619 children had completed skinfold thickness data (Supplementary Figure 1). A subset of 451 participants with completed FFQ at both time points were used to compare the trends of SSB intake volumes and types consumed and for sensitivity analyses.

**Exposure: Dietary assessment at ages 18 months and 5 years**

To assess dietary intakes at 18 months, the child’s primary caregiver (mostly mothers) were given a self-administered 94-item Food Frequency Questionnaire (FFQ) to complete. At age 5 years, the FFQ was extended to include additional food items that account for greater diet variety at later ages. At this time point, a 125-item FFQ was administered to primary caregivers by trained interviewers to ascertain the child’s diet. The food items in both FFQs were categorized into food groups: (i) bread; (ii) bread spreads; (iii) breakfast cereals; (iv) rice porridge and noodles; (v) potatoes and pasta; (vi) vegetables and bean curd; (vii) fruits; (viii) meat and ﬁsh; (ix) eggs; (x) cakes, biscuits and snacks; (xi) milk and dairy products; and (xii) other beverages. The FFQ also included general questions on food preparation methods, eating habits and practices and an open-ended section to capture additional food items not listed in the FFQ. Both FFQs were validated either against 24-hour recalls (24HR) at 18 months of age, or with 3-day food diaries (3DFD) at 5 years of age, and were previously described in detail. In our validation studies, the correlation analyses of total energy intakes were satisfactory at both time points with correlation values of r=0.4 ([19](#_ENREF_19), [20](#_ENREF_20)). Information for both of the FFQs were collected using the same standardized methods where mothers had to indicate the frequency of consumption over the past month as ‘never’, ‘number of times per month’, ‘number of times per week’ or ‘number of times per day’. In addition, they were also asked to indicate a typical serving size for each food consumed, and state the exact amount of beverage consumed in volumes, or units of measure (i.e.cups) or other standard serving sizes (i.e.1 bottle or 1 packet) which were all converted into volumes of intake (mls). Photographs of standardized household measuring utensils and food pictures were presented to assist mothers in quantifying their child’s food and beverage intakes.

Both the FFQs included 7 different beverage items that represent SSBs: the frequency of consumption and the volumes consumed per serving were recorded for each item. In the analyses, the definition of SSBs included those commonly described in literature ([4](#_ENREF_4), [5](#_ENREF_5)), i.e. carbonated, non-carbonated drinks and only pre-packaged fruit juices containing added sugar as well as commonly consumed sugar sweetened beverages in the Singaporean population such as malted drinks, cultured milk drinks, soy based drinks and traditional drinks (i.e. barley water, chrysanthemum tea, herbal tea) ([21](#_ENREF_21)). From the FFQs administered at age 18 months and 5 years respectively, the total daily intake (ml/day) of SSBs was quantitated by multiplying the frequency of consumption per day by the recorded volume of intakes for each specific beverage.

**Outcome: Anthropometric measures at age 6 years**

At age 6 years, measurements of weight, height and four skinfold thicknesses (triceps, biceps, subscapular and supra-iliac) were obtained by trained staff using standardized protocols as detailed previously ([22](#_ENREF_22)). Weight was measured to the nearest gram using calibrated SECA 803 weighing scale, and standing height measured using a stadiometer (SECA stadiometer 213). All measurements were taken in duplicate, and repeated a third time if there was a difference of 0.2 kg or 1 cm for the weight and height measures, respectively. The four skinfolds were measured in triplicate using Holtain skinfold calipers (Holtain Ltd, Crymych, UK) on the right side of the body and recorded to the nearest 0.2 mm, and repeated a fourth and fifth time if there was a difference of more than 1 mm within the first 3 readings. Average values across the repeated measurement were considered for all anthropometry measurement. Four averages were summed to derive the sum of skinfolds (SSF, in mm). Sex- and age-specific BMI z-scores were calculated using the World Health Organization references ([23](#_ENREF_23)). The cut-off for overweight and obesity combined were defined as +1SD above the reference distribution as per WHO recommendations ([24](#_ENREF_24)).

**Maternal and child covariates**

Maternal socio-demographic data (age, self-reported ethnicity, education level and parity) were obtained using a self-administered questionnaire at recruitment. Maternal weight and height (4 years post-partum) were measured using SECA 803 scales and a SECA 213 stadiometer (SECA Corp., Hamburg, Germany), respectively. These measurements were used to calculate body mass index (BMI, in kg.m-2). Gestational age (GA) was determined by a dating ultrasound scan in the first trimester. At birth and 18 months, weight was measured to the nearest gramme (SECA model 334; SECA Corp), and recumbent length was measured to the nearest 0.5cm from the top of the head to the soles of the feet (SECA model 210). The weight of children at 5 years of age was measured to the nearest 10g using a calibrated digital scale (SECAmodel803; SECACorp.), while standing height was measured using a stadiometer (SECA model 213). For reliability, all measurements were taken in duplicates and averaged. BMI at birth, 18 months and 5 years was calculated as weight divided by the square of recumbent length/height (kg/m2). Skinfold thickness at 18 months and 5 years (triceps, biceps, subscapular and suprailiac only at 5 years) were obtained to calculate sum of skinfolds (SSFs, in mm) ([22](#_ENREF_22)). We derived birth weight–for-GA and birth length–for-GA *z* scores by using references from our cohort ([25](#_ENREF_25)). From the obstetric records, we extracted information on child sex. Mothers were asked about the age at which their child had been introduced to solid foods and details on infant milk feeding (as detailed previously) ([26](#_ENREF_26)) using interviewer-administered infancy questionnaires. The early introduction to solids was defined as the introduction to foods other than milk before the age of 4 months (16 weeks of age) ([27](#_ENREF_27)). Screen time and outdoor play time were quantitated using a parental self-reported interviewer-administered questionnaire when the child turned 3 years of age ([28](#_ENREF_28)).

**Statistical analysis**

The distribution of SSB intakes was positively skewed and log transformation could not be performed due to a number of individuals (13%) with zero intakes at 18 months. With that we chose to analyze the SSB intakes as 1) a continuous variable modelled as 100 ml increments per day 2) a categorical variable of low, medium and high intakes based on the tertiles of the study sample at both time points, using separate regression models.

Continuous variables were described as mean ± standard deviation (SD), and categorical variables as frequencies and percentages. Linear regression models were conducted to examine SSB intakes (continuous or categorical) with adiposity outcomes (BMI z-scores and SSF) at age 6 years. Multivariable Poisson regression with robust variance ([29-31](#_ENREF_29)) was used to examine SSB intakes (continuous or categorical) with the relative risk of being overweight/obesity age 6 years. These were conducted separately for SSB intake at ages 18 months and 5 years. For both the linear and Poisson regression models, the regression coefficient was interpreted as an increase in outcome associated with every 100ml or with each tertile increase in SSB intake (relative to being in the lowest tertile). Trend tests to test the dose-response relationship across the tertiles of SSB intake were also performed by modelling the SSB tertiles as a continuous variable. We performed three models: An unadjusted model (model 1), a model adjusted for key potential confounders (model 2) and then additionally adjusted for baseline energy intake respective to the time point of exposure at age 18 months or at 5 years (model 3). The selection of potential confounders (maternal ethnicity, maternal education level, birth weight for gestational age, parity, breastfeeding duration) was based on the literature ([4](#_ENREF_4)), or if they changed the effect estimates of our univariate model with tertiles of SSB intake with BMI by >5% ([32](#_ENREF_32)). Screen time at age 3 years and maternal BMI 48 months postpartum was added to the list of confounders when examining the associations between SSB intake at age 5 years with adiposity and overweight/obese outcomes.

A sensitivity analysis was conducted to test the robustness of the findings using the subset of 451 participants with completed FFQ at ages 18 months and 5 years. Multiple imputations were used to account for only missing covariate values. Frequencies of missing values were <5% for breastfeeding duration and maternal education, and slightly higher for child screen time and maternal postpartum BMI (12-18%). All values were assumed to be missing at random based on the Little Missing Completely at Random(MCAR) test ( P value >0.05). Missing values were imputed 20 times using multiple imputation analysis, and chain equations. Imputations were based on available information on all exposure and outcome variables included in the study. Analyses were performed in each of the imputed datasets and the final results were pooled. We reported association estimates with their 95% confidence interval. All analyses were performed using SPSS IBM version 20.

**RESULTS**

**Characteristics of mothers and children**

Among the 767 participants who completed the FFQ at age 5 years, 98% reported their child consuming some form of SSBs. Children in the high intake tertile [median (interquartile range) 241 (197–328 ml)] had mothers who were more likely to be of Malay ethnicity, had lower educational attainment, were multiparous and had higher maternal postpartum BMI, compared to those in the low [40 (21–57 ml)] and middle [111 (93–137 ml)] tertile. Additionally, these children were most likely to have more than 4 hours of screen time viewing a day at age 3 years and had the highest total energy intake, BMI z-scores and sum of skinfolds at age 5 years (Table 1). The characteristics of participants who were (n=767) and were not (n=320) included in the analyses were similar except for maternal age (included mothers were slightly older) (Supplementary Table S1). All maternal and infant characteristics significantly associated with SSB intake tertiles at age 5 years were largely similar to SSB intakes at age 18 months in n=555 participants, except maternal parity, child screen time, BMI z-scores and skinfolds where no significant differences were observed with SSB intake tertiles at 18 months. While there was no significant difference between the intake tertiles at age 5 years with maternal age, higher SSB intake tertiles at 18 months were associated with younger mothers. Furthermore, the volume of intake in each tertile at age 18 months was lower compared to at age 5 years (Supplementary Table S2).

**Trends in SSB intake from age 18 month to 5 years**

In the subset of 451 children with data at both the 18 month and age 5 year time points, SSB intakes were compared. Overall, a higher proportion of participants were consuming higher volumes of SSBs (50->400ml versus 0-50ml) at age 5 years (5.8-32.4%) compared to 18 months of age (3.3-13.5%) (Figure 1). Furthermore, a higher proportion of children at age 5 years were consuming malted, cultured and sweetened drinks, but fewer consumed soya milk and traditional herbal tea compared to children aged 18 months (*p*<0.05) (Figure 2).

**SSB intake at ages 18 months with BMI z-score, sum of skinfold (SSF) thickness and overweight/obesity at age 6 years**

SSB intakes at 18 months modeled as 100ml/day increments were associated only with higher risk for overweight/obesity [(relative risk (RR):1.09; 95% confidence interval (CI): 1.02, 1.64)], but not with BMI z-scores or SSF. However, after further adjustment for energy intake, this association was no longer significant (Table 2). There were also no significant associations between the medium/high SSB intake tertiles at age 18 months for both adiposity outcomes (BMI z-score, SSF thickness) and risk for overweight/obesity at age 6 years, when compared to the lowest intake tertile (P trend >0.05). These findings remained even after further adjustment for energy intake (Table 2), and in the sensitivity analysis of 451 children (Supplementary Table S3).

**SSB intake at age 5 years with BMI z-score, sum of skinfold (SSF) thickness and overweight/obesity at age 6 years**

Higher volumes of SSB (per 100ml increment) were associated with higher BMI by 0.09 (95% CI: 0.03, 0.15) s.d. unit, higher SSF thickness by 0.75 (0.06, 1.44) mm, and increased risk for overweight/obesity by 1.2 times (1.07, 1.23) at age 6 years. Similar trends were seen with the categories of SSB intake, with the high SSB intake tertile being significantly associated with higher BMI z-scores by 0.33(0.11, 0.55) s.d. unit, and higher risk for overweight/obesity (RR (1.56(1.05, 2.33)) at age 6 years when compared to the low intake tertile (*P for trend* <0.05). There were no significant associations between the medium and high SSB intake tertiles and SSF thickness (*p* for trend >0.05). All associations remained even after further adjustment for energy intake (Table 3).

In sensitivity analyses (n=451), similar associations were observed between the volumes of SSB (per 100ml increment) with all outcomes. However, the results from the associations between the SSB intake tertiles and BMI z-scores and SSF thickness differed in this subset: Both the medium and high intake tertiles were associated with higher BMI z-scores [(β: 0.34 s.d. unit; 95% CI: 0.06, 0.62 s.d unit) and (β: 0.46 s.d. unit; 95% CI: 0.18, 0.74 s.d. unit), respectively]. Furthermore, the high intake tertile was associated with higher skinfold measures by 3.25(0.36, 6.14) mm (*p* for trend< 0.05) which was previously not observed in the 767 participants. All associations remained after further adjustment for energy intake (Supplementary Table S4).

**DISCUSSION**

Our study is the first prospective Asian Singaporean cohort study to observe that the consumption of SSBs in preschoolers (age 5 years) and young (18 months) children is associated with later childhood adiposity measures (BMI, and skinfold thickness) and overweight/obesity outcomes at age 6 years. At age 5 years, every 100 ml/day increment of SSB intake was associated with 0.09 s.d. unit higher BMI and 0.8mm higher SSF thickness, and 1.2 fold higher risk for overweight/obesity at 6 age years. Similarly, children in the highest intake tertile (median intake=~250ml) of SSB consumption at this age had a 0.3 s.d. unit higher BMI, and almost a 1.6 fold higher risk for overweight/obesity at age 6, compared to those in the low intake tertile (median intake= ~50ml/day), and there were positive non-significant trends with SSF thickness. All the associations were still significant after the adjustment for energy intake. In contrast, there were overall null findings between the SSB intake at 18 months with child adiposity at age 6 years.

The few studies in 1-2 year old children have observed positive associations between SSB intake and adiposity outcomes. A small sample of 97 Latino mother and toddler pairs aged between 1-2 years observed that higher consumptions of SSBs above the median was associated with higher BMI after a 6 month follow-up ([33](#_ENREF_33)). Two studies in the US examined SSB intakes in 2 year olds, with one reporting that children drinking SSBs (compared with infrequent/no drinkers) had higher BMI at 4 years of age ([11](#_ENREF_11)), while another reported that high intakes (>3 times/week) versus no intakes of SSBs was associated with higher odds for developing obesity at 6 years of age ([10](#_ENREF_10)). Lastly, high intakes (15 servings/week=320ml/day) of SSBs in 13 month old Dutch toddlers were significantly associated with increased BMI, but only in girls up to 6 years of age ([8](#_ENREF_8)). The overall null associations we observed with SSB intakes at 18 months could be attributed to the lower volumes of SSB consumed at this time point by the children in our cohort [(the median volume in our high intake tertile was only138ml/day (IQR: 98-231); the low volume was only 3ml/day (IQR: 0-7 ml)]. Alternatively, another potential concern regarding the SSB intake at 18 months is the use of the maternal report of dietary assessments which was self-administered at this time point. This will inevitably include some degree of measurement error due to recall bias or improper reporting that may have attenuated true associations in this study. With the exception of the study in Dutch toddlers mentioned earlier ([8](#_ENREF_8)), comparisons of the intake volumes across the studies in children within a similar age group is difficult as typical serving sizes of SSBs (in ml/day) are usually not ascertained, or different dietary assessment methods have been used (for e.g.: 24 hour-recall/interview questionnaire versus an FFQ) ([10](#_ENREF_10), [11](#_ENREF_11), [33](#_ENREF_33)).

Our findings related to children’s SSB intake at 5 years with BMI and overweight/obesity outcomes were consistent across the different measures, and the associations remained significant even in a subset analysis. These observations concur with the few existing cross-sectional studies ([34-36](#_ENREF_34)) and one prospective study ([11](#_ENREF_11)) in children of a similar age range. These studies have all used body weight measures (BMI or BMI Z-score) as a main outcome and have defined the overweight/obesity statuses of children ([11](#_ENREF_11), [14](#_ENREF_14), [34](#_ENREF_34), [35](#_ENREF_35)), but none have examined SSB intakes with skinfold measures. Sum of skinfold thickness (usually biceps, triceps, supra-iliac, and subscapular) has been shown to be a useful supplementary measure widely used to assess body fatness in children ([37](#_ENREF_37)). In our study, skinfold thickness was only significantly associated with adiposity outcomes when SSB intakes at 5 years were modelled as 100/ml day increments, but not with the tertile intakes, which could be due to the loss of power from the categorization into tertile groups ([38](#_ENREF_38)).

The underlying biological mechanism by which SSB intake affects adiposity has not been fully elucidated ([39](#_ENREF_39)). The hypothesized mechanism for the potential causal relationship between SSB intake and obesity is thought to be mediated through the increase in energy intake resulting from the reduction in satiety, and incomplete caloric compensation at subsequent meals ([5](#_ENREF_5)). However, the additional adjustment for total energy intake in our regression models did not affect most of the estimated significant associations in our main dataset, as well as in our subset analysis. Similar observations were seen in previous studies ([8](#_ENREF_8), [39](#_ENREF_39), [40](#_ENREF_40)), suggesting that the associations may be explained by non-energy effects of SSBs. High intakes of SSBs in preschoolers could be just markers of poor diet quality where these children are consuming high amounts of sweet foods, along with high intake of sugary beverages ([41](#_ENREF_41)). High glycaemic loads from the possible high sugar diet alone could induce hyperinsulinemia, leading to increase in fat deposition that is independent from energy intake ([42](#_ENREF_42)). Another alternate mechanism linking poor diet and obesity could be through the mediation effects of the microbiome profile in the gut. Poorer diet quality (e.g. Western dietary patterns of high-fat food and refined sugars) has been associated with a specific diversity of microbiota termed the “obesogenic microbiota”, which has been shown to display enhanced capacity for energy harvest from the diet that might lead to weight gain ([43](#_ENREF_43)). Overall, poorer diet quality that might lead to higher glycemic loads and the presence of obesogenic microbiota in the gut are all possible contributing factors to the non-energy effects between SSBs and adiposity.

In our current findings, we found a trend of higher volumes of SSB consumption in older children of preschool age (5 years versus 18 months). In our study, 98% of participants were consuming SSBs by ages 5 years, and this percentage is higher than the 80.5% children in China consuming SSBs by ages 3-7 years ([14](#_ENREF_14)). This observation is in line with a survey conducted in 800 Singaporean children in 2009 examining their beverage consumption habits: The survey showed that consumption of SSBs increased with age (7-10 years versus 3-6 years) ([21](#_ENREF_21)). Higher consumptions of SSBs were also observed to be displacing the consumption of healthier and more nutrient-dense beverages like milk ([21](#_ENREF_21)). Furthermore, the differences in the types of SSBs consumed between the two age groups (18 months versus 5 years) in this study suggest that age relevant interventions are needed to target the main types consumed. This may have implications on the current daily recommendations for sugar intake in Singaporean toddlers and preschoolers ([44](#_ENREF_44)).

Strengths of this study are the prospective data collection (1- year follow up) and assessment of a wide range of socio-demographic and lifestyle factors that were adjusted for in multivariable analyses. Although we only had FFQ data at year 5 for 71% of the mother child dyads who participated at the start, no differences between the participants who were excluded and included in the study were observed except for maternal age. We analyzed a number of adiposity measures, including sum of skinfold thickness (SSF) measures. Several limitations of this study needs to be acknowledged. Using an FFQ to assess dietary intake is the most appropriate tool because it provides an advantage over a single 24- hour dietary recall or food diaries by capturing habitual intake (over 30 days) ([45](#_ENREF_45)). However, like all dietary assessments, measurement errors in dietary intake may still be present ([46](#_ENREF_46)). For example, mothers of children attending daycare on weekdays might be less aware of their child’s exact food intake, and are thus less likely to report their daily intake accurately. Since this FFQ captured maternal self-reported dietary intake of children, a systematic bias might occur if mothers underreport intake of SSBs to meet more socially desirable intakes ([47](#_ENREF_47)). However, we attempted to control for this by using cutoffs within the 500 to 4,000kcal range for energy intake, suitable for children of this age range to identify dietary under-reporters and over-reporters ([48](#_ENREF_48)). Because of the observational design of our study, casual effects of SSB on child adiposity could not be determined. Furthermore, the relatively short follow-up (1 year) between SSB intakes at age 5 and adiposity outcomes at age 6 might not be sufficient for us to observe longer lasting effects of SSB on adiposity, and because of this short follow-up, we cannot rule out the possibility of reverse causation (i.e. higher BMI or energy intake leading to higher consumptions of SSB). Although we collected data on child outdoor activity, the self-reported method used quantify physical activity in this study can underestimate the strength of the relationship between this covariate and the risk factor of interest (SSB intake). Future studies will benefit from objective physical activity data measured with an accelerometer rather than questionnaire-based activity measures. Lastly, although we attempted to control for major confounders in this study, residual confounding (i.e. other lifestyles factors such as diet quality) could not be ruled out.

**Conclusion**

In conclusion, our observations suggest that SSB consumption should be limited in young children to address the raising prevalence of child obesity. It also further contributes to the emerging consensus of the negative effects of SSB consumption in children below school age. The development of public health strategies to educate childcare centers, and parents of toddlers and preschoolers about the health implications of high consumption of SSB should be a priority. Finally, further longitudinal analyses are needed to better understand the mechanisms involved and the effects on SSB intake of long-term weight gain.

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**CONFLICT OF INTEREST**

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**AUTHOR CONTRIBUTIONS**

FY, KMG, YSC, LPS and KHT designed and led the GUSTO cohort study. PLQ, JK and YYC contributed to the statistical analysis and writing of the manuscript. JYT cleaned and processed the data for SSB intakes. HXL and RS cleaned, processed and performed the validation of the FFQs at 18 months and Year 5. JB and PN cleaned the dataset for screen time and outdoor play. IMA, WLY, MTT and YSL contributed to the collection and processing of the anthropometric datasets. PLQ and MFFC were responsible for finalizing the manuscript. All authors contributed to and approved the final manuscript.

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**Figure legends**

**Figure 1: Proportion of children consuming SSBs at 18 months and 5 years of age by volume range (n=451).** χ2-analysis was used to determine the differences in the proportion of participants consuming and not consuming SSBs for each specific volume range.

**Figure 2: Types of SSB consumed at age 5 compared to 18 months (subset of n = 451).** χ2-analysis showed statistically significant differences between the proportion of children at 18 months and 5 years for all SSB types (p<0.05).

Table 1: Maternal and child’s characteristics according to tertiles of SSB intake at 5 years of age (n = 767)

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Low intake 40 (21 – 57 ml)2 (n = 255) | Medium intake 111 (93 – 137 ml)2  (n = 256) | High intake 241 (197 – 328 ml)2  (n = 256) | *P*3 |
| Maternal ethnicity |  |  |  | <0.0001 |
| *Chinese*  *Malay*  *Indian* | 161 (63.1)  44 (17.3)  50 (19.6) | 154 (60.2)  61 (23.8)  41 (16.0) | 118 (46.1)  87 (34.0)  51 (19.9) |  |
| Maternal educational level |  |  |  | 0.016 |
| *Post-secondary and below*  *University and above* | 146 (57.5)  108 (42.5) | 172 (67.7)  82 (32.3) | 173 (63.4)  80 (31.6) |  |
| Parity |  |  |  | 0.001 |
| *Primiparous*  *Multiparous* | 126 (49.4)  129 (50.6) | 131 (51.2)  125 (48.8) | 91 (35.5)  165 (64.5) |  |
| Maternal age at first birth (years) | 31.3 ± 5.1 | 30.9 ± 5.1 | 30.7 ± 5.4 | 0.193 |
| Postpartum BMI at 4 years (kg/m2) | 23.8 ± 4.7 | 24.9 5.8 | 25.1 ± 4.9 | 0.010 |
| Child sex |  |  |  | 0.332 |
| *Boy*  *Girl* | 125 (49.0)  130 (51.0) | 131 (51.2)  125 (48.8) | 142 (55.5)  114 (44.4) |  |
| Breastfeeding status |  |  |  | 0.695 |
| *Never breastfed*  *Breastfed for <6 months*  *Breastfed for >* 6 months | 11 (4.4)  207 (82.5)  33 (13.1) | 10 (4.0)  216 (86.4)  24 (9.6) | 13 (5.3)  207 (84.1)  26 (10.6) |  |
| \*Early introduction to solids (<16 weeks of age) |  |  |  | 0.143 |
| *Yes*  *No* | 227 (97.4)  6 (2.6) | 229 (99.5)  1 (0.5) | 224 (97.4)  6 (2.6) |  |
| Child screen time (hours/day) |  |  |  | 0.012 |
| *< 2 hours per day*  *2 – 4 hours per day*  *> 4 hours per day* | 117 (51.5)  64 (28.2)  46 (20.3) | 92 (41.4)  77 (34.7)  53 (23.9) | 82 (35.5)  82 (35.5)  67 (29.0) |  |
| Child outdoor playing (hours/day) |  |  |  | 0.776 |
| *< 2 hours per day*  *2 – 4 hours per day*  *> 4 hours per day* | 191 (86.8)  28 (12.7)  1 (0.5) | 186 (89.4)  21 (10.1)  1 (0.5) | 184 (85.6)  29 (13.5)  2 (0.9) |  |
| Birth weight for gestational age ( z-scores) | 0.02 ± 1.0 | 0.02 ± 1.03 | 0.16 ± 0.99 | 0.115 |
| Child energy intake at 5 years (kcal) | 1245 ± 431 | 1405 ± 435 | 1735 ± 627 | <0.0001 |
| BMI at 18 months (z-scores) | -0.16 + 0.96 | -0.08 + 1.09 | 0.11 + 1.02 | 0.259 |
| Σ skinfold at 18 months | 16.4 + 7.5 | 15.4 + 8.7 | 16.4 + 8.3 | 0.276 |
| BMI at 5 years (z-scores) | -0.19 ± 1.0 | 0.08 ± 1.3 | 0.20 ± 1.3 | <0.0001 |
| Σ skinfold at 5 years | 27.5 ± 8.8 | 28.9 ± 10.3 | 29.9 ± 11.1 | 0.013 |

GUSTO, Growing Up in Singapore Towards healthy Outcomes. There were missing data for education (n = 6), breastfeeding (n = 20), maternal postpartum BMI at 48 months (n = 97), birth weight for gestational age (n = 12), early introduction to solid foods (n = 74), SSF (n = 148), screen time (n = 87) and outdoor playing (n = 124).

\*Early introduction to solids was defined as the introduction to foods other than milk before the age of 4 months (16 weeks of age).

1*p*-value across the SSB tertile categories was determined with the use of a χ2-analysis (categorical) or trend tests using SSB intake tertile categories as continuous variable (continuous). Value was presented as mean ± s.d. for continous data or n (%) for categorical data.  
2 Values reflect median (IQR).  
3 *p*<0.05 is statistically significant

Table 2: Associations between sugar sweetened beverage (SSB) consumption at 18 months with adiposity measures at year 6.

|  |  |  |  |
| --- | --- | --- | --- |
| **SSB intake at 18 months** | **BMI z-scores1a(n=555)** | **SSF1b (n=407)** | **Overweight / obesity2a (n=555)** |
| **Model 1(unadjusted)** | **β(95% CI)** | **β(95% CI)** | **Relative risk(95% CI)** |
| 100ml/day increments | 0.09 (0.03, 0.15)\* | 1.04 (0.30, 1.77)\* | 1.10 (1.03, 1.17)\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | -0.04 (-0.29, 0.22) | -0.62 (-3.38, 2.14) | 0.92 (0.54, 1.56) |
| High intake | 0.23 (-0.02, 0.48) | 0.96 (-1.80, 3.72) | 1.40 (0.87, 2.24) |
| *p-trend* | *p* = 0.066 | *p* = 0.493 | *p* = 0.071 |
|  |  |  |  |
| **Model 2( adjusted)** |  |  |  |
| 100ml/day increments | 0.06 (-0.002, 0.12) | 0.68 (-0.05, 1.42) | 1.09 (1.02, 1.64)\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | -0.05 (-0.29, 0.20) | -0.91 (-3.57, 1.76) | 0.93 (0.55, 1.58) |
| High intake | 0.08 (-0.17, 0.33) | -0.42 (-3.18, 2.35) | 1.12 (0.68, 1.84) |
| *p-trend* | *p* = 0.532 | *p* = 0.758 | *p* = 0.194 |
|  |  |  |  |
| **Model 3(energy intake adjusted)** |  |  |  |
| 100ml/day increments | 0.05 (-0.01, 0.11) | 0.71(-0.05,1.47) | 1.09 (1.02, 1.17)\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | -0.05 (-0.29, 0.19) | -0.92(-3.59,1.75) | 0.93 (0.55, 1.58) |
| High intake | 0.06 (-0.20, 0,31) | -0.46(-3.27,2.34) | 1.10 (0.67, 1.81) |
| *p-trend* | *p* = 0.676 | *p*=0.850 | *p*= 0.204 |

Abbreviations: CI, confidence interval; RR, relative risk; SSF, Sum of skinfold 1Estimated regression coefficients and relative risk2 (95% CI) of the associations between SSB intake (high and medium compared with low as reference) with BMI z-scores and overweight/obesity outcomes at 6 years of age. Trend tests were performed using categories of SSB intake as continuous variable in the linear regression and Poisson regression models. Models are adjusted for ethnicity, education, birth weight for gestational age, screen time, breastfeeding duration and parity. Model 3 is model 2 additionally adjusted for energy intake at age 18 months \*\**p*-value < 0.01. \**p*-value < 0.05. a Median(IQR),n: low intake 2(0-6 ml),n=185; medium intake 28(18-43 ml), n=185; high intake 138(98-231 ml),n=185. b Median(IQR),n: 3(0-7 ml),n=135; medium intake 28(18-43 ml), n=136; high intake 136(99-238 ml),n=136

Table 3: Associations between sugar sweetened beverage (SSB) consumption at year 5 with adiposity measures at year 6.

|  |  |  |  |
| --- | --- | --- | --- |
| **SSB intake at year 5** | **BMI z-scores1a(n=767)** | **SSF1b(n=619)** | **Overweight /obesity2a(n=767)** |
| **Model 1(unadjusted)** | **β(95% CI)** | **β(95% CI)** | **Relative risk(95% CI)** |
| 100ml/day increments | 0.12 (0.05, 0.18)\*\* | 0.83 (0.15, 1.52)\* | 1.16 (1.09, 1.23)\*\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | 0.23 (0.01, 0.46)\* | 1.25 (-1.19, 3.69) | 1.25 (0.78, 1.98) |
| High intake | 0.42 (0.20, 0.65)\*\* | 2.62 (0.17, 5.06)\* | 1.74 (1.13, 2.69)\* |
| *p-trend* | *p* < 0.001 | *p* = 0.036\* | *p* = 0.005 |
|  |  |  |  |
| **Model 2(adjusted)** |  |  |  |
| 100ml/day increments | 0.09 (0.03, 0.15)\* | 0.75 (0.06, 1.44)\* | 1.15 (1.07, 1.23)\*\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | 0.17 (-0.05, 0.38) | 0.53 (-1.89, 2.94) | 1.13 (0.74, 1.71) |
| High intake | 0.33 (0.11, 0.55)\* | 1.93 (-0.52, 4.37) | 1.56 (1.05, 2.33)\* |
| *p-trend* | *p* = 0.003 | *p* = 0.123 | *p* = 0.025 |
|  |  |  |  |
| **Model 3( energy intake adjusted)** |  |  |  |
| 100ml/day increments | 0.09 (0.02, 0.16)\* | 0.87(0.14,1.60)\* | 1.15 (1.06, 1.24)\* |
| Low intake | Reference | Reference | Reference |
| Medium intake | 0.17 (-0.05, 0.39) | 0.62(-1.81,3.04) | 1.12 (0.74, 1.69) |
| High intake | 0.34 (0.11, 0.58)\* | 2.20(-0.38,4.78) | 1.54 (1.03 – 2.30)\* |
| *p-trend* | *p* = 0.004 | *p* =0.096 | *p* = 0.033 |
|  |  |  |  |

Abbreviations: CI, confidence interval; RR, relative risk; SSF, Sum of skinfold. 1Estimated regression coefficients and relative risk2 (95% CI) of the associations between SSB intake tertiles (high and medium with low as reference) with BMI z-scores and overweight/obesity outcomes at 6 years of age.Trend tests were performed using categories of SSB intake as continuous variable in the linear regression and Poisson regression models. Models are adjusted for ethnicity, education, birth weight for gestational age, screen time, breastfeeding duration, maternal BMI 48 months postpartum and parity, SSB intake at 18 months. Model 3 is model 2 additionally adjusted for energy intake at age 5 years. \*\**p*-value < 0.01. \**p*-value < 0.05. a Median(IQR),n: low intake 40 (21-57 ml/day),n=255; medium intake 111(93-137 ml),n=256; high intake 241(197-328 ml),n=256. b Median(IQR),n: low intake 37(21-54 ml),n=202; medium intake 108(91-134 ml),n=209; high intake 241(195-317 ml),n=208.