Title

Body mass index trajectories in early childhood in relation to cardiometabolic risk profile and body composition at 5 years of age

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Short running title

BMI trajectories and metabolic status at 5 years

Abbreviations

Body composition (BC), fat mass (FM), fat mass index (FMI), fat-free mass (FFM), fat-free mass index (FFMI), International Wealth Index (IWI), latent class trajectory (LCT), standard deviation (SD), World Health Organization (WHO).

Clinical Trial Registry

The birth cohort is registered in ISRCTN (https://www.isrctn.com/): identifier

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Abstract

Background: Both impaired and accelerated postnatal growth have been associated with adult risk of obesity and cardiometabolic diseases like type-2-diabetes and cardiovascular disease. However, the timing of the onset of cardiometabolic changes and specific growth trajectories linking early growth with later disease risk are not well understood.

Objective: To identify distinct trajectories of body mass index (BMI) growth from 0-5 years and examine their associations with markers of cardiometabolic risk at age 5 years.

Design: In a prospective birth cohort study of 453 healthy and term Ethiopian children with BMI assessed a median of 9 times during follow-up, we identified subgroups of distinct BMI trajectories in early childhood using latent class trajectory modelling. Associations of the identified growth trajectories with cardiometabolic markers and body composition at 5 years were analyzed using multiple linear regression analysis in four adjustment models for each outcome.

Results: Four heterogeneous BMI growth trajectories were identified: “stable low BMI” (19.2%), “normal BMI” (48.8%), “rapid catch-up to high BMI” (17.9%), and “slow catch-up to high BMI” (14.1%). Compared with the “normal BMI” trajectory, children in the “rapid catch-up to high BMI” trajectory had higher triglycerides (range of β-coefficients in model 1-4: 19-21%), C-peptide (23-25%), fat mass (0.48-0.60 kg) and fat-free mass (0.50-0.77 kg) across the four adjustment models. Children in the “stable low BMI” class had lower low-density lipoprotein cholesterol (0.14-0.17 mmol/L), high density lipoprotein cholesterol (0.05-0.09 mmol/L), fat mass (0.60-0.64 kg), fat-free mass (0.35-0.49 kg), but higher triglycerides (11-13%).

Conclusions: The development of obesity and cardiometabolic risk may be established already in early childhood and thus provides further basis for timely interventions targeted at young children from low-income countries with unfavorable growth patterns.

Key words

body composition; cohort study; child; developmental origins of health and disease; growth; latent class trajectory modelling; non-communicable diseases; Sub-Saharan Africa.

Introduction

The prevalence of childhood obesity is a major threat to public health worldwide (1, 2). While some progress has been seen in high-income countries, many low-income countries experience a dual burden of malnutrition where rising prevalence of childhood overweight coincides with high pre- and postnatal under-nutrition (3, 4). Both ends of this spectrum of malnutrition have been associated with adult risk of obesity, type 2 diabetes and cardiovascular disease (4, 5), and studies have shown that growth patterns in early childhood is associated with these outcomes (6-8).

Most of these studies have examined associations of variability in early-life nutrition with adult risk of disease and are therefore not designed to address the timing of onset of the cardiometabolic adaptations that may occur already in childhood (9-11). Another issue is that early-life growth is typically assessed at one or few timepoints in childhood. However, as childhood growth is a dynamic process and diverging longitudinal growth patterns are likely to associate differently with later cardiometabolic outcomes, studies with detailed and repeated assessment of body size are needed. Additionally, very few of the studies that have explored longitudinal growth patterns and their associations with adiposity and cardiometabolic risk were conducted in low-income populations. These populations are nonetheless likely to be particularly affected as a result of the dual burden of malnutrition seen in many countries currently undergoing rapid economic and nutritional transition (12). Stratification of children into different trajectories of growth associated with different levels of adiposity and cardiometabolic risk profiles may therefore provide an opportunity to identify early and targeted interventions in children from low-income populations.

Thus, we aimed to identify distinct trajectories of body mass index (BMI) growth from birth to 5 years and to estimate patterns of fat mass (FM) and fat-free mass (FFM) growth in infancy for each of the identified BMI trajectories. Furthermore, we examined the relationships of the identified BMI trajectories with cardiometabolic markers and BC at 5 years of age in a low-income urban population.

Subjects and methods

Study setting and participants

We used data from the Infant Anthropometry and Body Composition (iABC) birth cohort study, as described elsewhere (13, 14). Briefly, mother-child pairs were recruited within 48 hours after delivery at Jimma University Specialized Hospital, Jimma, Ethiopia between December 2008 and October 2012. Eligible mothers were residing in Jimma town and had given birth to a term (gestation >37 weeks) and apparently healthy child above 1500g without congenital malformations. From birth to 60 months of age, we planned a total of 12 visits (0, 1.5, 2.5, 3.5, 4.5, 6, 12, 18, 24, 36, 48, 60 months). The modelling of BMI trajectories from 0-5 years included children with BMI assessed at the birth visit and at least one time in each of the periods between the 1.5-6- and 12-60-months visits. For the subsequent regression analysis with the 5-year outcomes, the analyses were restricted to children with a valid measurement of the specific outcome in question and full covariate information. As some participants refused blood sampling or were unable to deliver enough blood for analysis of all biomarkers, the number of children included in the regression analyses differed by the specific outcome.

Data collection

Anthropometry and BC

Length was measured to the nearest 0.1 cm in recumbent position using a SECA 416 Infantometer for children below 2 years and in standing position using a SECA 213 portable height measurer for children above 2 years (SECA, Hamburg, Germany). Waist circumference was measured to the nearest 0.1 cm in standing position with feet together midway between

the iliac crest and lowest costal margin using a non-stretchable measuring tape. Weight from birth to 6 months was measured to the nearest 0.1 g using the built-in electronic scale of the infant air displacement plethysmography (ADP) instrumentation (PEA POD, COSMED, Rome, Italy), from 1-3 years to the nearest 0.1 kg using an electronic UNICEF scale (SECA, Hamburg, Germany) and from 4-5 years to the nearest 1 g using the attached electronic scale of the child/adult ADP instrumentation (BOD POD, COSMED, Rome, Italy). FM and FFM from birth to 6 months and at 5 years were measured using ADP in the PEA POD and the BOD POD with a pediatric chair insert, respectively. Both ADP systems are accurate, precise, feasible and safe methods for assessment of BC in infants and children (15, 16), and in the present cohort, the PEA POD has previously been validated against a 3-component model with deuterium dilution (13). In brief, these BC systems estimate total body density from weight and volume measurements, and assuming known values of the density of FM and FFM, calculate the proportion of FM in body weight. The theory and methods behind the PEA POD and BOD POD are described in detail elsewhere (17, 18). A full BC assessment lasted 5-10 minutes and during the volume measurements, the child was placed on a plastic bed (PEA POD) or in a pediatric chair insert (BOD POD) in an enclosed test chamber, not wearing any clothes besides a swim cap (PEA POD and BOD POD) or tight fitted underpants (BOD POD). Calibration of the BC equipment was performed each morning. All the calculations were performed by the built-in computer (PEA POD software version 3.3.0 and BOD POD software version 5.2.0). BMI, FM index (FMI), and FFM index (FFMI) were calculated by dividing weight, FM, and FFM with the squared height in meter, respectively.

Blood pressure at 5 years

After relaxing for 5 minutes, systolic and diastolic blood pressure were measured in sitting position using a blood pressure device with age-appropriate cuffs (Pressostabil model, Welch Allyn Inc., Skaneateles Falls, USA). Measurements were done in duplicate and averaged.

Cardiometabolic markers at 5 years

A 2 ml venous blood sample was drawn from the antecubital fossa as the last element of assessments of the child following a minimum of 3 hours of fasting. Glucose concentrations were measured in whole blood using the HemoCue Glucose 201 RT System (HemoCue, Ängelholm, Sweden). Glycosylated haemoglobin (HbA1c, mmol/mol) was measured on whole blood using a DCCT aligned Quo-Test® A1c Analyzer (EKF Diagnostics, Cardiff, Wales). Subsequently, serum was obtained by centrifuging the whole blood sample, aliquoted in 3x0.4 mL and frozen at -80°C until analyzed. The serum samples were analyzed at the Ethiopian Public Health Institute, using the module c501 of the COBAS 6000 analyzer (Roche Diagnostics International Ltd, Rotkreuz, Switzerland) for total-, LDL-, and HDL cholesterol, and triglyceride concentrations (all lipids in mmol/L), and the module e601 for insulin (µU/mL), and C-peptide (ng/mL). The homeostasis model assessment of insulin resistance index (HOMA-IR) was calculated as insulin × glucose / 22.5 (19).

Covariates

Information on birth order, child's sex, gestational age, maternal age, educational level, and socioeconomic status of the family was obtained at the birth visit. Maternal postpartum height was measured to the nearest 0.1 cm using a Seca 214 Stadiometer (SECA, Hamburg,

Germany). Birth order was self-reported as the number of previous pregnancies. Gestational age of the new-born was assessed by trained research nurses using the New Ballard Score test (20). The International Wealth Index (IWI) was used to assess socioeconomic status of the family. The IWI assesses the material well-being of households in low- and middle-income countries (21), and includes information of 12 material well-being dimensions, including seven household assets, access to two public services and three characteristics of the house. The IWI ranges from 0 to 100 (highest wealth). Information on breastfeeding status was assessed at 4 to 6 months post-partum and divided into four categories: exclusive (no other foods given), almost exclusive (no other foods given except water), predominant (breast milk as primary food) and partial/no (breast milk not the primary food/not breast feeding) (22).

Ethics

Ethical approval was granted by the Jimma University Ethical Review Committee (Ref. no. RPGC/279/2013). Written informed consent was obtained from parents or caregivers of all eligible children. Children with any medical condition observed by the research nurses were referred in accordance with local clinical guidelines.

Statistical methods

All descriptive data are presented as mean (SD) or median (interquartile range) for continuous variables and percentages for categorical variables. P-values <0.05 were considered statistically significant. All analyses were carried out in R version 3.4.1 (The R foundation for Statistical Computing).

Identification of latent BMI trajectories in childhood

Heterogeneity in repeated measures of BMI was analyzed using latent class trajectory (LCT) modelling to identify distinct subgroups of children with similar trajectories of BMI growth from 0-5 years (23, 24). We ran a series of LCT models with various specifications of BMI as a function of age and number of subgroups (classes). As described in detail in the **Supplemental Methods,** the best fitting model according to our "a priori" criteria was obtained with a four class model specified with natural cubic splines with knot points at 0, 3, 6, 24, 48, and 60 months. Using the class assignments from the LCT analysis, we re-estimated the BMI trajectories from 0-60 months for girls and boys separately to assess if there were any sex differences in BMI growth for each of the four assigned classes.

FMI and FFMI growth in infancy

For each of the identified BMI trajectory classes, we applied mixed-effects modelling to estimate the corresponding mean growth in FMI and FFMI from 0-6 months of age. We required children to have a minimum of three FMI and FFMI measurements during the first 0-6 months to be included in the modelling. The FMI and FFMI as a function of age were modelled separately and fitted with natural cubic splines with knot points at ages 0, 3 and 6 months.

Associations of BMI trajectories with BC and cardiometabolic markers at 5 years

We analyzed the relationship of the identified BMI trajectory classes with BC and cardiometabolic markers at 5 years using multiple linear regression analysis. The "normal BMI" trajectory class was used as reference. In all analyses, we log transformed outcomes

where the corresponding model residuals were not normally distributed, which resulted in normally distributed model residuals. The resulting estimates were back transformed and presented on the relative scale as percentwise change. We ran four separate models for each outcome. Model 1 was adjusted for sex, birth order, and gestational age. Model 2 was additionally adjusted for the child's exact age at the 5-year visit, maternal age at delivery, maternal postpartum height, maternal educational status, and family socioeconomic status (IWI). Model 3 was additionally adjusted for child birth weight. Model 4 was additionally adjusted for child BMI at the 5-year visit. Since BMI comprises both FM and FFM, the analyses of the outcomes FM and waist circumference were adjusted for the lean component of BMI (FFM and height at 5 years) instead of BMI in model 4. Similarly, the analysis of the outcome FFM was adjusted for FM and height at 5 years (the fat component of BMI). We compared the estimated associations across the four models for a given outcome and exposure using a complete case approach, limiting the analyses to data with complete information on all covariates in model 4. In further analyses, we accounted for multiple testing using the Benjamini-Hochberg approach (25), where the number of tests was set to 45 (15 outcomes for three exposure groups) (**Supplemental Figure 1**). To account for breastfeeding, we ran sensitivity analyses on a smaller sample with available information on breastfeeding status at 4 to 6 months post-partum (**Supplemental Figure 2**).

Results

At birth, the mothers were on average 24.6 years and 49% were primiparous. Half of the mothers had completed primary school or higher and 94% were breast feeding either exclusively, almost exclusively or predominantly during the first 4-6 months after birth (**Supplemental Table 1**). Low birth weight was seen in 9.4% of the children. At 5 years, the children were on average more than 1.2 SD scores below the World Health Organization (WHO) international growth standards for height, 15.3% were stunted, and 5.9% were overweight or obese. The average BMI of 15.0 kg/m^2 at 5 years was similar to the average of the WHO international growth standards (**Supplemental Table 2**) (26).

Latent BMI trajectories in early childhood

A total of 453 children were included in the modelling of the BMI trajectories (**Figure 1**). The children had their BMI assessed a median (interquartile range) of 9 (8-10) times during follow-up period, contributing a total of 3952 observations to LCT modelling. We identified four distinct BMI trajectory classes from birth to 5 years (**Figure 2** and **Supplemental Figure 3**): Trajectory class 1: “stable low BMI” (19.2%, n=87), 2: “normal BMI” (48.8%, n=221), 3: “rapid catch-up to high BMI” (17.9%, n=81), and 4: “slow catch-up to high BMI” (14.1%, n=64). The ability of the LCT modelling to discriminate the identified classes was acceptable with high median posterior probabilities of assigned class membership above 85% for all four classes (**Supplemental Figure 4**) (27). Children in the “stable low BMI” trajectory class presented on average slow initial BMI gains plateauing after 4 months and remained at a relatively low level in infancy with a small catch-up from around 8 to 27 months. The “normal BMI” trajectory was very similar to the WHO international growth standards with the infancy peak at around

5 months (26). The “rapid catch-up to high BMI” trajectory presented on average accelerated initial BMI gains peaking at around 9 months and steadily declining towards a normal BMI level at 60 months. The “slow catch-up to high BMI” trajectory presented on average slow BMI gains from birth until peaking at around 17 months.

Characteristics of the latent BMI trajectories

Background characteristics, BC measures, and cardiometabolic markers of the mother-child pairs according to the BMI trajectory class memberships are shown in **Table 1** and **Table 2**. At birth, we did not observe any statistically significant differences between the four trajectories besides for gestational age. The highest proportions of low birth weight (13-18%) were seen in the two catch-up trajectory classes. At 5 years, children in all groups were on average lighter, shorter, and thinner, compared with the WHO international growth standards (26). At 5 years, children in the “stable low BMI” class presented the highest proportions of stunting and underweight, and the lowest proportions of overweight, while children in the “rapid catch-up to high BMI” class presented the lowest proportions of stunting and underweight and had on average 1.2 kg more FM than the “stable low BMI” class.

FMI and FFMI growth in infancy

Each of the four BMI trajectory classes differed in terms of the average velocity of both FMI and FFMI growth from 0-6 months (**Figure 3**). Children in the “rapid catch-up to high BMI” class had the lowest FMI at birth but gained FMI at an accelerated rate through infancy resulting in the highest average FMI at 6 months. While growth in FMI largely reflected the patterns of the BMI classes the first 3-4 months (Figure 3 and Supplemental Figure 3), the

accretion patterns of FFMI were slightly concave with much smaller variation between the classes. At 6 months, the difference between the highest and lowest growth trajectory was 2.68 kg/m^2 for FMI and 1.32 kg/m^2 for FFMI.

Associations of BMI trajectories with BC and cardiometabolic markers at 5 years

Compared with children classified in the "normal BMI" trajectory (reference group), children in the "rapid catch-up to high BMI" class had higher triglycerides (range of β-coefficients in model 1-4: 19-21%) and C-peptide (23-25%) across the four models (**Figure 4** and **Supplemental Table 4**). Tendencies of positive associations were also seen for insulin, HbA1c, and HOMA-IR, although confidence bands were wide and included zero. Conversely, children classified in the "stable low BMI" class had lower LDL cholesterol (0.14-0.17 mmol/L), HDL cholesterol (0.05-0.09 mmol/L), but higher triglycerides (11-13%) across the four models. Tendencies of inverse associations were also seen for total cholesterol, and systolic blood pressure, although confidence bands were wide and included zero.

In relation to the BC and anthropometry measures, children in the "rapid catch-up to high BMI" class were taller (1.3-1.8 cm), had larger waist circumference (0.9-1.6 cm), higher FM (0.48-0.60 kg) and FFM (0.50-0.77 kg), while children in the "stable low BMI" class had lower height (1.2-1.5 cm), smaller waist circumference (1.0-1.3 cm), and less FM (0.60-0.64 kg) and FFM (0.35-0.49 kg) across the four models. Moreover, children in the "slow catch-up to high BMI" class had higher FM (0.46-0.58 kg), but not higher FFM. When accounting for multiple testing in the results presented in the fully adjusted model (model 4), the associations for the markers of lipid metabolism were no longer significant. However, the inverse associations of "stable low BMI" with waist circumference and FM, and the positive associations of "rapid

258 catch-up to high BMI" with height, FM and FFM remained significant (Supplemental Figure 1).

259 Adjusting all the models for breastfeeding status at 4 to 6 months postpartum did not alter

260 the associations markedly (Supplemental Figure 2).

Discussion

This is the first study from a sub-Saharan African population to examine how distinct trajectories of adiposity-related BMI the first years of life have different implications on markers of cardiometabolic risk and BC in early childhood. We found that accelerated BMI growth in infancy was associated with higher concentrations of triglycerides, C-peptide, height, waist circumference, FM, and FFM, while low BMI growth was associated with lower LDL, HDL, height, waist circumference, FM, and FFM, but higher triglycerides at 5 years in Ethiopian children. The effect estimates for the cardiometabolic markers did not change markedly across the different adjustment models. Thus, the associations were independent of maternal characteristics, socioeconomic status, and birth weight and were not mediated through size at 5 years. Since this investigation was exploratory rather than confirmatory, we did not account for multiple testing in the primary analysis.

Studies examining the health effects of variability in early-life growth have typically used predefined cut-offs to define low, normal or accelerated growth between two or more timepoints (7, 28). This *a priori* classification forces observations into specific categories that may not fully reflect the complex and dynamic trajectories of child growth. Using an exploratory data-driven approach, we were able to identify trajectories of low, normal and accelerated growth associated with markers of adiposity, insulin- and lipid metabolism without *a priori* categorization of growth variability. The generalizability of these trajectories was supported by their similarity to BMI trajectories identified in studies from high-income countries using similar data-driven modelling and overlapping age-periods (29-32). In line with the present findings, these studies also reported associations of accelerated or stable high BMI trajectories in early childhood with risk of obesity (29, 30, 32), elevated levels of

markers of adiposity (11, 30), and changes in cardiometabolic status (11, 31) in childhood or early adolescence.

The present findings and existing evidence are consistent with the capacity-load model of cardiometabolic risk (33), with early low BMI growth constraining metabolic capacity (e.g. lean mass deficits) while accelerated BMI growth increases metabolic load (e.g. excess FM and waist circumference). Both low metabolic capacity and high metabolic load challenge metabolic homeostasis. We found that FMI accretion from 0-6 months largely reflected the patterns of BMI growth. Children with accelerated BMI growth therefore had the highest FM accretion and on average ½ kg and 1 kg more FM at 5 years than the children with normal and low BMI growth, respectively. Compared with UK children, the Ethiopian children also had higher average FM (boys: 3.11 vs. 4.19 kg, and girls: 3.97 vs. 4.14 kg) and markedly lower FFM (boys: 16.35 vs. 12.27 kg, and girls: 14.60 vs. 12.04 kg) at 5 years (34). As the children with accelerated BMI growth were larger overall, they also presented the highest FFMI accretion and FFM at 5 years. However, the effect estimates for the association with FM and FFM were of almost similar size, despite that the FFM at 5 years was an almost 3 times larger body compartment than FM. Moreover, we have recently shown that FM accretion the first 4 months of life was positively associated with FMI at 4 years, but not FFMI (35), and also that a lower birth weight associates with a FM but not a FFM catch-up growth pattern (36). Altogether, it is therefore possible that accelerated BMI growth during a critical window the first 6 months of life induces disproportionally high accretion of FM in relation to FFM, that may result in unfavorable cardiometabolic changes already in childhood. Moreover, children with accelerated BMI growth may be particularly vulnerable to the effects of a high FM as they had the lowest weight at birth, indicating constrained development of metabolic

capacity during fetal life, and highest waist circumference at 5 years, related to increased metabolic load. Slow growth during infancy may continue to constrain the development of metabolic capacity, which is supported by a high proportion of stunting at 5 years in children with low BMI growth. However, these children did not appear to have a particularly unfavorable cardiometabolic status as they presented lower cholesterol concentrations and FM compared with the children with a normal BMI growth. These findings are similar to a study of survivors of severe-acute malnutrition in Malawi, where cardio-metabolic risk markers were not elevated despite several indices of low metabolic capacity (37). The likely explanation is that the accompanying lower FM is not sufficient to challenge the low metabolic capacity in these groups of children.

Continued follow-up of the present cohort will confirm whether these proposed relationships will persist in the longer term, but it is possible that children with either low or accelerated early growth are at highest risk of developing obesity and cardiometabolic diseases later in life. These new insights may help identify potential early-life targets for interventions that promote FFM accretion, linear growth, and normal birth weight (i.e. metabolic capacity) without increasing excess BMI growth and FM accretion (i.e. metabolic load). This is particularly relevant for public health professionals working in low-income countries where both under-nutrition and excess fat accumulation in early life are pertinent and discernible issues associated with an ongoing nutritional transition (12).

The strengths of this study include the 5-year longitudinal design with an average of nine repeated BMI assessments per child, detailed assessment of changes in BMI and BC in a critical window of development from birth to 6 months, and the assessment of multiple cardiometabolic markers and BC indices at 5 years of age. Another strength is the ability of

the LCT modelling to capture the complex and dynamic patterns of child growth and let the data speak for themselves by *a posteriori* identification of distinctive subgroups of BMI growth. Finally, over a 5-year period and 12 assessment waves, it was possible to retain 79% and 61% of the cohort of children included in the follow-up study for the growth modelling and regression analysis of the 5-year outcomes, respectively.

However, the study also had some limitations. First, longitudinal attrition inevitably makes the LCT modelling of the BMI trajectories more uncertain. Second, it cannot be excluded that children not able to participate in the 5-year follow-up visit have caused some selection bias in the estimated associations, although these mother-child pairs were largely similar to those included in the analysis (**Supplemental Table 3**). Third, the data-driven nature of the LCT modelling may limit generalizability of the present findings and future studies should assess whether the classification of the distinct BMI trajectories commonly apply to BMI growth in this age range. Moreover, as the LCT analysis is not forcing children into groups using predefined cut-offs, the size of each identified class may vary substantially, which may limit the power in the subsequent regression analyses. Although we obtained relatively large class sizes, the effect estimates presented in the regression analysis had relatively large confidence bands which may have resulted in type-2-errors. Fourth, we did not stratify the LCT analysis by sex, as this would have resulted in small class sizes and limit the power in the subsequent regression analyses. However, when re-estimating the BMI trajectories separately for boys and girls using the four identified classes from the whole study sample, we found no clinically meaningful sex differences in BMI growth for any of the four classes (**Supplemental Figure 5**). Fifth, as BMI comprises both weight and height, we were not able to assess how growth trajectories of these individual components was associated with later BC and cardiometabolic

risk markers. Finally, the observational nature of this study does not allow us to imply any causal effects. We cannot rule out that important unmeasured covariates such as pre-pregnancy maternal nutritional status, gestational weight gain, paternal BMI, fetal growth trajectories, comprehensive dietary assessment in infancy and childhood, and duration of breastfeeding have confounded our results. However, in a sensitivity analysis we adjusted our results for a crude measure of breastfeeding status at 4-6 months post-partum, which did not affect our results noticeably.

In a birth cohort of Ethiopian children, we identified considerable heterogeneity in BMI growth in early childhood, which was associated with FM accretion in early infancy and markers of cardiometabolic status and indices of BC at 5 years. Collectively, our findings offer an important contribution to understand pathways from early growth to later cardiometabolic health by suggesting that distinct trajectories of BMI growth in early life may be a key factor in the complex etiology of obesity and cardiometabolic risk development already from an early age. This is highly relevant for public health professionals in low-income countries, where the dual burden of malnutrition is a pertinent problem, as it offers potential early-life targets for interventions by identifying subgroups of children that may present unfavorable growth trajectories.

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Tables

Table 1 Description of the mother-child pairs attending the 5-year follow-up visit and included in the trajectory modelling according to the BMI trajectory class membership (n= 352) [1]

	Trajectory group					
	1: Stable low BMI (n=69)	**2: Normal BMI** (n=173)	**3: Rapid catch-up to high BMI** (n=60)	**4: Slow catch-up to high BMI** (n=50)	***p*-value** [2]	**Missing, n**
Maternal characteristics						
Age at birth (years)	24.8 (4.8)	24.7 (4.6)	24.1 (4.6)	24.4 (5.5)	0.825	0
Postpartum height (cm)	156.6 (5.8)	157.5 (6.3)	157.7 (5.5)	155.6 (6.3)	0.187	2
Postpartum body mass index (kg/m^2)	21.91 (3.06)	22.09 (3.65)	22.33 (3.39)	23.02 (3.78)	0.352	6
Birth order of current child						
First	44.9	44.5	66.7	54.0		
Second	27.5	29.5	16.7	26.0		
Third or above	27.5	26.0	16.7	20.0	0.122	0
Breastfeeding status at 4 to 6 months post-partum						
Exclusive	8.8	15.1	15.5	4.3		
Almost exclusive (water given)	20.6	20.1	24.1	23.4		
Predominant	66.2	57.9	56.9	63.8		
Partial or no	4.4	6.9	3.4	8.5	0.581	20
Maternal education						
No school	5.8	8.7	1.7	10.0		
Some primary school	47.8	47.4	41.7	34.0		
Completed primary school	11.6	14.5	20.0	20.0		
Completed secondary school	18.8	17.9	20.0	26.0		
Higher education	15.9	11.6	16.7	10.0	0.496	0
Socioeconomic status (International Wealth Index)	45.9 (17.2)	44.6 (16.7)	48.2 (18.6)	45.4 (16.5)	0.565	0
Child characteristics at birth						
Sex (boys)	47.8	50.3	51.7	52.0	0.965	0
Gestational age (weeks)	39.2 (0.9)	39.1 (1.0)	39.0 (1.0)	38.7 (0.8)	0.020	0
Weight (kg)	3.10 (0.40)	3.07 (0.38)	2.95 (0.43)	2.98 (0.47)	0.076	0
Length (cm)	49.2 (2.1)	49.3 (1.9)	49.0 (1.9)	48.7 (2.1)	0.291	0
Fat mass (kg)	0.25 (0.20)	0.23 (0.15)	0.18 (0.15)	0.22 (0.16)	0.076	2
Fat-free mass (kg)	2.85 (0.29)	2.85 (0.32)	2.78 (0.35)	2.78 (0.35)	0.322	2
Low birth weight (%) [3]	5.8	6.9	13.3	18.0	0.061	0
Child characteristics at 5 years						
Age at 5-year visit (months)	59.8 (1.7)	60.0 (1.4)	60.1 (1.5)	59.8 (1.4)	0.702	0
Weight (kg)	15.14 (1.72)	16.25 (1.88)	17.49 (2.32)	16.76 (2.05)	<0.001	0
Length (cm)	102.8 (4.8)	104.2 (4.2)	106.0 (4.5)	103.8 (3.8)	<0.001	0
Weight for age SDS [4]	-1.40 (0.81)	-0.90 (0.80)	-0.39 (0.89)	-0.67 (0.87)	<0.001	0
Height for age SDS	-1.44 (0.98)	-1.15 (0.86)	-0.78 (0.95)	-1.25 (0.79)	<0.001	0
BMI for age SDS	-0.73 (0.91)	-0.27 (0.79)	0.14 (0.79)	0.15 (0.84)	<0.001	0
Underweight [5]	21.7	8.7	0.0	6.0	<0.001	0
Stunted [6]	26.1	12.1	11.7	16.0	0.043	0
Wasted by BMI (Thinness) [7]	7.2	2.3	0.0	2.0	0.090	0
Overweight [8]	1.4	1.7	10	14.0	0.001	0
Obese [9]	0.0	0.6	3.3	2.0	0.164	0

[1] Data are mean (SD) (for continues normally distributed variables) and percentages (for categorical variables). [2] Differences between trajectory groups were calculated by One-way ANOVA F-test (for continuous variables), Pearson's Chi-Square test of independence (for categorical variables with expected counts above 5 in all cells) and Fisher's exact test of independence (for categorical variables with expected counts in any cell below 5). [3] Low birth weight is defined as birth weight <2500 g. [4] Standard deviation scores (SDS) are derived using the 2006 (aged <61 months) and 2007 (aged ≥61 months) World Health Organization (WHO) child growth standards. [5] Weight for age more than 2 SDS below the age- and sex-specific median of the WHO child growth standards. [6] Height for age more than 2 SDS below the age- and sex-specific median of the WHO child growth standards. [7] BMI for age more than 2 SDS below the age- and sex-specific median of the WHO child growth standards. [8] BMI-for-age more than 1 to 2 SDS above the sex-specific median of the WHO child growth standards. [9] BMI-for-age more than 2 SDS above the sex-specific median of the WHO child growth standards.

Table 2 Cardiometabolic markers and body composition at 5 years of age in the children attending the 5-year follow-up visit and included in the trajectory modelling according to the BMI trajectory class membership (n= 352) [1]

	Trajectory group					
	1: Stable low BMI (n=69)	**2: Normal BMI** (n=173)	**3: Rapid catch-up to high BMI** (n=60)	**4: Slow catch-up to high BMI** (n=50)	*p*-value [2]	Missing, n
Glucose metabolism						
Glucose (mmol/L)	5.84 (0.83)	5.90 (0.83)	5.98 (1.08)	5.89 (0.63)	0.844	26
HbA1c (mmol/mol)	37 (5)	37 (4)	38 (4)	38 (5)	0.736	83
Insulin (µU/mL) [3]	5.47 (2.62, 11.58)	5.89 (3.15, 11.08)	8.29 (4.12, 11.61)	6.39 (3.04, 9.43)	0.166	34
C-peptide (ng/mL) [3]	0.99 (0.57, 1.24)	1.05 (0.59, 1.54)	1.31 (0.77, 1.84)	1.01 (0.70, 1.35)	0.094	39
HOMA-IR [3,4]	1.12 (0.55, 2.53)	1.21 (0.63, 2.33)	1.86 (0.86, 2.58)	1.31 (0.65, 2.07)	0.196	34
Lipids						
Total cholesterol (mmol/L)	3.32 (0.63)	3.43 (0.58)	3.49 (0.71)	3.37 (0.55)	0.430	30
LDL (mmol/L)	1.54 (0.52)	1.68 (0.55)	1.70 (0.68)	1.66 (0.49)	0.322	31
HDL (mmol/L)	0.76 (0.23)	0.81 (0.26)	0.77 (0.27)	0.78 (0.24)	0.434	35
Triglycerides(mmol/L) [3]	1.00 (0.79, 1.29)	0.91 (0.68, 1.27)	1.04 (0.83, 1.53)	0.90 (0.72, 1.15)	0.042	35
Blood pressure						
Systolic (mmHg)	86.1 (6.7)	88.0 (7.2)	89.3 (8.0)	87.5 (7.5)	0.092	2
Diastolic (mmHg)	53.6 (6.8)	54.2 (8.2)	55.4 (10.3)	54.3 (9.3)	0.673	2
Anthropometry and body composition						
Body mass index (kg/m^2)	14.32 (1.16)	14.92 (1.10)	15.51 (1.21)	15.52 (1.20)	<0.001	0
Waist circumference (cm)	50.09 (2.69)	51.38 (2.89)	52.77 (3.21)	51.99 (2.85)	<0.001	1
Fat mass (kg)	3.50 (1.22)	4.13 (1.15)	4.68 (1.34)	4.64 (1.24)	<0.001	16
Fat-free mass (kg)	11.64 (1.14)	12.14 (1.33)	12.86 (1.69)	12.10 (1.43)	<0.001	16
Fat mass index (kg/m^2)	3.30 (1.10)	3.79 (0.99)	4.14 (1.05)	4.29 (1.03)	<0.001	16
Fat-free mass index (kg/m^2)	11.02 (0.78)	11.17 (0.80)	11.41 (1.01)	11.26 (0.96)	0.079	16

[1] Data are mean (SD) for continues variables that are normally distributed and median (interquartile range) for continuous variables that are not following a normal distribution. [2] Differences between trajectory groups were calculated by One-way ANOVA F-test for continues normally distributed variables. Variables found not to follow a normal distribution were log transformed prior to the tests of group differences. [3] Nonnormally distributed. [4] Homeostasis model assessment of insulin resistance (HOMA-IR) was calculated as insulin (µU/mL) × glucose (mmol/l) / 22.5.

Legends for figures

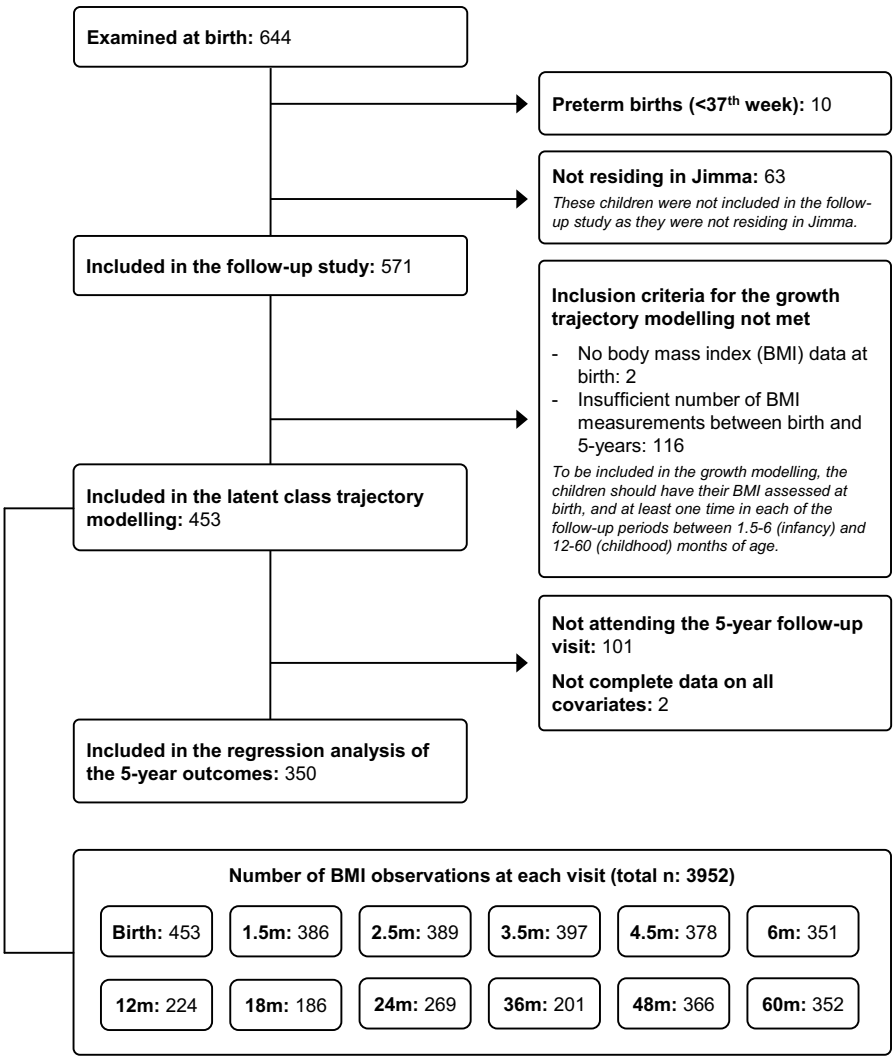
Figure 1. Flow diagram of the included children and number of BMI observations at each follow-up visit from birth to 60 months.

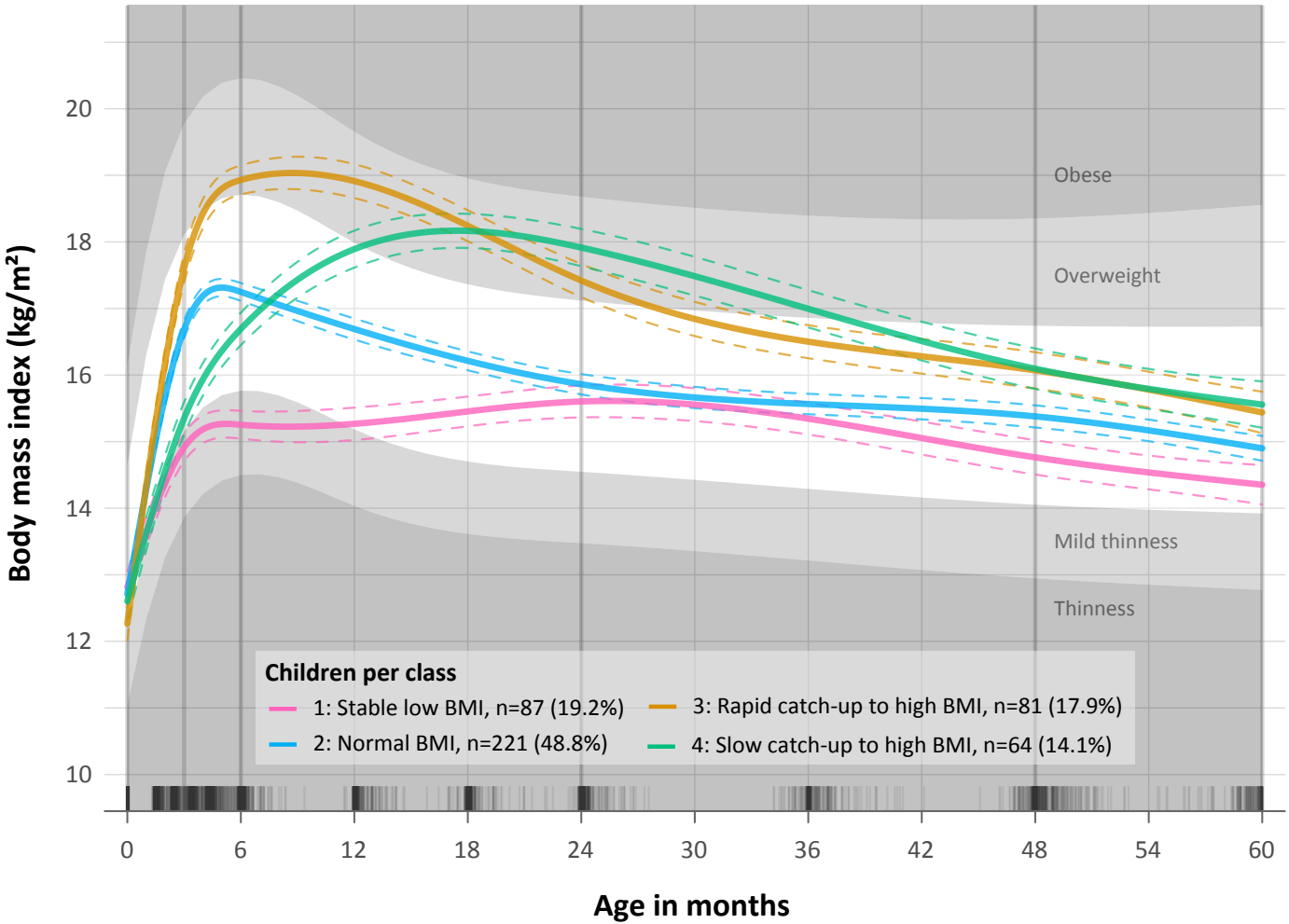
Figure 2. Distinct body mass index (BMI) trajectories from 0-5 years of children in the iABC birth cohort derived from a latent class trajectory analysis. Solid lines display the class-specific estimated average BMI as a function of age. The dashed lines show the estimated 95% confidence limits. The shaded areas indicate the reference in standard deviation scores (SDS) from the median BMI-for-age according to the international growth standards developed by the World Health Organization. Normal BMI (white) is defined as a BMI-for-age SDS from -1 to 1, mild thinness as ≥ -2 to <-1 SDS (light grey), thinness as < -2 SDS (grey), overweight as >1 to ≤ 2 SDS (light grey), and obese as > 2 SDS (grey). The density of BMI observations is shown as a rug plot along the x-axis.

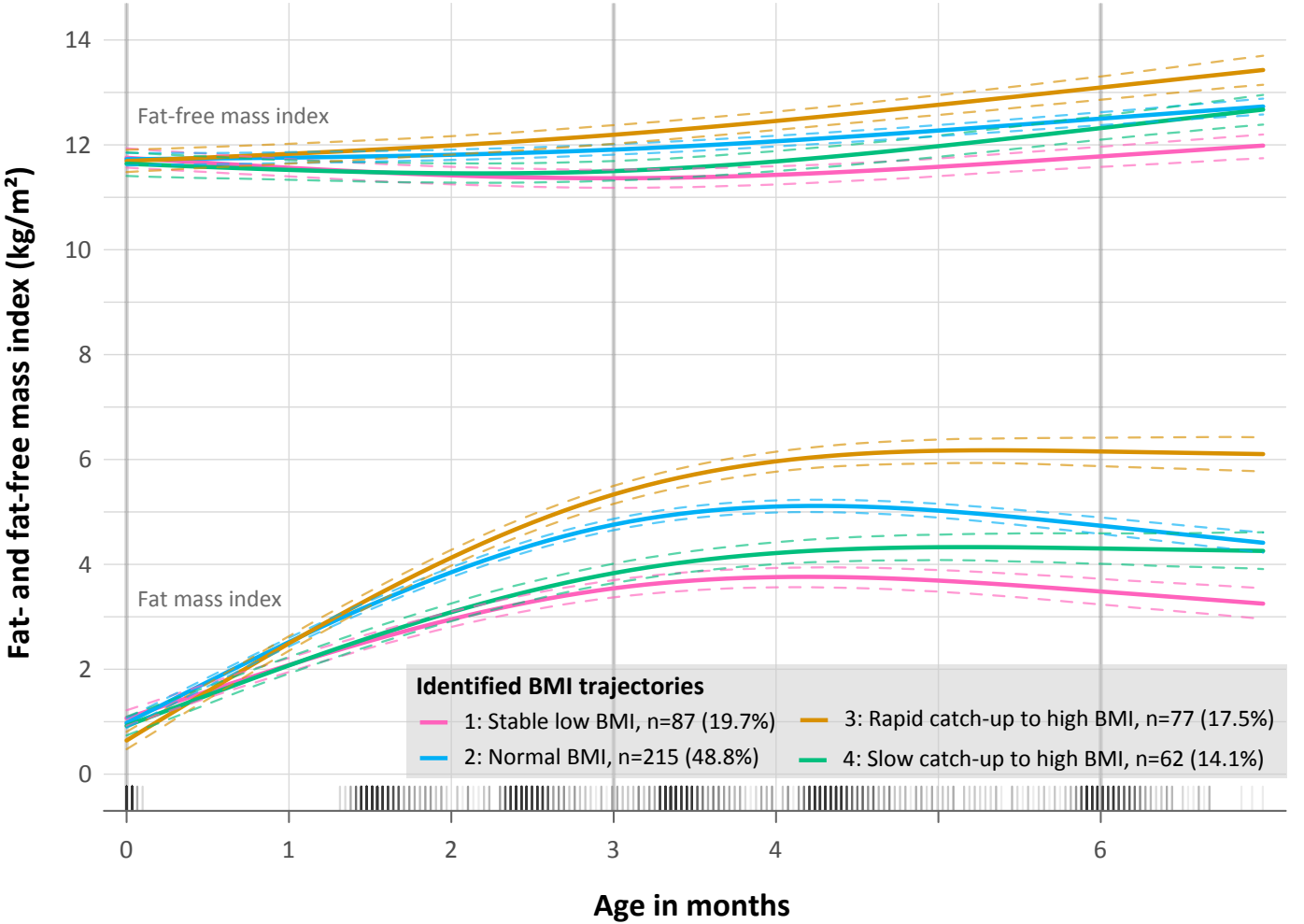
Figure 3. Estimated changes in fat mass index and fat-free mass index from 0-6 month for each of the identified latent body mass index trajectories classes.

Figure 4. The coefficients (95% CIs) displayed in the forest plots were derived from multiple linear regression models and represent the mean difference in concentrations of cardiometabolic markers and body composition indices between the reference trajectory 2: normal BMI, and the 3 BMI trajectory categories 1: stable low BMI, 3: rapid BMI catch-up and 4: slow BMI catch-up, respectively. The 4 distinct BMI trajectories (exposure variable) were

derived from a latent class trajectory analysis. The outcome variables insulin, C-peptide, HOMA-IR and triglycerides were log transformed prior to analyses. The resulting effect estimates were back-transformed and presented as percentwise change. Model 1 (the leftmost circle) was adjusted for sex, birth order and gestational age. Model 2 was additionally adjusted for child age at the 5-year visit, maternal age at delivery, maternal postpartum height, maternal educational status and family socioeconomic status (International Wealth Index). Model 3 was additionally adjusted for child birth weight. Model 4 (the rightmost circle) was additionally adjusted for child BMI at the 5-year visit. In model 4, the analyses of FM and waist circumference were adjusted for FFM and height at 5 years instead of BMI, and the analysis of FFM was adjusted for FM and height at 5 years instead of BMI. * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$.


Examined at birth: 644
Preterm births (<37th week): 10
Not residing in Jimma: 63
These children were not included in the follow-up study as they were not residing in Jimma.
Included in the follow-up study: 571
Inclusion criteria for the growth trajectory modelling not met
- No body mass index (BMI) data at birth: 2
- Insufficient number of BMI measurements between birth and 5-years: 116
To be included in the growth modelling, the children should have their BMI assessed at birth, and at least one time in each of the follow-up periods between 1.5-6 (infancy) and 12-60 (childhood) months of age.
Included in the latent class trajectory modelling: 453
Not attending the 5-year follow-up visit: 101
Not complete data on all covariates: 2
Included in the regression analysis of the 5-year outcomes: 350
Number of BMI observations at each visit (total n: 3952)
Birth: 453
1.5m: 386
2.5m: 389
3.5m: 397
4.5m: 378
6m: 351
12m: 224
18m: 186
24m: 269
36m: 201
48m: 366
60m: 352


Body mass index (kg/m²)
20
18
16
14
12
10
Obese
Overweight
Mild thinness
Thinness
Children per class
1: Stable low BMI, n=87 (19.2%)
2: Normal BMI, n=221 (48.8%)
3: Rapid catch-up to high BMI, n=81 (17.9%)
4: Slow catch-up to high BMI, n=64 (14.1%)
0
6
12
18
24
30
36
42
48
54
60
Age in months


Fat- and fat-free mass index (kg/m²)
14
12
10
8
6
4
2
0
Fat-free mass index
Fat mass index
0
1
2
3
4
6
Age in months
Identified BMI trajectories
1: Stable low BMI, n=87 (19.7%)
2: Normal BMI, n=215 (48.8%)
3: Rapid catch-up to high BMI, n=77 (17.5%)
4: Slow catch-up to high BMI, n=62 (14.1%)


Glucose (mmol/L), n=324
HbA1c (mmol/mol), n=267
Insulin (% change), n=316
C-peptide (% change), n=311
HOMA-IR (% change), n=316
Total cholesterol (mmol/L), n=320
LDL (mmol/L), n=319
HDL (mmol/L), n=315
Triglycerides (% change), n=315
Systolic blood pressure (mmHg), n=348
Diastolic blood pressure (mmHg), n=348
Height (cm), n=350
Waist circumference (cm), n=349
Fat mass (kg), n=334
Fat-free mass (kg), n=334
Estimate (95% CI)
1: Stable low BMI
3: Rapid catch-up to high BMI
4: Slow catch-up to high BMI
Growth trajectories (0-5 years)