Accretion of fat-free rather than fat mass in infancy is positively associated with linear growth in childhood

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List of abbreviations used: ADP, air-displacement plethysmography; ANOVA, One-way analysis of variance; BC, body composition; EBLUPs, estimated best linear unbiased predictors; FM, fat mass; FFM, fat-free mass; HAZ, height-for-age z-score; iABC, infant anthropometry and body composition cohort; JUSH, Jimma University Specialized Hospital; PCA, principal component analysis; REML, restricted maximum likelihood.

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Abstract

Background: We have previously shown that fat-free mass (FFM) at birth is associated with height at 2 y of age in Ethiopian children. However, the relationship between changes in body composition during early infancy and later linear growth has not been studied.

Objective: This study examined the associations of early infancy fat mass (FM) and FFM accretion with linear growth from 1 - 5 y of age in Ethiopian children.

Methods: In the infant anthropometry and body composition study (iABC), a prospective cohort study was carried out among children in Jimma, Ethiopia, followed from birth to 5 y of age. FM and FFM were measured up to 6 times from birth to 6 mo using air-displacement plethysmography. Linear mixed effects models were used to identify associations between standardized FM and FFM accretion rates during early infancy and linear growth from 1 - 5 y of age. Standardized accretion rates were obtained by dividing FM and FFM accretion by their respective SD.

Results: FFM accretion from 0 - 6 mo was positively associated with length at 1 y (β=0.64; 95% CI: 0.19, 1.09; *P*=0.005) and linear growth from 1 - 5 y (β=0.63; 95% CI: 0.19, 1.07; *P*=0.005). The strongest association with FFM accretion was observed at 1 y. The association with linear growth from 1 - 5 y was mainly engendered by the 1 y association. FM accretion from 0 - 4 mo was positively associated with linear growth from 1 - 5 y (β=0.45; 95% CI: 0.02, 0.88; *P*=0.038) in the fully adjusted model.

Conclusions: In Ethiopian children, FFM accretion associated with linear growth at 1 y and no clear additional longitudinal effect from 1 - 5 y was observed. FM accretion had weak

association from 1 - 5 y. This study was registered as ISRCTN46718296 at www.controlled-trials.com.

Keywords: fat-free mass, fat mass, body composition, linear growth, height, length, children

Introduction

Childhood stunting is associated with reduced human capital later in life (1). Undernutrition and multiple infections are considered main causes of stunting in low- and middle-income countries (2). Although timing of childhood growth faltering has been well documented (3,4), the evidence on predictors of childhood linear growth is largely based on cross-sectional studies (5,6).

Studies of body composition (BC) in infancy have long been hampered by the lack of feasible methods, but recent technological advancement has resulted in an increased number of studies that describe BC throughout childhood in both high (7–11), and low- and middle-income countries (12,13)**.** As a consequence of these improved methods, there is an increasing interest in clinical research to identify the effects of fetal and postnatal programming on childhood health outcomes.

Several studies have shown that low birth weight is a risk factor for shorter height (14,15), while rapid weight gain during infancy has been linked to a number of health outcomes including non-communicable diseases in later life (1,16). However, birth weight and weight gain do not differentiate between fat and fat-free tissue during infancy (17). Although few studies have addressed the association between growth and nutrition in early infancy and linear growth longitudinally (14,15,18), the role of early infancy fat mass (FM) and fat-free mass (FFM) accretion in childhood linear growth remains unknown.

A review by Toro-Ramos et al (10) revealed that FFM tissue increase steadily from birth to 1 y, whereas FM had variable accretion. These different FM and FFM accretion patterns during infancy might affect later health outcome in a different way.

Recently, in the Ethiopian infant anthropometry and body composition cohort (iABC), it was shown that FFM at birth, but not FM, was positively associated with height (19) and development (20) at 2 y of age. Further, based on the same cohort, it has also been shown that FFM was accreted linearly during the first 6 mo while FM accretion leveled out at 4 mo of age (12). However, the immediate and long-term health effects of postnatal BC accretion have not been studied. Thus, follow-up studies are required to identify the role of FM and FFM accretion rates during infancy on linear growth throughout childhood. In the present study we examined the longitudinal association of early infancy FM and FFM accretion rates with linear growth during childhood. We hypothesized that accretion of FM and FFM in early infancy correlated differently with linear growth from 1 - 5 y.

Subjects and methods

Study design, setting and population

A prospective cohort study was carried out among 644 apparently healthy children followed from birth to 5 y of age at Jimma University Specialized Hospital (JUSH), Ethiopia. This study is a follow-up of the iABC study, which involved a longitudinal birth BC cohort that has been described in detail elsewhere (12,21). In short, mothers and their newborns were recruited among mothers giving birth at the maternity ward in JUSH. Inclusion criteria were newborn having birth weight ≥1500 g, term delivery (gestational age ≥37 wk) according to the New Ballard score (22), without congenital malformation and with parents residing in Jimma Town.

Ethical considerations

This study was approved by Jimma University, College of Health Sciences Ethical Review Board with reference number (RPO/56/2001). Written informed consent was obtained from mothers/caregivers before enrollment. Participants received an information sheet explaining the objective of the study and the procedure written in the local language. Children found ill at each follow-up visit were referred to the pediatric ward of JUSH for further diagnosis and treatment.

Data collection

Child Anthropometry

At birth and at 1.5, 2.5, 3.5, 4.5, and 6 mo of age, BC was measured to the nearest 0.1 g by trained research nurses using a Peapod®, an air displacement plethysmograph (ADP) previously described in detail (12).

Anthropometric measurements were taken according to WHO guidelines (23). Child weight and length were measured at birth, 1.5, 2.5, 3.5, 4.5, 6, 12, 18, 24, 36, 48, and 60 mo of age. Trained research nurses measured length to the nearest 0.1 cm in recumbent position in children <2 y using a SECA 416 length board (SECA, Hamburg, Germany) and standing position for children ≥2 y using a SECA 213 portable stadiometer. Measurements were taken in duplicate and the means were used in the analyses.

Maternal anthropometry

At delivery and at 1.5, 2.5, 3.5, 4.5, 6, 12, 18, and 24 mo after delivery, maternal height was measured using a SECA 213 portable stadiometer to the nearest 0.1 cm while maternal weight was measured using Tanita 418 (Tanita Corp.,US) bioimpedance analyzer to the nearest 0.1 kg.

Questionnaire data at baseline

Background information including sex of the child, birth order, parental education and socioeconomic status were collected at baseline by trained research nurses using structured and pre-tested questionnaires. Parental wealth index was computed from data on socioeconomic indicators, which were collected within 48 hours after delivery and combined using principal component analysis (PCA). Then, the PCA score was classified into quintiles. Possession of household items, availability of drinking water and toilet facility were considered for the wealth index. A detailed description has been provided elsewhere (19).

Breast feeding status was assessed at 3 mo based on feeding related questions since birth. Children were categorized as exclusively breastfed (only breast milk with exception of taking medication in the form of syrups, drops and oral rehydration therapy), predominant breastfed (breast milk is predominant source of food, but water and liquid drinks added and allowed to take medicine), partial breast fed (breast feeding with additional solid/complementary food added), or no breast fed according to the WHO classification (24).

Statistical analysis

Data were double entered in Epidata version 3.1 (Odense, Denmark, 2003-2008). R version 3.3.5 was used to derive the exposure variables (25). STATA/IC 12.1 (StataCorp, College Station, TX, USA) was used for analysis of outcomes. Baseline characteristics were summarized using means ± SD for continuous variables, and frequencies and percentages for categorical variables. In order to calculate height-for-age z-score (HAZ) based on the WHO growth standard, we used the "Zscore06"(26) STATA command for ages up to 60.9 mo, whereas "WHO Anthro plus" was

used for children >60.9 mo. The calculated HAZ from 12 to 60 mo was used to compare the children's growth with the WHO standard.

Derivation of exposure variables

BC data obtained from birth to 4 or 6 mo were used to derive exposure variables in terms of standardized accretion rates for FM and FFM, respectively. Correspondingly, length measurements from birth to 4 or 6 mo were used to derive length accretion rates.

The FFM accretion rate was generated from birth to 6 mo of age. Since the linear trend for FM was not fulfilled from birth to 6 mo (12), we generated the FM accretion rate from birth to 4 mo and 4 - 6 mo separately. However, as the infant gained negligible fat from 4 - 6 mo with almost zero slope, only FM from 0 - 4 mo was used as an exposure variable. Specifically, linear mixed effects models were fitted separately for each of these three variables (FM, FFM, and length) using restricted maximum likelihood (REML) estimation. All data points, except those from children having a measurement only at birth, were included in the models. Separate models were fitted for boys and girls as growth during infancy differ between boys and girls (8,12,27). The models included age in mo as a linear fixed-effects term and child-specific random intercepts and slopes for age in mo. In order to improve the precision of the BC accretion rates, the models also included fixed-effects terms adjusting for baseline differences with respect to sex of the child, birth order, head circumference, maternal and paternal schooling.

Non-standardized estimates of FM, FFM, and length accretion rates for all children were obtained from the fitted linear mixed effects models through the estimated best linear unbiased predictors (EBLUPs) of the slopes for age in mo. The estimated accretion rates were by definition of EBLUPs centered around 0 and, thus, provided a relative ranking of the children in

the study population. Standardized accretion rates were obtained by dividing accretion rates by their standard deviation and expressed as standard deviation score (SD). Standardized FFM accretion from 0 - 6 mo and FM accretion from 0 - 4 mo entered as exposure variables in subsequent statistical analyses, whereas length accretion from birth to 4 and birth to 6 mo were used as a covariate in the analyses of FM and FFM, respectively. A summary of the above description was provided in supplemental file (**Supplemental Table 1**).

Characterization of exposure variables

Non-standardized FM, FFM, and length accretion rates, i.e., the EBLUPs of the slopes of age in mo, were used to obtain actual mean growth rates from birth to 4 or 6 mo. The birth to 6 mo mean growth rates were generated by calculating the difference between the 6 months' predicted value from the birth value while birth to 4 mo growth were obtained by calculating the mean difference between predicted values at 4 mo and birth value. The FM and FFM growth rates were expressed as gram/mo and the length growth rate was expressed as centimeter/mo. One-way analysis of variance (ANOVA) and two-sample t-tests were used to examine differences in FM, FFM, and length accretion rates from birth to 4 or 6 mo between sex, birth order, parental schooling and parental wealth index.

Analysis of outcomes

Linear mixed effects models (with REML estimation) were used to investigate the association between standardized FM and FFM accretion rates and linear growth from 1 - 5 y (height, cm). The models included child-specific random intercepts and robust standard errors were used. FM and FFM (kg) at birth, standardized length accretion (from 0 - 4 mo or 0 - 6 mo, depending on exposure of interest), sex, birth order, maternal age at delivery, parental education, and

household wealth index at birth, breast feeding status at 3 mo and maternal BMI at 3 mo postpartum were all considered potential confounders based on practice in previous publications (19,28–30). To obtain estimate changes (betas) due to the exposure between time points, the models also included interaction terms between time of follow-up and exposure. The accumulated changes in association from 1 - 5 y were derived by adding the betas for each time points within the interval from 1 - 5 y.

Specifically, for both standardized FM and FFM accretion rates, three models with different levels of confounder adjustment were fitted separately. For FFM, Model 1 included the interaction between time of follow-up and the standardized FFM accretion rate adjusted for FFM (kg) at birth. Model 2: Same as Model 1 plus standardized length accretion from 0 - 6 mo, sex, birth order and breast feeding at 3 mo adjusted for. Model 3 (fully adjusted model): Same as Model 2 plus maternal age at delivery, parental wealth index and schooling at delivery, and maternal BMI at 3 mo postpartum. For FM, Model 1 included the interaction between time of follow-up and FM accretion rate adjusted for FM (kg) at birth. Model 2 and 3 included same adjustment as described for FFM. Additionally, sex-stratified analyses were carried out. Model assumptions were assessed using residual plots and QQ plots. A significance level of 0.05 was used. Unless indicated otherwise, values in the text are mean ± SDs.

Results

A total of 644 mother-infant pairs were recruited within 48 hours after birth. Of these, 10 of the neonates were excluded as they were found to be preterm. Out of 634 infants, 99 had either no BC measurement or had only one BC measurement at any of the follow-up visits up to 6 mo and, therefore, they were excluded. In total 535 children had two or more measurement of BC from

birth to 6 mo and hence were used for estimation of BC accretion rates. Of these 454 were followed up one or several times during their first 1 - 5 y and had length/height data from 1 - 5 y. A total of 354 children with 1225 observations that had complete data were used in this analysis (**Figure 1**). Background characteristics did not differ significantly between children with and without BC data (**Supplemental Table 2**).

Baseline characteristics of children and their parents are shown in **Table 1**. About 51% were males. At birth, FFM was 2.8 ± 0.3 kg, while FM was 0.2 ± 0.2 kg. At delivery, 12.1% of mothers were below 20 y with age range from 15 - 40 y. Only 31.3 % of children were exclusively breast fed at 3 mo.

FM, FFM and length accretion rates

FM, FFM, and length accretion rates from birth to 6 mo were 316 ± 97 g/mo, 429 ± 59 g/mo, and 2.8 ± 0.3 cm/mo, respectively. FM accretion from birth to 4 mo was 386 ± 112 g/month, while the length accretion rate from birth to 4 mo was 3.3 ± 0.4 cm/mo. However, the FM accretion from 4 - 6 mo was only 102 ± 46 g/mo.

As seen in **Table 2**, FFM accretion rate from 0 - 6 mo differs between sex, birth order, maternal and paternal education, while FM accretion rate from 0 - 4 mo marginally differed by birth order only. Sex and paternal education were the only variables that resulted in differences in length accretion from 0 - 6 mo (Table 2). Length accretion from birth to 4 mo differed by sex (data not shown).

Description of length/height

The mean age of the child at outcome assessment were 12.4 ± 0.6, 18.2 ± 0.7, 24.3 ± 0.6, 36.4 ± 1.2, 48.4 ± 0.9 and 59.9 ± 1.5 for age 1, 1.5, 2, 3, 4 and 5 y, respectively. Mean length/height at 1, 1.5, 2, 3, 4 and 5 y were 72.8 ± 2.9, 78.3 ± 3.2, 82.9 ± 3.6, 90.2 ± 3.9, 97.7 ± 4.1 and 104.2 ± 4.4 cm, respectively.

The HAZ from 12 - 60 mo is shown in **Figure 2**. Mean HAZ declined from 12 - 36 mo while it increased from 36 - 60 mo showing that children were growing at a slower rate compared to the WHO growth standard until 36 mo with slight catch up growth afterwards. Males had significantly lower HAZ from 12 - 24 mo (P<0.05) but not between 36 and 60 mo.

Standardized FFM accretion rate and linear growth during childhood

At 1 y

In (**Table 3,** model 1- 3), FFM accretion from 0- 6 mo of life was positively associated with length at 1 y. As expected the effect estimate of standardized FFM accretion on length at 1 y was reduced substantially if length accretion rate from 0 - 6 mo was included as a covariate, but without losing its significance. In the fully adjusted model (Table 3, model 3), each increase by 1 SD in FFM accretion rate resulted in an increase in length at 1 y by 0.64 cm (β=0.64; 95% CI: 0.19, 1.09; P=0.005).

Longitudinally over the period from 1 - 5 y

Standardized FFM accretion rate during the first 6 mo of life was positively associated with linear growth from 1 - 5 y in all models (Table 3). In the fully adjusted model (Table 3, model 3), for each increase by 1 SD in FFM accretion rate from 0 - 6 mo linear growth from 1 - 5 y

increased by 0.63 cm (β=0.63; 95% CI: 0.19, 1.07; *P*=0.005). Furthermore, FFM accretion rate from 0 - 6 mo was positively associated with linear growth from 4 - 5 y in all models (β=0.20; 95% CI: 0.03, 0.37; *P*=0.019). However, FFM accretion was not associated with linear growth from 1 - 1.5, 1.5 - 2, 2 - 3, and 3 - 4 y in any model. FFM accretion from 0 - 6 mo showed the strongest association with linear growth at 1 y and the accumulated change in association from 1 - 5 y seems to be mainly stimulated by the 1 y association (Table 3).

Standardized FM accretion rate and linear growth during childhood

At 1 y

FM accretion during the first 4 mo of life was positively associated with length at 1 y in model 1. However, the association disappeared when adjusting for length accretion rate from 0 - 4 mo, child and parental characteristics in subsequent models (**Table 4,** model 2 and 3).

Longitudinally over the period from 1 - 5 y

Standardized FM accretion rate from 0- 4 mo of life was positively associated with linear growth from 1 - 5 y in model 3 only. In the fully adjusted model (Table 4, model 3), for each increase by 1 SD in FM accretion rate from 0 - 4 mo linear growth from 1 - 5 y increased by 0.45 cm (β=0.45; 95% CI: 0.02, 0.88; *P*=0.038). However, FM accretion was not associated with linear growth in any of the periods 1 - 1.5, 1.5 - 2, 2 - 3, 3 - 4 and 4 - 5 y.

Stratified analysis

There was sex difference between FM and FFM accretion rates and linear growth from 1 - 5 y (**Supplemental Table 3 and 4**). For females, FFM accretion was positively associated with

length at 1 y in all models (Supplemental Table 3 model 1-3). For males, there were also significant associations except in the fully adjusted model (**Supplemental Table 4**, model 3).

The association between FFM accretion from 0 - 6 mo on linear growth during the period from 1 - 5 y was significant for males (β=0.92; 95% CI: 0.31, 1.53; *P*=0.003), whereas no association was seen between FFM accretion and linear growth from 1 - 5 y for females (β=0.32; 95% CI: -0.28, 0.92; *P*=0.29).

FM accretion from 0 - 4 mo was only associated with length at 1 y in model 1 for males (β=0.72; 95% CI: 0.19, 1.24; *P*=0.007); while for females no association was found. Moreover, both for males and females no association was found between FM accretion and linear growth from 1 - 5 y **(Supplemental Table 5 and 6).**

Other correlates

FFM at birth and early infancy length accretion rates were consistently and positively associated with linear growth from 1 - 5 y. Children from uneducated mother and ≥2nd born had lower linear growth from 1 - 5 y. Children from mothers with higher BMI at 3 mo postpartum had faster linear growth from 1 - 5 y. However, FM at birth, sex of the child, family wealth index, breast feeding at 3 mo, maternal age and paternal schooling were not associated with linear growth from 1 - 5 y (data not shown).

Discussion

The present study examined the association of FM and FFM accretion during early infancy with linear growth from 1 - 5 y. So far, this relation has received minimal attention in either high-income or low- and middle- income countries.

In this study, we found that FFM accretion from 0 - 6 mo was consistently and positively associated with length at 1 y and linear growth from 1 - 5 y. Early infancy FFM accretion had the strongest impact on length at 1 y. The effect of the FFM accretion rate on accumulated linear growth decreased slightly for the models with fewer covariates. Postnatal FFM tissue accretion might be related to protein intake of the infants and might mediate the effect of infancy nutritional intake on linear growth. In this regard Butte *et al* (31) suggested that FFM accretion during infancy is lower in exclusively breastfed infants when compared with formula fed (solid food, beverage and formula milk) infants. Correspondingly, Brunner *et al* showed that FFM at 4 mo was lower in children who were fed high concentrations of human milk leptin (32). However, nearly one third of the children included in this study were exclusively breastfed (neither water nor food added) at 3 mo.

Recent papers investigated the relationship between infant feeding practices with length. Fields *et al* (33) showed an inverse association between human breast milk leptin (satiety hormone) at 1 mo and infant length. Another study showed that infants who were breastfed for >1 y were shorter compared to formula fed infants at 1 y (31). The observed association between feeding pattern and linear growth in childhood might be mediated by the postnatal FFM accretion. This could be due to the effect of breast milk hormones and maternal BC during lactation (34) which could alter the infant FFM accretion and thereby affect linear growth in childhood.

FFM consists of proteins, minerals (both osseous and non-osseous) and water in the two compartment model and has been linked to a number of health outcomes in later life. In this study, we have shown that FFM accretion in infancy was positively associated with linear growth from 1 - 5 y. However, the importance of FFM in relation to bone mass in young adults

has been explored previously. In relation to this, Young *et al* (35) found that FFM assessed during early adolescence (post menorrheal) was positively associated with change in bone mass among female adolescents aged 8 - 26 y in Australia. Similarly, a longitudinal study by Bakker *et al* (36) in Amsterdam showed that FFM measured during young adulthood was positively associated with lumbar bone in young adults aged 27 - 36 y. The findings imply that FFM might operate as a proxy indicator for skeletal muscle mass in later life.

FM accretion from 0 - 4 mo showed a positive association with linear growth from 1 - 5 y after controlling for potential covariates in the fully adjusted model. The effect of FM accretion on linear growth was not particularly strong at 1 y, but instead the estimate appears to increase over the period from 1 - 5 y. A study conducted by Dewey *et al* (37) showed that infant weight-for-length was positively associated with linear growth in the first year of life across different populations in both breastfed and formula fed infants. The authors conclude that linear growth is partly regulated by body mass or fatness in infancy. However, weight-for-length is not a precise and an accurate measurement of body fatness during infancy (17). In the present study we used FM accretion from birth to 4 mo of age, as FM increased little from 4 - 6 mo or showed negligible change over the specified period (12). Although the association with FFM accretion from 0 - 6 mo was stronger than FM accretion from 0 - 4 mo with linear growth from 1 - 5 y, this study is breaking new ground in suggesting that postnatal FM tissue accretion may contribute energy to fund subsequent linear growth in childhood period. An earlier review by Norgan (38) suggested body fat as an important component serving as energy store during undernutrition or starvation. Another study by Savastano *et al* (39) showed that subcutaneous body fat helps as insulation to the body. Postnatal FM gain could be stimulated by breast milk adiponectin and n-3 long chain polyunsaturated fatty acid, as supported by other studies (32,40). This FM gain could

mediate the association of breast milk fatty acid and adiponectins levels with linear growth. However, in a high-income population, disproportionate FM gain during infant 'catch-up growth' has been linked with risk factors for chronic diseases such as type 2 diabetic and cardiovascular disease later in life (41).

Postnatal FM and FFM accretion had different patterns with linear growth from 1 - 5 y. For FFM accretion, the point estimate decreased until 2 - 3 y and then increased again afterwards with significant difference between 4 and 5 y (P=0.019). On the contrary, for FM the point estimate increased up to 2 y then decreased between 2 and 4 y, and then increased again from 4 - 5 y. These different patterns between FM and FFM might be related to the pattern of growth of children and needs further investigation.

In this study, FFM at birth had positive association with linear growth from 1 - 5 y. We have previously reported from the same cohort that FFM at birth was positively associated with height at 2 y (19). The association of linear growth in early childhood with FFM but not with FM at birth could be due to tissue accretion patterns observed in intrauterine life. FFM accretion takes place throughout pregnancy, but FM accretion mainly occur in late pregnancy (42). This study shows that both prenatal and postnatal FFM tissue accretion rates during infancy are independent predictors of linear growth from 1 - 5 y.

FM at birth was not associated with linear growth from 1 - 5 y of age. This finding is in line with our earlier finding where FM at birth showed no association with height at 2 y of age (19). Though FM at birth is an energy source for neonate growth, we did not find any association. The lack of association with FM at birth could be due to fetal FM accretion occurring mainly in the third trimester of pregnancy (42). In addition, FM at birth was very small with little or no

changes over the duration thus the data were all clustered together resulting in lower ability to detect association.

Our study was consistent with previous research in identifying other covariates as predictors of linear growth in early childhood. Children who had higher length gain during infancy (43) and who had mother with higher BMI 3 mo postpartum (13,28,29,44) had faster linear growth from 1 - 5 y, whereas children who were $\geq 2^{nd}$ born (30) and from uneducated mothers (45) had lower linear growth from 1 - 5 y.

Strengths and Limitations

This study is the first of its kind to examine the longitudinal association of BC accretion patterns in infancy with linear growth during childhood. We used ADP to repeatedly measure BC during early infancy which is known to be an accurate and precise BC measurement in infancy (7), and measured linear growth from 1 - 5 y. We calculated BC accretion rates by predicting BC using all data points except those having only birth measurement. This approach is helpful to obtain precise BC accretion rates for all children who had at least 2 measurements from the 6 measurement times and thereby increases the power in the prediction model. Furthermore, we used mixed effects models to account for random effects.

The observed association between FM and FFM accretion with linear growth could be confounded by other potential confounders not accounted for. First, as the study enrolled mothers immediately after delivery, we could not assess the pre-pregnancy and pregnancy related data such as anthropometry, maternal dietary intake during early gestation and pregnancy weight gain which could affect the birth BC. Secondly, confounding by morbidity and complementary feeding during infancy were not considered which could have potential effect on

postnatal BC. In principle, loss to follow-up could also have affected our findings. However, dropouts were most likely unrelated to having been in the study (observations were missing at random) as we found no significant differences between children who had BC measurements and those who did not. Therefore, the use of linear mixed models ensured unbiased estimates of the effects of the exposures. Further, we carried out available-case analyses using linear mixed models such that all measurements recorded between 1 - 5 y of age were utilized despite the fact that there was some variation in the number of measurements from time point to time point. The main reasons for this variation were due to participants moved away during the follow-up period, changed or lost their telephone address, and not willing to come for examination. These reasons are not likely to cause systematic bias and we therefore assume that missing data were missing at random, implying that trends over time remained unchanged also for time points where data were missing, i.e., we could predict trends based on the observed data only. Length/height measurement errors could also affect the finding however, two measurements of length/height were taken to minimize the error and mean values were used in the analysis. Thus, the finding should be interpreted with caution by considering the aforementioned limitation of the study.

Conclusion

Both FM and FFM accretion in infancy were positively associated with linear growth from 1 - 5 y. This indicates that both postnatal fat and fat-free tissues are concurrently involved in influencing linear growth during childhood. In addition, FFM at birth had the strongest positive association with linear growth from 1 - 5 y of age, whereas FM did not show any such association. This finding highlights the importance of promoting and advising healthy maternal and infant dietary intake and nutrition during the prenatal and postnatal period.

Future studies should include pre-pregnancy and pregnancy related data, infancy and childhood dietary intake and morbidity. Possible mechanisms underlying the association between prenatal and postnatal BC accretion rates with childhood linear growth should be studied. In addition, further long-term follow-up of this and other cohorts study is needed to assess the role of FM and FFM accretion during early infancy on childhood BC and cardio-metabolic risk.

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References

1. Victora CG, Adair L, Fall C, Hallal PC, Martorell R, Richter L, Sachdev HS, Maternal and Child Undernutrition Study Group. Maternal and child undernutrition: consequences for adult health and human capital. Lancet 2008;371:340–57.

2. Wei C, Gregory JW. Physiology of normal growth. Paediatr Child Health 2009;19:236–40.

3. Shrimpton R, Victora CG, de Onis M, Lima RC, Blössner M, Clugston G. Worldwide timing of growth faltering: implications for nutritional interventions. Pediatrics 2001;107:E75.

4. Victora CG, de Onis M, Hallal PC, Blössner M, Shrimpton R. Worldwide timing of growth faltering: revisiting implications for interventions. Pediatrics 2010;125:e473-480.

5. Oliveira VA de, Assis AMO, Pinheiro SMC, Barreto ML. Determinants of weight and linear growth deficits in children under two years of age. Rev Saúde Pública 2006;40:874–82.

6. Mamabolo RL, Alberts M, Steyn NP, Delemarre-van de Waal HA, Levitt NS. Prevalence and determinants of stunting and overweight in 3-year-old black South African children residing in the Central Region of Limpopo Province, South Africa. Public Health Nutr 2005;8:501–8.

7. Ellis KJ, Yao M, Shypailo RJ, Urlando A, Wong WW, Heird WC. Body-composition assessment in infancy: air-displacement plethysmography compared with a reference 4-compartment model. Am J Clin Nutr 2007;85:90–5.

8. Rigo J, Ziegler EE. Protein and energy requirements in infancy and childhood. Nestlé Nutr Workshop Ser Pediatr Program. Nestec Ltd., Vevey/S. Karger AG, Basel Switzerland. 2006;58:65–78.

9. Andres A, Hull HR, Shankar K, Casey PH, Cleves MA, Badger TM. Longitudinal body composition of children born to mothers with normal weight, overweight, and obesity. Obesity 2015;23:1252–8.

10. Toro-Ramos T, Paley C, Pi-Sunyer F, Gallagher D. Body composition during fetal development and infancy through the age of 5 years. Eur J Clin Nutr 2015;69:1279–89.

11. Paley C, Hull H, Ji Y, Toro-Ramos T, Thornton J, Bauer J, Matthews P, Yu A, Navder K, Dorsey K, et al. Body fat differences by self-reported race/ethnicity in healthy term newborns. Pediatr Obes 2016;11:361–8.

12. Andersen GS, Girma T, Wells JC, Kæstel P, Leventi M, Hother A-L, Michaelsen KF, Friis H. Body composition from birth to 6 mo of age in Ethiopian infants: reference data obtained by air-displacement plethysmography. Am J Clin Nutr 2013;98:885–94.

13. Kulkarni B, Mamidi RS, Balakrishna N, Radhakrishna KV. Body composition assessment in infancy and early childhood: comparison of anthropometry with dual-energy x-ray absorptiometry in low-income group children from India. Eur J Clin Nutr 2014;68:658–63.

14. Lourenço BH, Villamor E, Augusto RA, Cardoso MA. Determinants of linear growth from infancy to school-aged years: a population-based follow-up study in urban Amazonian children. BMC Public Health 2012;12:265.

15. Padonou G, Le Port A, Cottrell G, Guerra J, Choudat I, Rachas A, Bouscaillou J, Massougbodji A, Garcia A, Martin-Prevel Y. Factors associated with growth patterns from birth to 18 months in a Beninese cohort of children. Acta Trop 2014;135:1–9.

16. Adair LS, Fall CH, Osmond C, Stein AD, Martorell R, Ramirez-Zea M, Sachdev HS, Dahly DL, Bas I, Norris SA, et al. Associations of linear growth and relative weight gain during early life with adult health and human capital in countries of low and middle income: findings from five birth cohort studies. Lancet 2013;382:525–34.

17. Wells JCK. A critique of the expression of pediatric body composition data. Arch Dis Child 2001;85:67–72.

18. Espo M, Kulmala T, Maleta K, Cullinan T, Salin ML, Ashorn P. Determinants of linear growth and predictors of severe stunting during infancy in rural Malawi. Acta Paediatr 2002;91:1364–70.

19. Admassu B, Wells JCK, Girma T, Andersen GS, Owino V, Belachew T, Michaelsen KF, Abera M, Wibaek R, Friis H, et al. Body composition at birth and height at 2 years: a prospective cohort study among children in Jimma, Ethiopia. Pediatr Res 2017;82:209–214

20. Abera M, Tesfaye M, Girma T, Hanlon C, Andersen GS, Wells JC, Admassu B, Wibaek R, Friis H, Kæstel P. Relation between body composition at birth and child development at 2 years of age: a prospective cohort study among Ethiopian children. Eur J Clin Nutr [Internet]. 2017 Sep 27 [cited 2017 Sep 29]; Available from: http://www.nature.com/ejcn/journal/vaop/ncurrent/full/ejcn2017129a.html?foxtrotcallback=true

21. Andersen GS, Girma T, Wells JCK, Kæstel P, Michaelsen KF, Friis H. Fat and fat-free mass at birth: air displacement plethysmography measurements on 350 Ethiopian newborns. Pediatr Res 2011;70:501–6.

22. Ballard JL, Khoury JC, Wedig K, Wang L, Eilers-Walsman BL, Lipp R. New Ballard Score, expanded to include extremely premature infants. J Pediatr 1991;119:417–23.

23. World Health Organization. Training course on child growth assessment. Geneva, WHO; 2008.

24. World Health Organization. The World Health Organization's infant feeding recommendation [Internet]. 2017. [cited 2017 July 7]. Available from: http://www.who.int/nutrition/topics/ infant feeding recommendation/en/.

25. R Development Core Team. R: A language and environment for statistical computing. The R Foundation for Statistical Computing, Vienna, Austria. [Internet]. 2016. [cited 2017 May 16]. Available from: http://www.R-project.org/.

26. zscore06: Stata command for the calculation of anthropometric z-scores using the 2006 WHO child growth standards http://www.stata.com/statalist/archive/2011-04/msg01386. html. 2011.

27. Fields DA, Krishnan S, Wisniewski AB. Sex differences in body composition early in life. Gend Med 2009;6:369–75.

28. Reynolds RM, Osmond C, Phillips DIW, Godfrey KM. Maternal BMI, parity, and pregnancy weight gain: influences on offspring adiposity in young adulthood. J Clin Endocrinol Metab 2010;95:5365–9.

29. Widen EM, Bentley ME, Kayira D, Chasela CS, Jamieson DJ, Tembo M, Soko A, Kourtis AP, Flax VL, Ellington SR, et al. Maternal weight loss during exclusive breastfeeding is associated with reduced weight and length gain in daughters of HIV-infected Malawian women. J Nutr 2013;143:1168–75.

30. Myrskylä M, Silventoinen K, Jelenkovic A, Tynelius P, Rasmussen F. The association between height and birth order: evidence from 652 518 Swedish men. J Epidemiol Community Health 2013;571–7.

31. Butte NF, Wong WW, Hopkinson JM, Smith EO, Ellis KJ. Infant feeding mode affects early growth and body composition. Pediatrics 2000;106:1355–66.

32. Brunner S, Schmid D, Zang K, Much D, Knoeferl B, Kratzsch J, Amann-Gassner U, Bader BL, Hauner H. Breast milk leptin and adiponectin in relation to infant body composition up to 2 years. Pediatr Obes 2015;10:67–73.

33. Fields DA, George B, Williams M, Whitaker K, Allison DB, Teague A, Demerath EW. Associations between human breast milk hormones and adipocytokines and infant growth and body composition in the first 6 months of life. Pediatr Obes 2017;78–85.

34. Kulkarni B, Shatrugna V, Nagalla B, Rani KU. Regional body composition changes during lactation in Indian women from the low-income group and their relationship to the growth of their infants. J Am Coll Nutr 2011;30:57–62.

35. Young D, Hopper JL, Macinnis RJ, Nowson CA, Hoang NH, Wark JD. Changes in body composition as determinants of longitudinal changes in bone mineral measures in 8 to 26-year-old female twins. Osteoporos Int 2001;12:506–15.

36. Bakker I, Twisk JWR, Van Mechelen W, Kemper HC. Fat-free body mass is the most important body composition determinant of 10 year longitudinal development of lumbar bone in adult men and women. J Clin Endocrinol Metab 2003;88:2607–13.

37. Dewey KG, Hawck MG, Brown KH, Lartey A, Cohen RJ, Peerson JM. Infant weight-for-length is positively associated with subsequent linear growth across four different populations. Matern Child Nutr 2005;1:11–20.

38. Norgan NG. The beneficial effects of body fat and adipose tissue in humans. Int J Obes Relat Metab Disord J Int Assoc Study Obes 1997;21:738–46.

39. Savastano DM, Gorbach AM, Eden HS, Brady SM, Reynolds JC, Yanovski JA. Adiposity and human regional body temperature. Am J Clin Nutr 2009;90:1124–31.

40. Much D, Brunner S, Vollhardt C, Schmid D, Sedlmeier EM, Brüderl M, Heimberg E, Bartke N, Boehm G, Bader BL, et al. Breast milk fatty acid profile in relation to infant growth and body composition: results from the INFAT study. Pediatr Res 2013;74:230–7.

41. Dulloo AG, Jacquet J, Seydoux J, Montani JP. The thrifty 'catch-up fat' phenotype: its impact on insulin sensitivity during growth trajectories to obesity and metabolic syndrome. Int J Obes 2006;30:S23–35.

42. Haggarty P. Placental regulation of fatty acid delivery and its effect on fetal growth—a review. Placenta 2002;23:S28–38.

43. Krishnaveni GV, Veena SR, Srinivasan K, Osmond C, Fall CH. Linear growth and fat and lean tissue gain during childhood: associations with cardiometabolic and cognitive outcomes in adolescent Indian children. PLoS ONE 2015;10: e0143231.

44. Brown KH, Robertson AD, Akhtar NA. Lactational capacity of marginally nourished mothers: infants' milk nutrient consumption and patterns of growth. Pediatrics 1986;78:920–7.

45. Svefors P, Rahman A, Ekström EC, Khan AI, Lindström E, Persson LÅ, Selling KE. Stunted at 10 years. Linear growth trajectories and stunting from birth to pre-adolescence in a rural Bangladeshi cohort. PLOS ONE 2016;11:e0149700.

TABLE 1 Birth, parental and dyad characteristics at 3 mo postpartum in the iABC cohort (*n* = 454)[1, 2]

		n
Birth characteristics[3]		
Weight, kg	3.1 ± 0.4	453
Length, cm	49.2 ± 1.9	453
Fat-free mass, kg	2.8 ± 0.3	452
Fat mass, kg	0.2 ± 0.2	452
Male sex	229 (50.4)	454
Birth order		447
1st	233 (52.1)	
2nd	108 (24.2)	
≥3rd	106 (23.7)	
Parental characteristics at birth[3]		
Maternal age at delivery, y	24.2 ± 4.6	446
Maternal education		452
None	29 (6.4)	
Primary	277 (61.3)	
≥Secondary	146 (32.3)	
Paternal education		440
None	11 (2.5)	
Primary	261 (59.3)	
≥Secondary	163 (38.2)	
Dyad characteristics 3 mo postpartum[4]		
Breast feeding status at 3 mo of age		390
Exclusive	122 (31.3)	
Predominant	163 (41.8)	
Partial (breast milk and food)	96 (24.6)	
Not breastfed	9 (2.3)	
Maternal BMI 3 mo postpartum, kg/m^2	22.2 ± 3.4	383

[1] The numbers are mean ± SD or number (%).

[2] BMI; body mass index, iABC; infant anthropometry and body composition cohort.

[3] Data were obtained within 48 hours after delivery.

[4] Data were obtained at 3 mo (± 7 d).

TABLE 2 Description of FFM, FM and length accretion rates by sex, birth order, parental education and household wealth index[1]

Variables	*n*	FFM 0 - 6 (g/mo)[4]	*p*	*n*	FM 0 - 4 (g/mo)[4]	*p*	*n*	Length 0 - 6 (cm/mo)[4]	*p*
Sex[2]			<0.001			0.67			<0.001
Male	171	449 ± 61		213	388 ± 117		203	2.9 ± 0.3	
Female	178	409 ± 49		211	383 ± 106		201	2.7 ± 0.2	
Birth order[3]			0.009			0.053			0.07
1st	176	437 ± 59		209	398 ± 111		205	2.9 ± 0.3	
2nd	89	414 ± 59		112	382 ± 119		104	2.8 ± 0.3	
3rd	84	426 ± 56		103	366 ± 102		95	2.9 ± 0.3	
Maternal education[3]			0.001			0.67			0.06
No education	27	389 ± 56		30	373 ± 111		30	2.7 ± 0.2	
Primary	216	429 ± 59		265	385 ± 111		245	2.8 ± 0.3	
≥Secondary	106	438 ± 56		129	392 ± 113		129	2.9 ± 0.3	
Paternal education[3]			0.010			0.39			0.043
No education	9	397 ± 65		11	385 ± 126		10	2.8 ± 0.3	
Primary	204	423 ± 58		250	380 ± 107		235	2.8 ± 0.3	
≥Secondary	136	439 ± 58		163	395 ± 117		159	2.9 ± 0.3	
Wealth index quintile[3]			0.46			0.85			0.86
1st (lowest)	78	421 ± 63		86	385 ± 105		88	2.8 ± 0.3	
2nd	79	423 ± 66		96	375 ± 105		89	2.8 ± 0.3	
3rd	79	432 ± 52		107	388 ± 124		96	2.8 ± 0.3	
4th	64	430 ± 56		72	388 ± 115		74	2.9 ± 0.3	
5th (highest)	43	439 ± 52		54	395 ± 111		50	2.9 ± 0.3	

[1] FFM; fat-free mass, FM; fat mass.
[2] Two-sample t-test.
[3] One-way ANOVA.
[4] The numbers are mean ± SD.

TABLE 3 Standardized FFM accretion rate from 0 - 6 mo as a correlate of linear growth from 1 - 5 y in the iABC cohort (n = 354)[1]

	(Height, cm)[2]		
	Model 1[3]	Model 2[4]	Model 3[5]
FFM accretion rate from 0 - 6 mo (SD)			
at 1 y	1.16 (0.75,1.56)***	0.76 (0.31,1.21)***	0.64 (0.19,1.09)**
1 - 1.5 y	0.25 (-0.16,0.65)	0.24 (-0.17,0.64)	0.28 (-0.12,0.67)
1.5 - 2 y	0.16 (-0.18,0.50)	0.17 (-0.17,0.52)	0.14 (-0.20,0.48)
2 - 3 y	-0.08 (-0.42,0.26)	-0.06 (-0.40,0.27)	-0.09 (-0.42,0.24)
3 - 4 y	0.07 (-0.16,0.29)	0.07 (-0.16,0.29)	0.10 (-0.12,0.31)
4 - 5 y	0.20 (0.05,0.36)**	0.20 (0.05,0.35)**	0.20 (0.03,0.37)*
Accumulated change in association (SD)			
1 - 5 y	0.60 (0.18,1.03)**	0.61 (0.19,1.03)**	0.63 (0.19,1.07)**
FFM at birth, kg	2.36 (1.25,3.47)***	2.31 (1.17,3.44)***	2.21 (1.09,3.33)***

[1] BMI; body mass index, FFM; fat-free mass, iABC; infant anthropometry and body composition cohort, SD; standard deviation.

[2] Data are shown as regression coefficients (β) and 95% confidence intervals obtained from linear mixed effect model. *P <0.05, **P <0.01, ***P <0.001.

[3] Model 1: Interaction between time of follow-up and FFM accretion rate from 0 - 6 mo adjusted for FFM at birth.

[4] Model 2: Model 1 plus sex, standardized length accretion rate from 0 - 6 mo, birth order and breast feeding at 3 mo.

[5] Model 3: Model 2 plus maternal age at delivery, parental wealth index and schooling at delivery, and maternal BMI 3 mo postpartum.

TABLE 4 Standardized FM accretion rate from 0 - 4 mo as a correlate of linear growth from 1 - 5 y in the iABC cohort (n = 354)[1]

	(Height, cm)[2]		
	Model 1[3]	Model 2[4]	Model 3[5]
FM accretion rate from 0 - 4 mo (SD)			
at 1 y	0.54 (0.15,0.94)**	0.05 (-0.35,0.46)	0.10 (-0.34,0.53)
1 - 1.5 y	0.13 (-0.27,0.53)	0.12 (-0.27,0.52)	0.13 (-0.27,0.53)
1.5 - 2 y	0.23 (-0.08,0.55)	0.24 (-0.08,0.55)	0.23 (-0.08,0.54)
2 - 3 y	-0.08 (-0.38,0.22)	-0.06 (-0.36,0.24)	0.003 (-0.30,0.31)
3 - 4 y	-0.05 (-0.28,0.18)	-0.06 (-0.29,0.17)	-0.04 (-0.28,0.21)
4 - 5 y	0.12 (-0.06,0.30)	0.12 (-0.06,0.30)	0.12 (-0.09,0.33)
Accumulated change in association (SD)			
1 - 5 y	0.36 (-0.04,0.77)	0.36 (-0.05,0.76)	0.45 (0.02,0.88)*
FM at birth, kg	1.37 (-1.43,4.17)	1.42 (-1.15,3.98)	0.30 (-2.17,2.76)

[1] BMI; body mass index, FM; fat mass, iABC; infant anthropometry and body composition cohort, SD; standard deviation.

[2] Data are shown as regression coefficients (β) and 95% confidence intervals obtained from linear mixed effect model. *P <0.05, **P <0.01, ***P <0.001.

[3] Model 1: Interaction between time of follow-up and FM accretion rate from 0 - 4 mo adjusted for FM at birth.

[4] Model 2: Model 1 plus sex, standardized length accretion rate from 0 - 4 mo, birth order and breast feeding at 3 mo.

[5] Model 3: Model 2 plus maternal age at delivery, parental wealth index and schooling at delivery, and maternal BMI 3 mo postpartum.

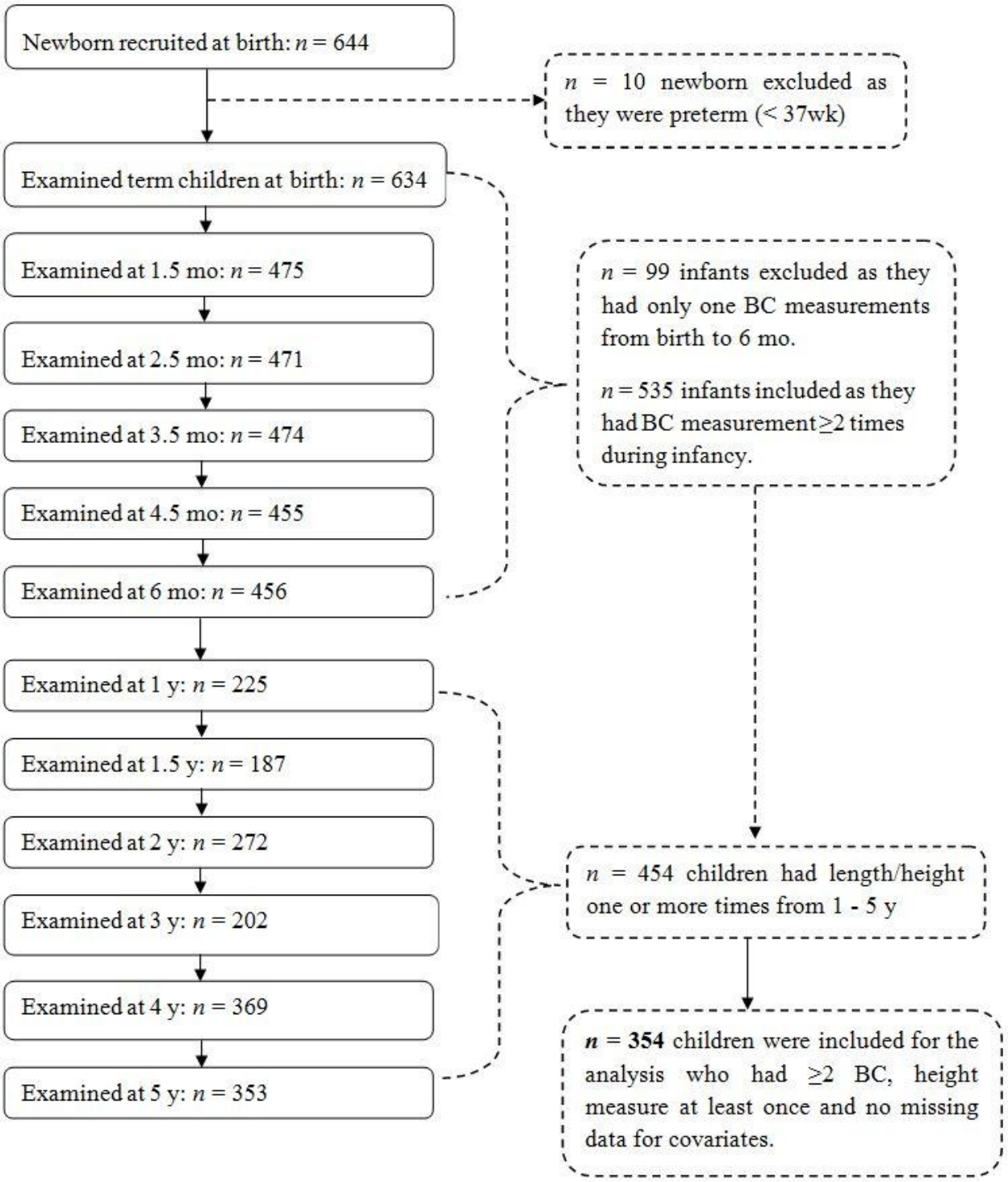

FIGURE 1 Flow diagram showing selection of children for the present study.

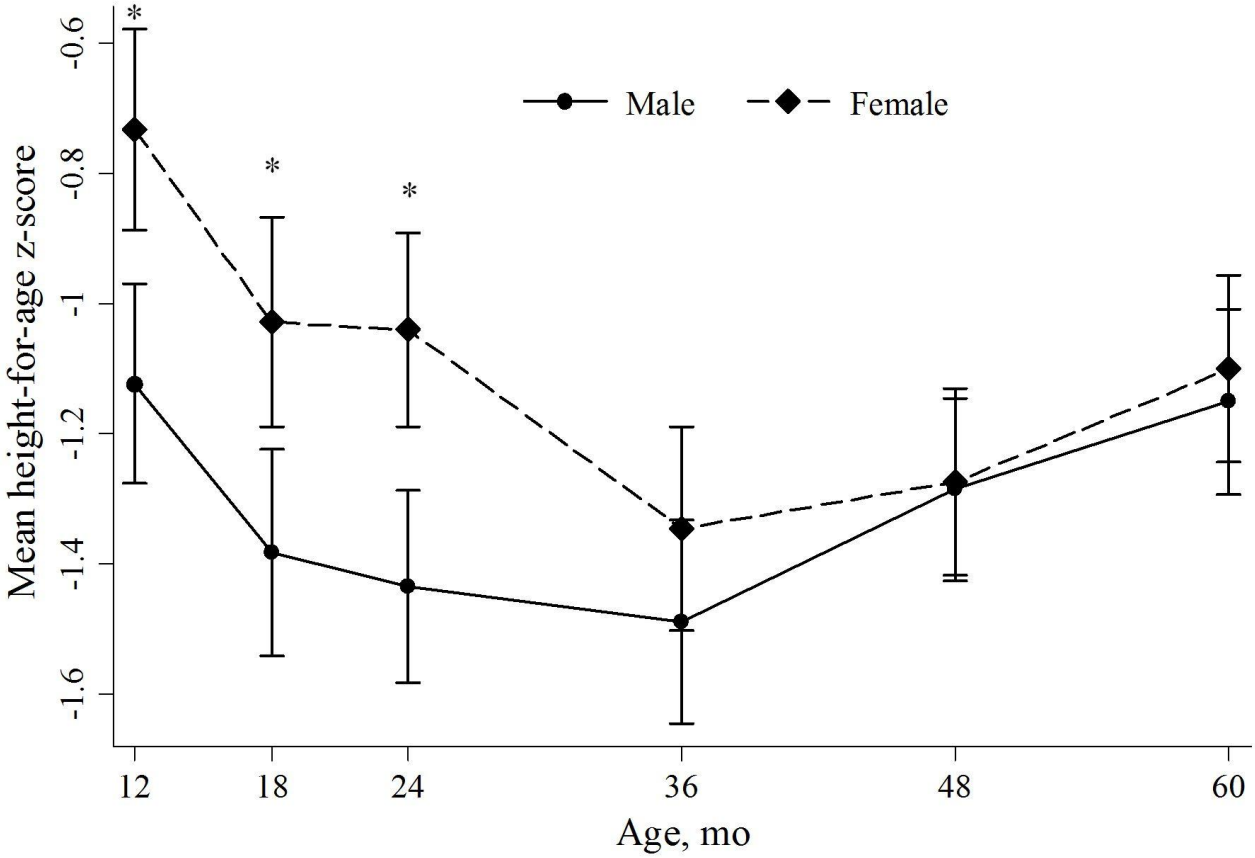

FIGURE 2 Comparing height-for-age z-score from 12 - 60 mo by sex among children included in the current study. Values are mean and 95% CI, n = 454. P value shows significant difference between male and female in height-for-age z-score, * P <0.05.