

2014

## Early infant feeding and adiposity risk: from infancy to adulthood

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This article was originally published as:

Oddy, W. H., Mori, T. A., Huang, R., Marsh, J. A., Pennell, C. E., Chivers, P., Hands, B., Jacoby, P., Rzehak, P., Koletzko, B. V., & Beilin, L. J. (2014). Early infant feeding and adiposity risk: from infancy to adulthood. *Annals of Nutrition and Metabolism*, 64 (3-4), 262-270.  
<http://doi.org/10.1159/000365031>

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**This is the author's version of an article published as:**

Oddy, W. H., Mori, T. A., Huang, R. C., Marsh, J. A., Pennell, C. E., Chivers, P. T., . . .  
Beilin, L. J. (2014). Early Infant Feeding and Adiposity Risk: From Infancy to  
Adulthood. *Annals of Nutrition and Metabolism*, 64(3-4), 262-270. doi:  
10.1159/000365031

Early infant feeding and adiposity risk: from infancy to adulthood.

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Running head: short breastfeeding duration and adiposity risk...

## ABSTRACT

**Introduction:** Systematic reviews suggest a longer duration of breastfeeding associates with a reduction in the risk of later overweight and obesity. Most studies examining breastfeeding in relation to adiposity have not used longitudinal analysis. In our study we aimed to examine early infant feeding with adiposity risk in a longitudinal cohort from birth to young adulthood using new as well as published data.

**Methods:** Data from the Western Australian Pregnancy Cohort (Raine) Study in Perth, Western Australia, were used to examine associations between breastfeeding and measures of adiposity at 1, 2, 3, 6, 8, 10, 14, 17 and 20 years.

**Results:** Breastfeeding was measured in a number of ways. Longer breastfeeding (in months) was associated with reductions in weight z-scores between birth and one year ( $\beta = -0.027$ ;  $p < 0.001$ ) in the adjusted analysis. At 3 years, breastfeeding for  $< 4$  months increased the odds of infants experiencing early rapid growth (OR=2.05; 95% CI: 1.43, 2.94;  $p < 0.001$ ). From 1 to 8 years, children breastfed  $\leq 4$  months compared to  $\geq 12$  months had a significantly greater probability of exceeding the 95th percentile of weight. The age breastfeeding stopped and milk other than breast milk was introduced (introduction of formula milk) played a significant role in the trajectory of BMI from birth to 14 years; the 4-month cut-point consistently associated with a higher BMI trajectory. The introduction of milk other than breast milk  $< 6$  months compared to 6 or more months, was a risk for being overweight or obese at 20 years of age (OR 1.47; 95% CI 1.12, 1.93;  $p = 0.005$ ).

**Discussion:** Breastfeeding to 6 months of age and beyond should be encouraged and is recommended for protection against increased adiposity in childhood, adolescence and young adulthood. Adverse long-term effects of early growth acceleration are fundamental in later overweight and obesity. Formula-feeding stimulates a higher postnatal growth velocity, whereas breastfeeding promotes slower growth and a reduced likelihood of overweight and obesity. Biological mechanisms underlying the protective effect of breastfeeding against obesity are based on the unique composition and metabolic and physiological responses to human milk.

**Keywords:** adiposity, breastfeeding, formula-feeding, obesity, Raine Study

## **INTRODUCTION**

Metabolic syndrome represents a cluster of risk factors for cardiovascular disease and type 2 diabetes mellitus, that include central adiposity, impaired fasting glucose tolerance, hypertriglyceridemia, decreased high-density lipoprotein cholesterol, hypertension [1] and often, a pro-inflammatory state [2], oxidative stress and fatty liver disease [3].

Population-based surveys show that metabolic risk factors are increasingly prevalent among children and adolescents and progress into adulthood [4]. This phenomenon is increasing worldwide largely as a consequence of the ongoing obesity epidemic [5].

Gale et al [6] recently showed that formula feeding affected fat mass in childhood and that compared with the body composition of breastfed infants, formula fed infants had an altered body composition in infancy. Systematic reviews have shown strong associations between early infant feeding and obesity risk [7-9] however sociodemographic, psychological, behavioural, ethnic and cultural influences also impact on childhood obesity emergence [10]. We have reported that breastfeeding protects against childhood obesity in a population cohort [11-13].

In this paper we collate the evidence of this association between early infant feeding and adiposity risk in longitudinal analysis from infancy to 20 years of age with a presentation of already published as well as new (not yet reported) work from the West Australian Pregnancy Cohort Study.

## **METHODS**

The West Australian Pregnancy Cohort (Raine) Study commenced in 1989 with the recruitment of 2,900 pregnant women between 16 and 20 weeks gestation [14]. A total of 2,868 live infants (96%) available for follow-up at birth provided a wide range of maternal and offspring data during pregnancy and at 1, 2, 3, 6, 8, 10, 14, 17 and 20 years of age. The 20 year follow-up was complete by December 2012.

### **Exposures**

***Duration of breastfeeding:*** Diaries were maintained by the mother during the first three years of life. The age that other milk was introduced and the age at which breastfeeding was stopped were recorded, providing good prospectively collected measures of infant feeding as continuous variables in months. Breastfeeding duration was dichotomised using four months as the cut-point, which is consistent with the World Health Organization recommendation prior to 2004 for mothers to exclusively breastfeed their infants for four months, although this has since been increased to six months [15]. Full breastfeeding was defined as breastfeeding without the regular introduction of milk other than breast milk but it did not preclude the intake of solid foods.

***Other factors:*** Comprehensive medical and obstetric data were obtained at recruitment. Questionnaires ascertained demographic information and maternal factors including usual pre-pregnancy weight. Antenatal information was obtained from maternal records and maternal height was measured at the first visit. Infant birthweight (g) and gestational age (in days) were obtained from medical notes. Maternal pre-pregnancy body mass index (BMI) was calculated by weight (kg)/height<sup>2</sup> (m). Social, demographic and family data including family income, structure and parental education during the mother's pregnancy were prospectively collected by questionnaire.

### **Outcomes measured at 1, 2, 3, 6, 8, 10, 14, 17 and 20 year follow-ups**

At the ages of 1, 2, 3, 6, 8, 10, 14, 17 participants attended a clinic assessment at the Telethon Kids Institute (formerly known as the Telethon Institute for Child Health Research), initially with primary caregiver and independently from 17 years on. At 20 years participants attended the clinical assessment at an adult health facility. Weight was measured to the nearest 100g using Wedderburn digital chair scales with children wearing light clothing, and height to the nearest 0.1 cm with a Holtain stadiometer. Body mass index (BMI) was calculated as weight (kg)/height(m)<sup>2</sup>. Additional anthropometric measures at all follow-up visits included calibrated measurements of waist, hip and arm circumference and skin folds (suprailiac, triceps and abdominal) using Holtain skinfold calipers (Holtain, Crymych, UK). Blood pressure was taken at all follow-up visits according to standardized methodology and venous blood samples were taken after an overnight fast for serum insulin, glucose, triglycerides, total cholesterol, HDL-cholesterol, LDL-cholesterol and high sensitivity C reactive protein. HOMA-IR was calculated as insulin [uU.ml] x glucose [mmol/L]/22.5. These additional anthropometric and metabolic results have previously been reported elsewhere [16].

### **Statistical Analyses**

*One year:* At one year of age, the population for analysis was defined as unrelated, full-term, singleton births of European descent with no congenital abnormalities. Weight at birth and one year of age were standardised for sex and age (age at birth was represented by a single time point) using the WHO Growth Standards [17, 18]. These reflect ideal growth patterns in children who have been breastfed exclusively for four months and are still breastfed at 12 months. Weight z-scores were calculated using the formula  $((X/M)^L - 1)/LS$ , where X is the weight measurement and L, M and S are the age and sex specific values for the power in the Box-Cox transformation, the median and the coefficient of variation, respectively [19].

The difference in weight z-score from 0-1 year was calculated as birth weight z-score minus year one weight z-score. Fetal sex-specific summary statistics were calculated for maternal anthropometrics, pregnancy and early life factors using mean and standard deviation for symmetric distributions and median and inter-quartile range (Q1-Q3) for asymmetric distributions.

The cross-sectional analysis of the difference in weight z-score from 0-1 year was performed using multivariate linear regression. Covariates were selected for the multivariate model using both forwards and backwards stepwise procedures. Covariates included: maternal (pre-pregnancy) age, height, weight, and BMI; maternal smoking indicator (at any time during the pregnancy); family income; parity (coded as an ordered categorical variable); pregnancy weight gain up to 34 weeks gestation, maternal self-reported diabetes and hypertension during pregnancy; gestational age (GA) at birth; and duration of breastfeeding. All analyses were performed using the statistical graphics software R version 2.6.2 [18].

***Growth trajectories to three years:*** Longitudinal growth in BMI over the first 3 years of life and the influence of breastfeeding duration, maternal pre-pregnancy BMI, parental educational status and maternal smoking during pregnancy were analyzed by Latent Growth Mixture Modelling (LGMM)(PR, BK, WO). Using this random effects extension of a Latent Class Growth Curve Model we identified homogeneous subgroups of typical BMI-trajectory classes among the heterogeneous individual growth curves. The number of BMI-trajectory-classes was identified by test statistics. Use of this model allowed effects of breastfeeding duration and other predictors of our identified typical BMI-trajectory classes and their effects, to be estimated. Detailed information on the LGMM is available [20]. The significance of breastfeeding duration was evaluated by 95% confidence intervals around the OR and the size



of the OR or mean outcomes and related P-values. Gender was not included in any LGMM as WHO standardized BMI-SDS scores are standardized to age and sex-specific “ideal growth”. All statistical analyses were performed with either the SAS 9.3 or the Mplus 7.1 statistics software.

***Year 1 to 8 [11]:*** Using the National Center for Health Statistics/Centers for Disease Control website (<http://www.cdc.gov/nchs/>), weight-for-length was calculated for 1-year-olds and BMI Z scores for 3-, 6-, and 8-year-olds and overweight was defined according to the sex-specific 95th percentiles. Categories of breastfeeding were defined as: never breastfed, breastfed  $\leq 4$  months, breastfed 5 to 8 months, breastfed 9 to 12 months, and breastfed  $>12$  months. These categories were used in mixed-effects models with BMI Z-score as the response variable and age as the explanatory variable. Final models included adjustment for birth-weight, gestational age, ethnicity, sex, maternal BMI, smoking status during pregnancy, parity and educational level. Generalized Estimating Equations modeled overweight from the ages of 1 to 8 years using the sets of covariates. A significance level of 0.05 was used.

***At 14 years [12]:*** We have reported adiposity rebound, defined as the last minimum (nadir) BMI before the continuous increase with age [21], and calculated in a subset of individuals (n=171) for whom a complete set of BMI data were available for all eight measured time points (birth to 14 years) and was based on the child’s age in months. We used linear mixed modeling which accounted for correlated errors normally associated with repeated, continuous and correlated observations. The variables for age at which breastfeeding was stopped and age at which other milk was introduced were categorized using a 4-month cut-off point. These were compared across weight status groups[22]. The effect of duration of breastfeeding (based on the four month cut-point) was assessed across the age range based on group mean differences

tested using Pearson Chi-Square. Adjustment for potential socioeconomic status confounding was based on maternal education.

***At 17 years:*** A cross-sectional analysis was performed (n=1053) using multivariate linear regression models with continuous BMI the response variable and breastfeeding duration and age milk other than breast milk introduced in months, as dichotomized explanatory variables, adjusting for child gender and maternal factors: pre-pregnancy BMI, education, age and family income.

***At 20 years:*** The non-random loss of disadvantaged participants is to be expected in a longitudinal cohort and may decrease generalizability of any findings. We developed a method that could be applied to account for such attrition bias using inverse probability weighting [23].

Our aim was to investigate exclusive breastfeeding and prevalence of BMI, overweight and obesity at 20 years. Of the original cohort (n=2868) 73% were available at the 20 year follow-up. Our follow-up response rate was 46% of those available for follow-up (n=1053).

Participants at 20 years were compared to non-participants to identify a priori candidates to predict missingness [24]. A missingness model was defined based on socio-demographic factors that predicted continued participation (maternal factors of age, education, BMI before pregnancy, and family income at birth). Final models (i.e. overweight and obesity vs breastfeeding) incorporated generalised estimating equations and our final predictive model provided an odds ratio and 95% confidence interval.

## RESULTS

**At one year:** By the time their infant was six months of age 39% of mothers had stopped breastfeeding and 55% had introduced milk other than breast milk. At one year of age a longer duration of breastfeeding was independently associated with reductions in weight z-scores between birth and the first year of life ( $\beta$  -0.027; SE = 0.004;  $p < 0.001$ ) following adjustment for maternal diabetes, increasing weight gain up to 34-weeks gestation, birth order and duration of gestation (Table 1).

**At three years:** Using BMI-SDS, the growth pattern of infants identified in a class 2 pattern - comprising 35.8% of the cohort begins at 0.3 standard deviation above the ideal growth standard and gains almost 1 standard deviations within the first 2 years (Figure 1). The curve remains 1 standard deviation above the ideal, normative growth development up to 3 years of age. We showed that breastfeeding for less than 4 months significantly increased the odds of infants being assigned to the early rapid growth class 2 (OR=2.05; 95% CI:1.43, 2.94;  $p < 0.001$ ) following adjustment for maternal BMI before pregnancy, socioeconomic status and gestational age. Although the growth patterns of infants identified in a class 1 or class 3 pattern showed an even stronger increase in BMI-SDS deviation from ideal growth within the first 2 years than those in class 2, breastfeeding was not significantly associated with these growth patterns.

**At 1, 3, 6 and 8 years [11]:** As reported previously (11) at 1 year, infants breastfed for more than 12 months were the leanest group (mean Z score -0.16, 95% CL -0.28, -0.04; not breastfed 0.16, 95% CL 0.02, 0.29; breastfed  $\leq 4$  months 0.31, 95% CL 0.22, 0.40; 5-8 months 0.17, 95% CL 0.06, 0.27; 9-12 months 0.11, 95% CL 0.01, 0.22). From 1 to 8 years, children

breastfed  $\leq 4$  months compared to breastfed for  $> 12$  months had the greatest probability of exceeding the 95th percentile for BMI (OR 1.87, 95% CL 1.21,2.89;  $p=0.005$ ) and was associated with the highest prevalence of maternal obesity, smoking and lower education.

**At 14 years:** The age ‘breastfeeding stopped’ and ‘milk other than breast milk was introduced’ played significant roles in the trajectory of BMI from birth to 14 years, especially at the 4-month cut-point where differences for BMI peak were apparent at one year of age, with this difference consistent over time into adolescence (Figures 2A & B).

**At 17 years:** Breastfeeding stopped before six months and the introduction of milk other than breast milk before six months compared to six months or later were associated with increased BMI at 17 years. BMI was consistently associated with early cessation of breastfeeding following adjustment for gender, maternal age, maternal education, family income at birth and maternal pre-pregnancy BMI (Table 2).

**At 20 years [13]:** In recent work at 20 years, our final predictive weighted model showed that the cessation of exclusive breastfeeding before six months, compared to six months or later was associated with an increased prevalence of overweight and obesity (BMI 25+) at 20 years (OR 1.47; 95% CI: 1.12, 1.93;  $p=0.005$ ). The full multivariate generalised estimating equation model for being overweight or obese compared to normal weight at 20 years, by milk introduced before 6 months of age is shown in Table 3.

## DISCUSSION

Breastfeeding, preferably of long duration (longer than six months), and late introduction of formula milk is recommended for protection against increased adiposity in childhood, adolescence and young adulthood. Our results provide an opportunity to examine the influence of infant feeding on weight status, the relationship with adiposity rebound and subsequent adolescent and young adult BMI. Overall, our findings suggest that formula feeding compared to breastfeeding results in accelerated weight gain in the infant, with likely upward BMI centile crossing [25]. Our results support other studies that showed overweight in adolescence increased as time spent being exclusively breastfed decreased [26] and extend the findings of Burke et al [11] which showed a higher BMI at eight years in children breastfed for four months or less. Statistical modelling [12] showed that breastfeeding and the age other milk introduced play an important role and may contribute to the timing of the adiposity rebound, .

A number of observational studies and meta-analyses have shown small protective effects of breastfeeding on obesity. However other studies have found no effect and raise the possibility of unknown confounders and publication bias [27]. Three comprehensive meta-analyses were conducted over the past decade. A meta-analysis that included 9 studies with over 69,000 participants [7] showed that breastfeeding has a consistent protective effect against childhood obesity (OR:0.78; 95% CI: 0.71, 0.85), with 4 studies showing a dose-response effect for the duration of breastfeeding. A meta-analysis of 17 studies in 2005 [8] found a dose-response relationship, where an increased duration of breastfeeding related to a decreased risk of overweight later in life. More specifically this meta-analysis reported a 4% decrease in risk with each additional month of breastfeeding. When restricted to exclusive breastfeeding, the risk of overweight decreased by 6% per month [8] suggesting that exclusivity of breastfeeding

may be central to the mechanism whereby it protects against obesity [28]. A meta-analysis by Owen et al [9] concluded that breastfeeding reduces the risk of obesity compared to formula feeding (OR: 0.87; 95% CI: 0.85–0.89). However, the same authors have since suggested that any observed protective effect of breastfeeding on BMI may be due to unadjusted confounding [27]. Beyerlein and von Kries [29] suggest that discrepancy in the findings of studies examining breastfeeding and obesity may be due to the different effects of breastfeeding on normal-weight versus overweight populations, showing a protective effect of breastfeeding in those within the highest BMI percentiles (>90th) [30].

A German study of 9,357 children, found that breastfeeding had a protective effect against obesity and overweight which remained significant after adjusting for social class and lifestyle [31]. A dose-response relationship was shown with the duration of breastfeeding, indicating a possible causal effect associated with a shorter duration.

A recent study, consisting of 822 young adults (18-28 years) from the Netherlands [32], demonstrated that exclusive breastfeeding had a significant protective dose-response effect on measures of body fat mass and visceral fat mass (BMI, waist circumference and waist-hip ratio). Furthermore, findings from a much smaller prospective randomized controlled trial of pre-term infants support a causal link between breast milk feeding and lipoprotein profile later in life [33].

A crucial period during post-natal growth relating to obesity risk has been hypothesized. The timing of this period remains uncertain with some suggesting the first few days of life [34], the first few weeks [28] or up to two years, may be the crucial period [35]. Plausible mechanisms for the development of fatness during this early sensitive window of exposure

include permanent structural changes such as decreased beta cell mass, accelerated cellular aging, telomere shortening with cell division and oxidative damage, and epigenetic programming changes from transcription to translation of protein [36]. Our results demonstrate that the early months of life are the crucial period in the development of adiposity later in life.

We showed that in terms of feeding patterns the time period of greatest risk for obesity later in life was prior to six months and this finding supports the recommendation of continued exclusive breastfeeding to six months and beyond. The theory is that a high nutrient diet in infancy adversely programs the cardio-metabolic system by promoting growth acceleration (upward centile crossing) [37]. Therefore slower growth benefits later CVD and its risk factors [33]. Early growth acceleration programs the abnormal vascular biology associated with early atherosclerosis. Therefore infants who grow rapidly during infancy or are at the highest end of the weight or BMI distribution are at increased risk of subsequent obesity [38]. Growth velocity may be a relevant influence in the causal pathway of obesity as suggested for fetal programming of metabolic disease [39] with associations between protein intake, growth velocity and weight gain reported [40, 41].

The theory of reverse causation in relation to growth suggests that infants who have lower growth trajectories, and therefore lower energy requirements, are satisfied with breastfeeding for longer. Children 'programmed' to be larger require and demand higher energy intake, resulting in the mother supplementing with formula or solid food earlier [42]. Rapid growth in the first few months of life leads to elevated risk of obesity in breastfed as well as formula fed infants, however as breastfeeding has been shown to promote slower growth, obesity is less likely in breastfed infants [43, 44].

Findings from a recent study followed up at two years and six years respectively [45, 46] compared the BMI of infants fed high-protein formula versus low-protein formula and a breastfeeding comparison group and is supportive of the early protein hypothesis. These authors reported a higher BMI in the high-protein formula group compared with the low-protein group who had BMI values closer to that of the breastfed infants. In regards to risk of later obesity, the most important differences between breast milk and formula appear to be related to the lower protein content and presence of hormones, growth factors and bioactive factors in breast milk [47]. In addition, biological mechanisms underlying the protective effect of breast milk feeding compared to formula feeding are based on the unique composition of human milk and the metabolic and physiological responses to human milk [48]. Breastfed infants may absorb less energy per volume than formula fed infants as well as receiving modifying growth factors that may inhibit adipocyte differentiation [48, 49].

Evidence exists for programming by nutrition in early infancy [50]. Feeding methods may prime offspring dietary behaviours, possibly by influencing development of the hypothalamus which occurs through gestation and into the postnatal period and is thought to play an important role in appetite control [51]. In humans, breastfed babies have been shown to exhibit better appetite control than bottle-fed babies [52]. This evidence supports the link between a longer duration of breastfeeding and a decreased risk of obesity later in life.

### **Strengths**

Our study has a number of strengths that include the use of a prospective pregnancy cohort and early infant feeding data, and anthropometric data collected to 20 years. Breastfeeding data were collected close to the period of breastfeeding cessation and a diary card was maintained to improve data collection. The study design and large community sample are



clear strengths, generating adequate statistical power to measure the association between dichotomized breastfeeding variables and anthropometric outcomes while controlling for possible confounders.

## **Conclusion**

As with other programming effects, the effect of early diet on later obesity risk may amplify over time. The early postnatal period is a particularly important time for the risk of the development of obesity and its cardio-metabolic sequelae which may potentially be addressed through the promotion of breastfeeding. Therefore this period is particularly important for targeting interventions and our findings suggest that the promotion of breastfeeding may have long-term protective benefits against future obesity.

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### Key points:

- Stopping breastfeeding before six months may lead to an increased risk of overweight and obesity later in life.
  - Introducing milk other than breast milk before six months may lead to an increased risk of obesity 20 years later.
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### To summarise our current knowledge:

- Overweight infants are more likely to become overweight children, adolescents and adults.
  - Breastfeeding preferably of long duration, preferably for at least 6 months and beyond and late introduction of formula milk is recommended for protection against increased adiposity in childhood, adolescence and adulthood.
  - Increasing the prevalence of exclusive breastfeeding to 6 months would be a worthwhile public health measure.
  - A possible adverse effect of formula feeding on postnatal weight gain and infant health remains of contemporary public health significance.
-

## **Acknowledgements**

We gratefully acknowledge the Raine Study participants and their families, and the Raine Study Team, for cohort coordination and data collection. Core funding for the Western Australian Pregnancy Cohort (Raine) Study is provided by the University of Western Australia; the Faculty of Medicine, Dentistry and Health Sciences at the University of Western Australia; the Telethon Kids Institute (formerly known as the Telethon Institute for Child Health Research); the Women and Infants Research Foundation; Curtin University of Technology; Edith Cowan University and the Raine Medical Research Foundation. We thank the National Health and Medical Research Council (NHMRC) for their provision of funding (Project ID#1022134 and European Union collaborative project ID#1037966).

Acknowledgments are extended to NHMRC funded participation in the European Union project 'Long-term effects of early nutrition on later health' FP7-289346-EarlyNutrition, Coordinator: Ludwig-Maximilians-University Munich, Germany Project Director: Professor Berthold Koletzko, MD PhD. The research leading to these results has received funding from the European Union's Seventh Framework Programme (FP7/2007-2013), project EarlyNutrition under grant agreement n°289346.

Prof WH Oddy wrote the paper. All co-authors have contributed to the content, read and approved the final manuscript and no conflicts of interest are declared.

**Table 1: Analysis of change in weight z-score from birth to one year**

	<b>Estimate</b>	<b>Standard Error</b>	<b>P</b>
Length of gestation* (days)	-0.037	0.003	<0.001
Parity: 0	1		<0.001
2	-0.287	0.072	
Maternal smoker : No	1		
Yes	0.319	0.056	<0.001
Maternal diabetes : No	1		
Yes	-0.435	0.142	0.002
Maternal hypertension : No	1		
Yes	0.123	0.057	0.031
Pregnancy weight gain <sup>a</sup> (kgs)	-0.015	0.004	<0.001
Pre-pregnancy maternal BMI* (kg/m <sup>2</sup> )	-0.019	0.006	0.001
Duration breast feeding (months)	-0.027	0.004	<0.001
Family Income <\$11,999	1		
\$12,000-\$23,999	0.070	0.086	0.412
\$24,000-\$35,999	0.084	0.086	0.331
>\$36,000	0.207	0.087	0.018

<sup>a</sup>Continuous covariates were centered prior to analysis: length of gestation minus 280 days, pregnancy weight gain minus 14 kg, pre-pregnancy maternal BMI minus 22 kg/m<sup>2</sup>.

**Table 2: Multivariate linear regression models for continuous BMI at 17 years with breastfeeding duration and age milk other than breast milk introduced in months, as dichotomized variables.**

	<b>BMI at 17 years (kg/m<sup>2</sup>)</b>
	Unstandardised beta coefficient (95% CI) p value
<b>Breastfeeding stopped by</b>	
3 months	1.26 (0.43, 2.10) 0.003
4 months	1.14 (0.37, 1.92) 0.004
5 months	1.01 (0.26, 1.75) 0.008
6 months	0.95 (0.23, 1.67) 0.100
<b>Milk other than breast milk introduced before</b>	
3 months	1.17 (0.43, 1.91) 0.002
4 months	1.15 (0.45, 1.84) 0.001
5 months	1.11 (0.42, 1.80) 0.002
6 months	1.07 (0.37, 1.76) 0.003

\* Adjusted for gender, maternal age <20, maternal education < yr12, family income <\$12000 at birth and maternal pre-pregnancy BMI ≥ 25kg/m

**Table 3: Multivariate generalised estimating equation model for being overweight or obese compared to normal weight at 20 years, by milk introduced before 6 months of age<sup>1</sup>**

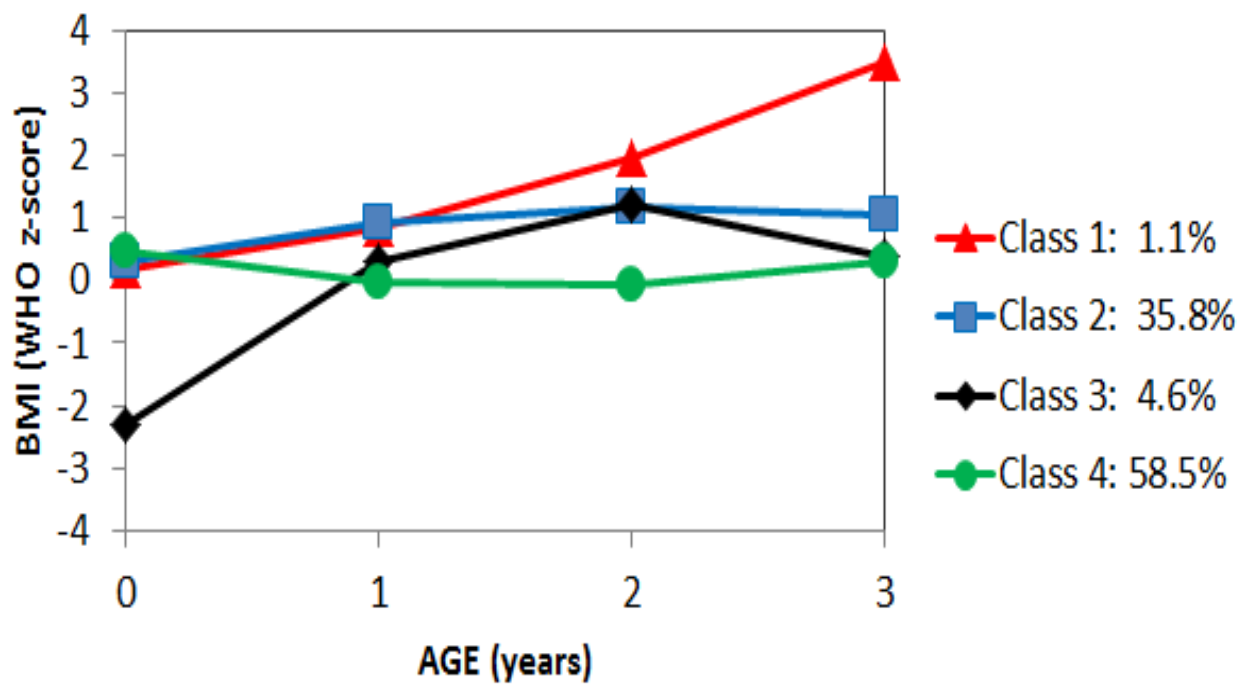
Exposures	Overweight or obese at 20 years OR (95% CI) p-value
Milk other than breast milk introduced < 6 months of age yes vs no	1.47 (1.12, 1.93) 0.005
Gender Male vs female	1.14 (0.87, 1.49) 0.34
Maternal age at birth <20 years vs ≥ 20 years	1.49 (0.80, 2.80) 0.21
Maternal education <yr12 vs ≥ yr12	1.27 (0.97, 1.67) 0.08
Family income at birth <\$12000 vs ≥ \$12000	1.59 (1.05, 2.40) 0.03
Maternal BMI before pregnancy ≥ 25 kg/m <sup>2</sup> vs < 25 kg/m <sup>2</sup>	3.11 (2.22, 4.34) <0.001

<sup>1</sup> Sample is weighted according to principles of inverse probability weighting [23].

**Supplemental Table Optional: Characteristics of study participants with breastfeeding information at birth and body mass index data at 20 years of age**

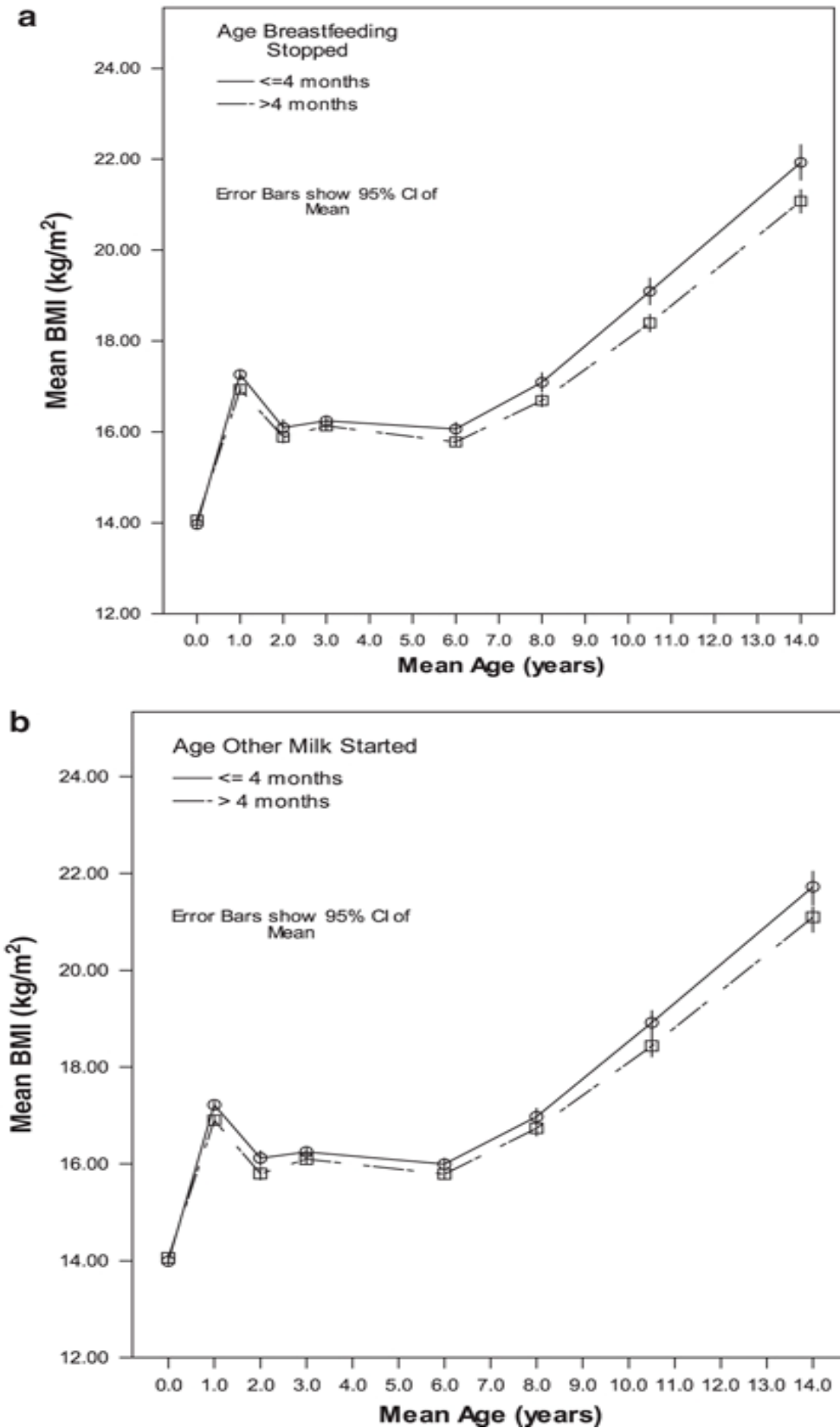
<b>Child factors</b>	<b>No</b>	<b>Column %</b>
Birthweight		
< 2500g	59	6.6
≥ 2500g	831	93.4
Gestational age		
< 37 weeks	75	8.4
≥ 37 weeks	815	91.6
Gender		
male	457	51.3
female	433	48.7
<b>Maternal and family factors</b>		
Maternal age at birth		
< 20 years	37	4.2
≥ 20 years	853	95.8
Maternal education		
< year 12	484	54.4
≥ year 12	406	45.6
Family income at birth		
< \$12000	106	11.9
≥ \$12000	784	88.1
Maternal pre-pregnancy BMI		
≥ 25kg/m <sup>2</sup>	152	17.1
< 25kg/m <sup>2</sup>	738	82.9
Breastfeeding stopped by		Cumulative %
4 months	266	29.9
6 months	350	39.3
Other milk introduced before		
4 months	369	41.5
6 months	489	54.9

Complete data available for breastfeeding duration, time other milk introduced, body mass index at 20 years and covariates of maternal age and education at birth, family income at birth and maternal pre-pregnancy BMI.



**Figure 1: Identified BMI trajectory classes from birth to 3 years by latent growth mixture modelling in the Raine Study.**

**Figure 2. Mean BMI by mean age based on Age Breastfeeding Stopped (a) (n=1330) and Age Other Milk Started (b) groups  $\leq 4$  months and  $> 4$  months (n=1320) [12]**





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